

Formal Draft Biological Opinion.

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**Biological Opinion
on
EPA's Proposed Program of Continuing Approval or Promulgation
of New Cyanide Criteria in State and Tribal Water Quality Standards**

**U.S. Fish and Wildlife Service
Arlington, Virginia
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1.0 LIST OF ACRONYMS

ACR	Acute to Chronic Ratio
BE	Biological Evaluation
BMP	Best Management Practice
CCC	Criteria Continuous Concentration (chronic criterion)
CFR	Code of Federal Regulations
CMC	Criteria Maximum Concentration (acute criterion)
CN	Cyanide
CPP	Continuing Planning Process
CWA	Clean Water Act
EC	Effects Concentration
EPA	Environmental Protection Agency
ESA	Endangered Species Act
FAV	Final Acute Value
FR	Federal Register
HCN	Hydrogen Cyanide
ICE	Interspecies Correlation Estimate
ISO	International Organization for Standardization
LC	Lethal Concentration
LCL	Lower Confidence Limit
LOEC	Lowest Observable Effects Concentration
MATC	Maximum Acceptable Toxicant Concentration
MLE	Maximum Likelihood Estimate
NOEC	No Observable Effects Concentration
NPDES	National Pollution Discharge Elimination System
NPS	Non-Point Source
OECD	Organisation for Economic Co-operation and Development
SETAC	Society for Environmental Toxicology and Chemistry
TMDL	Total Maximum Daily Load
WQM	Water Quality Management
WQS	Water Quality Standard

2.0 INTRODUCTION

This document transmits the U.S. Fish and Wildlife Service's (Service) biological opinion based upon our review of EPA's proposed program of continuing approval or promulgation of acute and chronic cyanide criteria in State and Tribal water quality standards and their effects on endangered and threatened species and designated critical habitats in accordance with section 7 of the Endangered Species Act of 1973, as amended (16 U.S.C. 1531 *et seq.*)(ESA). Your March 23, 2007, request for formal consultation was received on March 26, 2007.

Your request for formal consultation also included a request for our concurrence that the proposed acute and chronic cyanide criteria are not likely to adversely affect any endangered and threatened species or their critical habitats listed in section 9.2.1 of your March 23, 2007, *Biological Evaluation of Aquatic Life Criteria—Cyanide* (CN BE). According to information presented in the CN BE, available data suggest that cyanide is toxic to aquatic plants only at concentrations well above the proposed criterion concentrations. For that reason, we concur with your finding. We determined the proposed action may affect but is not likely to adversely affect numerous threatened and endangered animal species. The species for which we make this determination and our rationale are provided in Appendix B.

This biological opinion is based on information provided in: (1) your March 23, 2007, CN BE; (2) your July 31, 2006, *Draft Framework for Conducting Biological Evaluations of Aquatic Life Criteria: Methods Manual* (BE Methodology); (3) your October 29, 2004, *Draft Methodology for Conducting Biological Evaluations of Aquatic Life Criteria: Methods Manual*; (4) the February 22, 2001, *Memorandum of Agreement Between the Environmental Protection Agency, Fish and Wildlife Service and National Marine Fisheries Service Regarding Enhanced Coordination Under the Clean Water Act and Endangered Species Act; Notice* (66 FR 11202); and (5) additional information contained in Service files. A complete administrative record of this consultation is on file at the Service's Headquarters Office in Arlington, Virginia.

Scope of this Biological Opinion

This biological opinion evaluates the effects of EPA's program of continuing approval of cyanide criteria adopted in State water quality standards under the Clean Water Act (CWA) on listed species and their critical habitats. More precisely, this biological opinion evaluates at a program level the potential effects to listed species and critical habitat from exposure to cyanide at EPA's recommended CWA section 304(a) criteria. The assumptions made in this biological opinion about the potential for exposure at the criteria values may not apply consistently to all State and Tribal water quality standards or even all waters within a State or Tribal boundary. State and Tribal water quality standards are comprised of 4 elements: (1) designated uses; (2) criteria sufficient to protect those uses; (3) an antidegradation policy; and (4) general policies. The other components of a water quality standard, besides the criteria, may invalidate the assumptions made here about exposure.

Because the assumptions made in this biological opinion about the potential for listed species' exposure at the criteria values may not apply consistently to all State and Tribal water quality standards or even all waters within a State or Tribal boundary, this biological opinion does not include incidental take exemptions. Given the uncertainty about such exposure, we are unable to analytically determine at the program scale the likelihood (as opposed to the potential) of such take occurring. Therefore, it will be necessary for EPA to conduct subsequent, step-down ESA section 7 consultations with the Service on individual State and Tribal water quality standards to determine if incidental take exemptions associated with State and Tribal implementation of cyanide criteria in their water quality standards are warranted.

We anticipate much of the analysis presented in this biological opinion will carry over, so that the tiered consultation on State and Tribal water quality standards need only focus on potential effects of elements that were not fully considered here. Because we address criteria in this biological opinion, tiered consultation would likely focus on the various General Policies being implemented by each State and Tribe and the need to incorporate considerations for listed species and critical habitats into those policies, where necessary.

The Service expects that the EPA will notify affected States and Tribes of the need, in their next triennial review, to review their water quality standards for consistency with the findings presented in this biological opinion. EPA should work with States and Tribes to coordinate and consult directly with the Service's local Field Offices in conducting this review. In this review, EPA will, in consultation with the Service, evaluate waters of the United States within the range of applicable listed species and critical habitats considered in this biological opinion to determine whether:

1. there exists a designated use to which aquatic life criteria apply;
2. State-adopted criteria are consistent with this biological opinion; and,
3. general policies adopted by States or Tribes and approved by EPA would alter the effects analysis in this biological opinion.

This tiered consultation will involve a review of the effects of the proposed cyanide criteria on listed species and critical habitats at a more refined scale that incorporates the four elements of the State's or Tribe's water quality standards as approved. As noted above, it is at this scale that we will determine if the effects analysis includes findings that support an incidental take exemption(s).

3.0 CONSULTATION HISTORY

On February 22, 2001, the Service and the National Marine Fisheries Service (NMFS; collectively, the Services), and EPA noticed in the Federal Register the 2001, Memorandum Of Agreement (MOA) referenced above which, among other things, described our plan for conducting ESA section 7 consultations on EPA's recommended aquatic life criteria.

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In January 2004, the Services and EPA decided to proceed with a data call for the first batch of pollutants that would be reviewed in consultation, while continuing to work on the *Draft Methodology for Conducting Biological Evaluations of Aquatic Life Criteria--Methods Manual*.

On May 14, 2004, the Services and EPA issued data calls to regional staff and NMFS science center staff requesting information and data on cyanide, ammonia, chromium III and chromium VI. The data call requested the regions and science centers to send relevant studies to our Headquarters Offices by June 30, 2004.

On November 12, 2004, the Services received by email from EPA a November 5, 2004, revised *Draft Methodology for Conducting Biological Evaluations of Aquatic Life Criteria--Methods Manual* (dated October 29, 2004, on the document). This version represented a methodology developed collaboratively, and which had been peer reviewed by subject experts outside of the Federal government.

In December 2004, the Service and EPA exchanged comments on recommended revisions to the November draft methodology. EPA also informed the Services that they had received a draft BE for cyanide from their contractor and were reviewing the document to ensure the contractor had followed the BE methodology accurately.

On January 19, 2005, EPA transmitted by email the January 19, 2005, *Biological Evaluation of Aquatic Life Criteria—Cyanide, Part 1: Toxicity Analysis and Preliminary Effects Assessment*.

On May 3, 2005, the Services transmitted by email comments on EPA's January 19, 2005, draft biological evaluation for cyanide criteria.

On January 31, 2006, the Service received a January 26, 2006, letter from EPA requesting the Service to review a January 26, 2006, draft CN BE for its "completeness" in fulfilling the information requirements for ESA section 7 consultation.

On April 18, 2006, the Service transmitted a letter to EPA responding to EPA's January 26, 2006, consultation request.

In a June 29, 2006, letter, EPA requested the Service's concurrence that the proposed approvals of cyanide criteria were not likely to adversely affect 455 threatened and endangered species and their critical habitats.

On August 1, 2006, EPA electronically transmitted a July 31, 2006, *Draft Framework for Conducting Biological Evaluations of Aquatic Life Criteria: Methods Manual*, which they used to support their effects determinations.

On November 28, 2006, the Service responded by letter to EPA's consultation request with a recommendation that EPA initiate formal consultation.

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On March 23, 2007, EPA requested formal consultation and provided the CN BE, which concluded their action was not likely to adversely affect listed species or designated critical habitats.

On May 29, 2007, the Service transmitted a letter acknowledging the initiation of formal consultation.

On August 6, 2007, the Service transmitted a letter to EPA requesting an extension of formal consultation to November 6, 2007.

On August 17, 2007, the Service received an August 15, 2007, letter from EPA offering to extend the consultation period to September 30, 2007.

On May 5-9, 2008, the Services met with EPA to conduct a “Kaizen” “lean event.” The purpose of the meetings was to analyze the cyanide consultation process from the development of a biological assessment through the anticipated completion of formal consultation in an effort to find efficiencies in the process. The Services and EPA also discussed coordination and communication with respect to the national consultation on cyanide and local consultation on EPA promulgation of Oregon water quality standards.

On June 12, 2008, the Services and EPA met to follow up on the Kaizen lean event. Subsequent follow up meetings were cancelled until the Services completed draft biological opinions.

BIOLOGICAL OPINION

This biological opinion does not rely on the regulatory definition of “destruction or adverse modification” of critical habitat at 50 C.F.R. 402.02. Instead, we have relied upon the statutory provisions of the ESA to complete the following analysis with respect to critical habitat.

4.0 DESCRIPTION OF THE PROPOSED ACTION

Background

Section 304(a)(1) of the CWA directs EPA to publish criteria for water quality that accurately reflect the latest scientific knowledge on a number of factors, including “...the kind and extent of all identifiable effects on health and welfare including, but not limited to, plankton, fish, shellfish, wildlife, plant life, shorelines, beaches...which may be expected from the presence of pollutants in any water body, including groundwater;” “on the concentration and dispersal of pollutants, or their byproducts,” and on “the effects of pollutants on biological community diversity, productivity, and stability.” EPA’s CWA section 304(a) cyanide aquatic life criteria recommendations are published in *Ambient Water Quality Criteria for Cyanide – 1984* (EPA 1985). EPA describes the published

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criteria as a criterion maximum concentration (CMC) and criterion continuous concentration (CCC) for freshwater and saltwater:

Freshwater CMC (as free cyanide) = 22 µg/L

Freshwater CCC (as free cyanide) = 5.2 µg/L

Saltwater CMC (as free cyanide) = 1.0 µg/L

Saltwater CCC (as free cyanide) = 1.0 µg/L

For cyanide, the CMC (“acute” criterion) represents an estimated concentration in fresh or salt water to which aquatic organisms and their uses should not be affected unacceptably if the one-hour average concentration does not exceed this value more than once every three years on average, except possibly where locally important species are more sensitive (EPA 1985). The CCC (“chronic” criterion) represents an estimated concentration in either fresh or salt water to which aquatic organisms and their uses should not be affected unacceptably if the four-day average concentration does not exceed the CCC more than once every three years on average, except possibly where locally important species are more sensitive (EPA 1985).

Section 303(c)(2)(B) of the CWA requires States, Tribes, and U.S. territories (hereafter referred to collectively as States as defined by EPA in 40 CFR 131.3(j)) to adopt into their water quality standards numeric criteria for toxic pollutants listed under section 307(a) of the CWA for which section 304(a) criteria have been published if the presence of these pollutants is likely to affect a water body’s use. States can adopt criteria under section 303(c) that differ from EPA’s 304(a) criteria values whenever adequately justified, but States generally choose to adopt, verbatim, EPA’s 304(a) criteria and rely on the criteria document for their scientific justification. Once adopted into State or Tribal water quality standards, criteria help form the legal basis for implementing CWA programs to control pollution and achieve the goals and requirements of the CWA.

Proposed Program Action

EPA would approve State or Tribal-adopted water quality standards for cyanide criteria, or promulgate Federal water quality standards for cyanide criteria for U.S. waters that are identical to or more stringent than EPA’s recommended CWA section 304(a) aquatic life criteria for cyanide (EPA 1985). EPA’s cyanide criteria document (EPA 1985) serves as the States’, Tribes’ and EPA’s scientific justification that the recommended criteria values are sufficient to achieve designated uses that protect aquatic life under the CWA.

All States and Tribes have already either adopted into their state water quality standards cyanide criteria identical to EPA’s recommended 304(a) cyanide aquatic life criteria (EPA 1985) and EPA has approved them, or EPA promulgated cyanide criteria for the States and Tribes identical to their 304(a) criteria. For some water bodies, States and Tribes have adopted and EPA has approved site-specific criteria less stringent than the CWA 304(a) criteria, but EPA has not included continuing approval of less stringent criteria as part of the proposed action considered in this consultation.

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This biological opinion does not evaluate or provide ESA section 7 compliance for past EPA actions involving cyanide criteria for water quality standards or the past, present, and future effects of those actions on listed species and critical habitats. Past actions and their effects are relevant to describing the Status and Environmental Baseline of listed species and critical habitats. In this biological opinion the Service evaluates the proposed action in the context of EPA's continuing oversight role under the CWA and its discretion and authority to approve or promulgate State and Tribal water quality standards and affect implementation of CWA programs to achieve the goals and requirements of the CWA and the requirements of section 7(a)(2) of the ESA.

Interrelated and Interdependent Activities

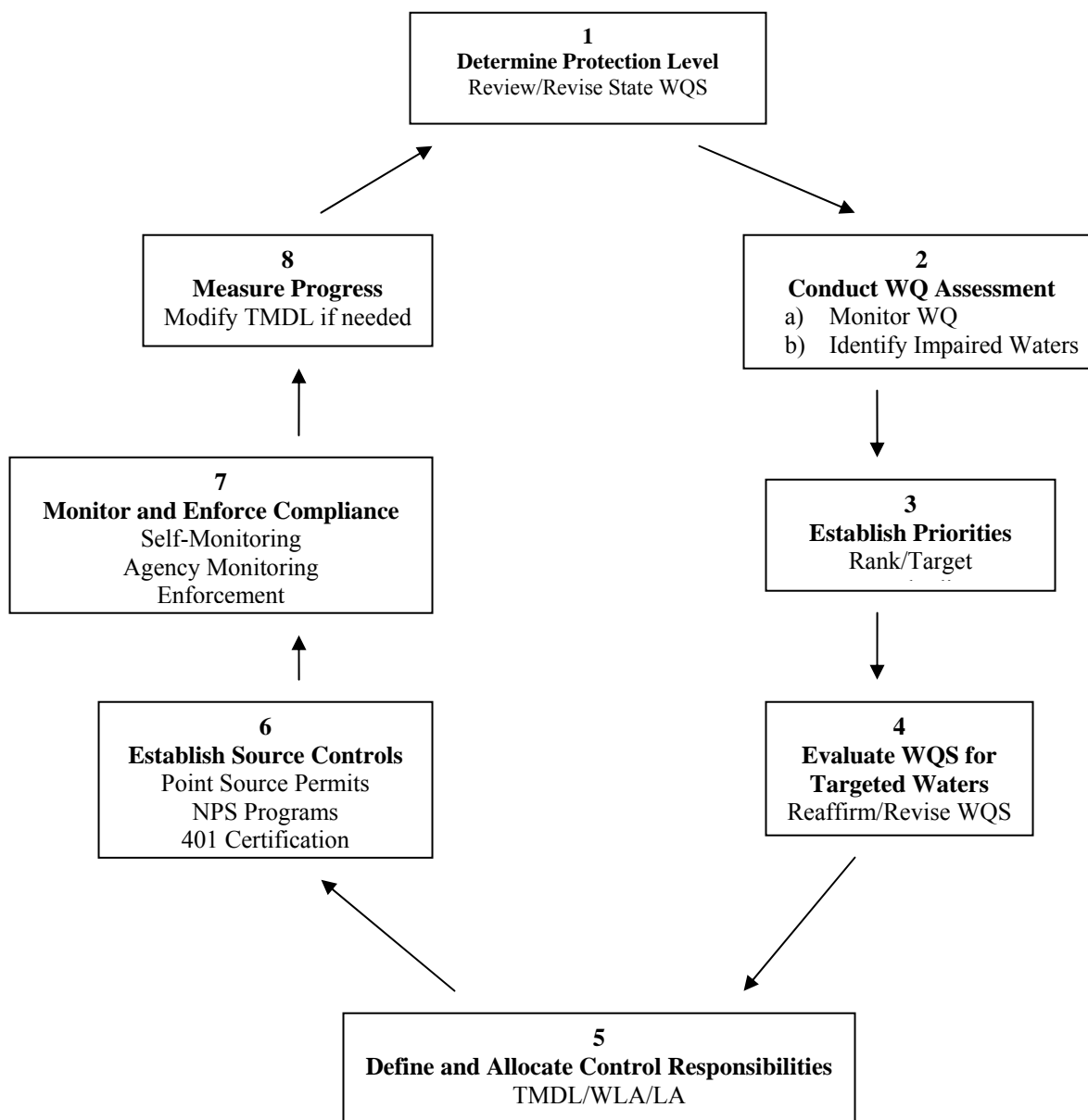
The effects of EPA's approval or promulgation of cyanide criteria in State and Tribal water quality standards must be understood in the larger context of the CWA. This larger context is framed by Congress' stated objective, goals, and policies of the CWA and the programs and activities authorized by the CWA and implemented by EPA, States, Tribes and local governments.

The objective of the CWA is to restore and maintain the chemical, physical, and biological integrity of the Nation's waters (CWA section 101(a)). In order to achieve this objective, the CWA articulates goals to ultimately eliminate the discharge of pollutants into navigable waters, with an interim goal of water quality that provides for the protection and propagation of fish, shellfish, and wildlife. Congress also articulated in section 101 of the CWA its policy to prohibit the discharge of pollutants in toxic amounts, and its policy that programs be developed and implemented so that the goals of the CWA could be met through the control of point and nonpoint sources of pollution.

Water quality standards are the States' and Tribes' goals for individual water bodies and provide the regulatory basis for control decisions under the CWA (40 CFR 130.0(b)). States, Tribes and EPA plan and implement CWA programs to manage point and non-point sources of pollution to attain the goals established in State and Tribal water quality standards. Control measures are implemented by issuing permits, building publicly-owned treatment facilities, instituting best management practices for nonpoint sources of pollution, and other means.

In the Water Quality Standards Handbook (EPA 1994), EPA illustrates how the various elements of the CWA and its regulations work together to manage pollution. Figure 1, reproduced from the handbook, describes the eight stages of the water quality-based approach to pollution control. Each stage represents a major CWA program with specific regulatory requirements and guidance. This figure is intended to illustrate how the different programs fit into the overall water quality control scheme.

Figure 1. Eight stages of EPA's approach to water quality-based pollution control.



1. **Determine Protection Level.** States and Tribes adopt, and EPA approves, water quality standards to protect public health or welfare, enhance the quality of water, and serve the purposes of the CWA. A water quality standard defines the water quality goals of a water body, or portion thereof, by designating a use or uses, by setting criteria necessary to protect the uses, and by preventing degradation of water quality through antidegradation provisions. States and Tribes may, at their discretion, include general policies which allow for exceedances in discharges

under specific circumstances. Development and review/revision of standards is directed by the Water Quality Standards Regulation (40 CFR part 131), described in greater detail below.

2. **Conduct Water Quality Assessment.** Once State and Tribal water quality standards establish the level of protection to be afforded to a water body, States and Tribes are required to conduct water quality monitoring to identify waters not meeting the standards. Section 305(b) of the CWA requires States and Tribes to prepare a water quality inventory every 2 years to document the status of water bodies that have been assessed. Under section 304(l), States and Tribes identify all surface waters adversely affected by toxic, conventional, and nonconventional pollutants from point and non-point sources. Under section 314(a), States and Tribes identify publicly-owned lakes for which uses are known to be impaired by point and non-point sources. Section 319 requires States and Tribes to perform nonpoint source (NPS) assessments of navigable waters, including the identification of impaired and threatened waters and the activities causing impairment. NPS assessment reports and management programs are subject to EPA approval and oversight. The collective assessment efforts contribute to the States' and Tribes' section 303(d) listing of waters for which effluent limitations and other pollution control requirements are not stringent enough to implement a water quality standard.
3. **Establish Priorities.** Once waters needing additional controls have been identified, States and Tribes are required to submit for EPA review their priority rankings of waters in need of total maximum daily load (TMDL) development.
4. **Evaluate WQS for Targeted Waters.** At this point in the water quality management process, States and Tribes have targeted priority water quality-limited water bodies. EPA recommends that States and Tribes re-evaluate the appropriateness of the water quality standards for the targeted waters if: 1) States or Tribes have not conducted in-depth analyses of appropriate uses and criteria; 2) changes in the uses of the water body may require changes in the standard; 3) more recent water quality monitoring show the standard is being met; and, 4) site-specific criteria may be appropriate because of specific local environmental conditions or the presence of species more or less sensitive than those included in the national criteria data set.
5. **Define and Allocate Control Responsibilities.** For water quality limited waters, States and Tribes must establish a TMDL that quantifies pollutant sources, and a margin of safety, and allocates allowable loads to the contributing point and non-point source discharges so that the water quality standards are attained. EPA recommends States and Tribes develop TMDLs on a watershed basis.
6. **Establish Source Controls.** Once a TMDL has been established and the appropriate source loads developed, implementation should proceed. The first step is to update the "water quality management plan," described below. Next, point

and nonpoint source controls should be implemented to meet waste load allocations and load allocations, respectively. The NPDES permitting process is used to limit effluent from point sources. Construction decisions regarding publicly-owned treatment works must also be based on the more stringent of technology-based or water quality-based limitations. In the case of nonpoint sources, State, Tribal and local laws may authorize the implementation of nonpoint source controls, such as best management practices (BMPs) or other management measures.

7. **Monitor and Enforce Compliance.** Monitoring is essential to water quality-based decision making. Point source dischargers are required to provide reports on compliance with NPDES permit limits. A monitoring requirement can be put into the permit as a special condition as long as the information is collected for the purposes of writing a permit limit. Effective monitoring programs are also required for evaluating nonpoint source control measures and EPA provides guidance in implementing and evaluating nonpoint source control measures. EPA and States and Tribes are authorized to bring civil or criminal action against facilities that violate their NPDES permits. State nonpoint source programs are enforced under State law and to the extent provided by State law.
8. **Measure Progress.** If the water body achieves the applicable State or Tribal water quality standards it may be removed from the section 303(d) list of waters needing TMDLs. If water quality standards are not met, the TMDL and allocation of load and waste loads must be modified.

The water quality-based approach to pollution control is implemented by EPA, and the States and Tribes consistent with specific statutory and regulatory requirements. The regulatory requirements are articulated in: the *Water Quality Planning and Management* regulation (40 CFR 130) (50 FR 1779, January 11, 1985, as amended 54 FR 14359 April 11, 1989; 57 FR 33049, July 24, 1992; 59 FR 13817, March 23, 1994; 65 FR 17170, March 31, 2000; and, 66 FR 53048, October 18, 2001); and the *Water Quality Standards* regulation (40 CFR 131)(48 FR 51405, November 8, 1983, as amended 56 FR 64893, December 12, 1991; 59 FR 64344, December 14, 1994; 60 FR 15386, March 23, 1995; 65 FR 24653, April 27, 2000). Separately, the *Water Quality Guidance for the Great Lakes System* regulation (40 CFR 132)(60 FR 15387, March 23, 1995, as amended 65 FR 47874, August 4, 2000; 65 FR 59737, October 6, 2000; 65 FR 66511, November 6, 2000; 65 FR 67650, November 13, 2000) constitutes the guidance for States in the Great Lakes system; and other regulations promulgated by EPA.

State Water Quality Standards

The CWA directs States and authorized Tribes to adopt water quality standards for all their waters and submit their standards to EPA for review and approval or disapproval (of all or part)(CWA section 303(a)(1)-(2), (c)). The CWA further requires that a State shall at least once every three years hold public hearings for the purpose of reviewing applicable water quality standards and, as appropriate, modifying and adopting standards (i.e., conduct triennial reviews). Consultation with EPA is one of the first steps States and Tribes take

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when beginning the triennial review process (EPA 1994). The triennial review process is a forum through which EPA would review and approve or disapprove proposed revisions to State and Tribal water quality standards.

By statute, State and Tribal water quality standards consist of the designated uses of waters and the water quality criteria needed to achieve the designated uses. Section 303(d) and 118(c) of the CWA requires States and Tribes to establish an antidegradation policy. The *Water Quality Standards Regulation* (40 CFR part 131) defines the regulatory requirements of the water quality standards program and adds a provision for States and Tribes to adopt general policies that affect the application and implementation of State and Tribal water quality standards. For example, States and Tribes may adopt policies concerning mixing zones, water quality standards variances, and critical flows for water quality-based permit limits. General policies provide a mechanism for permitting a discharge where numeric water quality criteria may be exceeded, or low flows below which criteria do not apply, or where a standard is not being attained, a variance from the standard would enable permit issuance as long as progress towards attainment is being made. The designated uses, criteria, antidegradation policy, and general policies, together, define the level of protection afforded to a water body or portion thereof under the CWA.

In a January 27, 2005, memorandum (EPA 2005) EPA concluded that ESA section 7 consultation does not apply to EPA's approvals of State and Tribal antidegradation policies because EPA's approval action does not meet the "Applicability" standard defined in the regulations implementing section 7 of the ESA (50 CFR 402.03). Section 402.03 of the ESA section 7 consultation regulations (50 CFR part 402) states that section 7 and the requirements of 50 CFR part 402 apply to all actions in which there is discretionary Federal involvement or control. EPA concluded that they are compelled to approve State and Tribal antidegradation policies if State or Tribal submissions meet all applicable requirements of the *Water Quality Standards Regulation* (40 CFR part 131) and lack discretion to implement measures that would benefit listed species. Consequently, in its analyses the Service can not rely on antidegradation policies in State and Tribal water quality standards to provide protection over and beyond approved water quality criteria. EPA will treat new or revised antidegradation policies as being inapplicable to consultation and thus existing antidegradation policies can be changed by States or Tribes independent of any review of existing water quality criteria.

Water Quality Monitoring

States and Tribes are required to establish water quality monitoring, including collection and analysis of physical, chemical, and biological data to determine abatement and control priorities; developing and reviewing water quality standards, total maximum daily loads, wasteload allocations and load allocations; assessing compliance with NPDES permits by dischargers; reporting information to the public through the section 305(b) report and reviewing site-specific monitoring efforts (40 CFR 130.4).

Continuing Planning Process

Section 303(e) of the CWA requires States and Tribes to have in place a “continuing planning process” (CPP) approved by EPA and requires EPA to periodically review a State’s or Tribe’s planning process for conformity to the requirements of the CWA. Section 303(e)(3) states that EPA shall approve any CPP that will result in plans for all navigable waters that include, but are not limited to, eight elements. EPA’s regulations implementing this section (40 CFR 130) add a ninth element:

1. effluent limitations and schedules of compliance at least as stringent as those required by section 301(b)(1), section 301(b)(2), section 306, and section 307, and at least as stringent as any requirements contained in any applicable water quality standard in effect under section 303;
2. the incorporation of all elements of any applicable areawide waste management plans (CWA section 208) and applicable basin plans (CWA section 209);
3. total maximum daily load for pollutants in accordance with section 303(d);
4. procedures for revision;
5. adequate authority for intergovernmental cooperation;
6. adequate implementation, including schedules of compliance, for revised or new water quality standards under CWA section 303(c);
7. controls over the disposition of all residual waste from any water treatment processing;
8. an inventory and priority ranking of need for waste treatment works to meet applicable requirements of CWA sections 301 and 302; and,
9. a process for determining the priority of permit issuance.

By statute and regulation, an approved CPP is a prerequisite for EPA’s approval of a permitting program under Title IV of the CWA.

Water Quality Management Plans

The CWA and its implementing regulations (40 CFR 130) require States and Tribes to establish Water Quality Management (WQM) Plans to prioritize and direct implementation of CWA programs. The following are required elements of State or Tribal WQM Plans:

1. total maximum daily loads;
2. effluent limitations;
3. municipal and industrial waste treatment;
4. nonpoint source management and control;
5. identification of management agencies capable of carrying out responsibilities;
6. implementation measures necessary to carry out the plan;
7. programs for the control of dredge or fill material;
8. identification of relationship to basin plans developed under CWA section 209;
9. identification of programs for control of ground water pollution.

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WQM Plans are produced and regularly updated on the basis of water quality assessments. Consistency with WQM Plans is a requirement for issuance of permits under CWA section 402 and grants made under the municipal construction grants program. States and Tribes must certify by letter for EPA approval that WQM Plan updates are consistent with all other parts of the plan (40 CFR 130).

In summary, the effects of EPA's approval or promulgation of cyanide criteria in State or Tribal water quality standards must be understood in the larger context of the CWA. This larger context is framed by Congress' stated objective, goals, and policies of the CWA and the programs and activities authorized by the CWA and implemented by EPA, States, Tribes and local governments to achieve these objectives, goals, and policies.

State water quality standards establish the level of protection afforded to water bodies or water body segments. Water quality standards include the designated uses assigned to water bodies, criteria to achieve the designated uses, an antidegradation policy to protect higher quality waters, and general policies which provide for exceedances of criteria values in specified circumstances. The approval or promulgation of cyanide criteria is either interrelated to and/or interdependent with the other elements of State or Tribal water quality standards. The proposed action analyzed in this biological opinion must be compared with the water quality standards as they are approved for individual States and Tribes. Separate, tiered consultations would be necessary to address discrepancies and quantify and exempt incidental take, as appropriate.

The programs of the water quality-based approach to pollution control are targeted to assessing compliance with standards and instituting changes to achieve compliance through modifications to allowable discharges or to the standards themselves. The adaptive management of water quality, articulated in the water quality-based approach to pollution control, is driven by continuing planning processes and water quality management planning and implementation. Because the planning and implementation of CWA programs by EPA, States, Tribes and local governments is targeted towards achieving State or Tribal water quality standards, these activities are necessarily part of a larger action, and thus interrelated to the approval of criteria in State or Tribal water quality standards.

5.0 ACTION AREA

The action area for this consultation consists of all waters subject to the CWA, including "territorial seas," which extend seaward a distance of three miles from the coast, and habitats effected by administration of the CWA. The action area also includes all other parts of watersheds that serve as sources to waters subject to the CWA where activities contribute to point and non-point sources of pollution and are subject to compliance with the CWA. This action area includes such waters and habitats within and surrounding Indian country, the 50 States, and all U.S. territories.

6.0 STATUS OF THE SPECIES AND CRITICAL HABITAT

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Information on the status of affected listed species and critical habitat is presented in two parts. The species and critical habitats considered and the states encompassing their distribution are listed in Table 1, and individual species and critical habitat accounts are presented in Appendix A. Because the action area for this consultation encompasses the entire range of affected listed species and critical habitats, no Environmental Baseline section is needed or presented.

In the Status of the Species and Critical Habitat section of a biological opinion the Service presents biological or ecological information relevant to formulating the opinion, and characterizes the current condition of the species or critical habitat, the factors responsible for that condition, and the survival and recovery needs of the species and the intended conservation function of critical habitat. This information is derived from listing documents and recovery plans or subsequent biological opinions as new information becomes available. The Status of the Species and Critical Habitat evaluation establishes key biological context for evaluating the effects of a proposed Federal action and any cumulative effects for purposes of making an ESA section 7(a)(2) determination(s).

Aquatic life criteria address the CWA goals and policy of attaining water quality that provides for the protection and propagation of fish, shellfish, and wildlife, and prohibiting the discharge of toxic pollutants in toxic amounts (CWA Sec. 101(a)(2) & (3)). The CWA policy of prohibiting the discharge of toxic pollutants in toxic amounts provides a foundation for characterizing EPA's discretion in recommending aquatic life cyanide criteria under CWA section 304(a) and approval of cyanide criteria in state water quality standards. Although most approved water quality standards include a statement prohibiting the discharge of toxic pollutants in toxic amounts, the criteria by which this standard presumably is enforced, the CCC and the CMC, are defined in such a way that adverse effects are limited but not prohibited. \

To fully assess the impacts of the proposed action, it is important to consider the status of the species and the current state of their aquatic environments as this provides a context for the jeopardy analysis. Although the baseline condition includes the past and present impacts of activities in the action area, it does not include the future impacts of the action under review in this consultation. Because the action area for this consultation (i.e. all waters of the U.S., including territories) encompasses the entire ranges for the affected listed species and critical habitats, it is difficult to fully assess 1) the past and present impacts of all Federal, State, or private actions and other human activities in the action area; 2) the anticipated impacts of all proposed Federal projects in the action area that have already undergone formal or early section 7 consultation; and 3) the impact of State or private actions occurring simultaneously with this consultation for each species included in this opinion. As such, we will broadly describe the status of the aquatic environments within the action area as a means for establishing a baseline condition from which to analyze the effect of the action.

Section 304(a)(1) of the CWA requires EPA to develop criteria for water quality for the protection of aquatic life as well as for human health. EPA develops these criteria as numeric limits on the amounts of chemicals that can be present in river, lake, or stream

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water to protect aquatic organisms from death, slower growth, reduced reproduction, and the accumulation of harmful levels of toxic chemicals in their tissues that may also adversely affect consumers of such organisms. Aquatic life criteria have been developed for almost 50 pollutants to date and serve as a basis for the development of state water quality standards. Waters that do not meet these standards are considered to be “impaired” under section 303(d) of the CWA. The most recent National Water Quality Inventory Report to Congress (EPA 2009) presents a summary finding of the 2004 state water quality reports covering 16% of the nation’s 3.5 million miles of rivers and streams, 39% of the nation’s 41.7 million acres of lakes, ponds, and reservoirs, and 29% of the nation’s 87,791 square miles of bays and estuaries. For waters included in the assessment, 44% of the rivers and streams, 64% of the lakes ponds and reservoirs, and 29% of the bays and estuaries were reported as impaired or not clean enough to support their designated uses.

The sources of degradation vary by water type but included pathogens, habitat alterations, mercury, and organic enrichment/oxygen depletion from both known and unknown or unspecified sources. An assessment by the U.S. Geological Survey (USGS) on the presence and concentration of pesticide in the nation’s streams and ground water found that concentrations of pesticides were frequently greater than water-quality benchmarks for aquatic life and fish eating wildlife. Specifically, USGS found that 57% of 83 agricultural streams, 83% of 30 urban streams, and 42% of 65 mixed-land-use streams had concentrations of at least one pesticide that exceeded one or more aquatic-life benchmarks at least one time during the year. The most frequently detected pesticide compounds included atrazine, deethylatrazine, and metolachlor (USGS 2006).

Over 83,000 chemicals are currently listed in the Toxic Substances Control Act Inventory and more than 87,000 chemicals including pesticides, commodity chemicals, naturally occurring non-steroidal estrogens, food additives, cosmetics, nutritional supplements, and representative mixtures have been identified as potential endocrine disruptors by the Endocrine Disruptor Screening and Testing Advisory Committee (1998) convened by EPA. For most of these chemicals, scientific data is insufficient or unavailable for evaluating their potential individual and combined impacts on aquatic environments.

Table 1. Listed species and critical habitats considered in this biological opinion along with their listing status and distribution on a state-by-state basis. E=endangered; T=threatened; CH=critical habitat; XN=(non-essential experimental population).

Common Name	Scientific Name	Status	Distribution
	FISH		
	<i>Acipenseridae</i>		
Gulf sturgeon	<i>Acipenser oxyrinchus desotoi</i>	T, CH	AL, FL, LA, MS
Kootenai River white sturgeon	<i>A. transmontanus</i>	E, CH	ID, MT
Pallid sturgeon	<i>Scaphirhynchus albus</i>	E	AR, IA, IL, KS, KY, LA, MO, MS, MT, ND, NE, SD, TN

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Common Name	Scientific Name	Status	Distribution
Alabama sturgeon	<i>S. suttkusi</i>	E, CH	AL, MS
	<i>Amblyopsidae</i>		
Ozark cavefish	<i>Amblyopsis rosae</i>	T	AR, MO, OK
Alabama cavefish	<i>Speoplatyrhinus poulsoni</i>	E, CH	AL
	<i>Atherinidae</i>		
Waccamaw silverside	<i>Menidia extensa</i>	T, CH	NC
	<i>Catostomidae</i>		
Modoc sucker	<i>Catostomus microps</i>	E, CH	CA, OR
Santa Ana sucker	<i>Catostomus santaanae</i>	T, CH	CA
Warner sucker	<i>Catostomus warnerensis</i>	T, CH	OR
Shortnose sucker	<i>Chasmistes brevirostris</i>	E	CA, OR
Cui-ui	<i>Chasmistes cujus</i>	E	NV
June sucker	<i>Chasmistes liorus</i>	E, CH	UT
Lost River sucker	<i>Deltistes luxatus</i>	E	CA, OR
Razorback sucker	<i>Xyrauchen texanus</i>	E, CH	AZ, CA, CO, NM, NV, UT
	<i>Cotidae</i>		
Pygmy sculpin	<i>Cottus paulus</i>	T	AL
	<i>Cyprinidae</i>		
Blue shiner	<i>Cyprinella caerulea</i>	T	AL, GA, TN
Beautiful shiner	<i>Cyprinella Formosa</i>	T, CH	AZ, NM
Devils River minnow	<i>Dionda diaboli</i>	T, CH	TX
Spotfin chub	<i>Erimonax monachus</i>	T, CH, XN	AL, NC, TN, VA
Slender chub	<i>Erimystax cahni</i>	T, CH, XN	TN, VA
Mojave tui chub	<i>Gila bicolor mohavensis</i>	E	CA
Owens tui chub	<i>Gila bicolor snyderi</i>	E, CH	CA
Borax Lake chub	<i>Gila boraxobius</i>	E, CH	OR
Humpback chub	<i>Gila cypha</i>	E, CH	AZ, CO, UT
Sonora chub	<i>Gila ditaenia</i>	T, CH	AZ
Bonytail chub	<i>Gila elegans</i>	E, CH	AZ, CA, CO, NV, UT
Gila chub	<i>Gila intermedia</i>	E, CH	AZ, NM
Yaqui chub	<i>Gila purpurea</i>	E, CH	AZ
Pahranagat roundtail chub	<i>Gila robusta jordani</i>	E	NV
Virgin River Chub	<i>Gila seminuda (=robusta)</i>	E, CH	AZ, NV, UT
Rio Grande silvery minnow	<i>Hybognathus amarus</i>	E, CH	NM, TX
Big Spring spinedace	<i>Lepidomeda mollispinis pratensis</i>	T, CH	NV
Little Colorado spinedace	<i>Lepidomeda vittata</i>	T, CH	AZ
Spikedace	<i>Meda fulgida</i>	T, CH	AZ, NM
Moapa dace	<i>Moapa coriacea</i>	E	NV
Palezone shiner	<i>Notropis albizonatus</i>	E	AL, KY
Cahaba shiner	<i>Notropis cahabae</i>	E	AL
Arkansas River shiner	<i>Notropis girardi</i>	T, CH	AR, KS, NM, OK,

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Common Name	Scientific Name	Status	Distribution
			TX
Cape Fear shiner	<i>Notropis mekistocholas</i>	E, CH	NC
Pecos bluntnose shiner	<i>Notropis simus pecosensis</i>	T, CH	NM
Topeka shiner	<i>Notropis topeka (=tristis)</i>	E, CH	IA, KS, MN, MO, NE, SD
Oregon chub	<i>Oregonichthys crameri</i>	E	OR
Blackside dace	<i>Phoxinus cumberlandensis</i>	T	KY, TN
Woundfin	<i>Plagopterus argentissimus</i>	E; CH, XN	AZ, NM, NV, UT
Colorado pikeminnow (=squawfish)	<i>Ptychocheilus lucius</i>	E; CH, XN	AZ, CA, CO, NM, UT
Ash Meadows speckled dace	<i>Rhinichthys osculus nevadensis</i>	E, CH	NV
Kendall Warm Springs dace	<i>Rhinichthys osculus thermalis</i>	E	WY
Loach minnow	<i>Tiaroga cobitis</i>	T, CH	AZ, NM
	Gasterosteidae		
Unarmored threespine stickleback	<i>Gasterosteus aculeatus williamsoni</i>	E	CA
	Gobiidae		
Tidewater goby	<i>Eucyclogobius newberryi</i>	T, CH	CA
	Goodeidae		
White River springfish	<i>Crenichthys baileyi baileyi</i>	E, CH	NV
Hiko White River springfish	<i>Crenichthys baileyi grandis</i>	E, CH	NV
Railroad Valley springfish	<i>Crenichthys nevadae</i>	T, CH	NV
	Osmeridae		
Delta smelt	<i>Hypomesus transpacificus</i>	T, CH	CA
	Percidae		
Slackwater darter	<i>Etheostoma boschungii</i>	T, CH	AL, TN
Vermilion darter	<i>Etheostoma chermocki</i>	E	AL
Relict darter	<i>E. chienense</i>	E	KY
Etowah darter	<i>E. etowahae</i>	E	GA
Fountain darter	<i>E. fonticola</i>	E, CH	TX
Niangua darter ¹	<i>E. nianguae</i>	T, CH	MO
Watercress darter	<i>E. nuchale</i>	E	AL
Okaloosa darter	<i>E. okaloosae</i>	E	FL
Duskytail darter	<i>E. percnurum</i>	E, XN	KY, TN, VA
Bayou darter	<i>E. rubrum</i>	T	MS
Cherokee darter	<i>E. scotti</i>	T	GA
Maryland darter	<i>E. sellare</i>	E, CH	MD
Bluemask darter	<i>E. sp.</i>	E	TN
Boulder darter	<i>E. wapiti</i>	E, XN	AL, TN
Amber darter	<i>Percina antesella</i>	E, CH	GA, TN
Goldline darter	<i>P. aurolineata</i>	T	AL, GA

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Common Name	Scientific Name	Status	Distribution
Conasauga logperch	<i>P. jenkinsi</i>	E, CH	GA, TN
Leopard darter	<i>P. pantherina</i>	T, CH	AR, OK
Roanoke logperch	<i>P. rex</i>	E	VA, NC
Snail darter	<i>P. tanasi</i>	T, CH	AL, GA, TN
	<i>Poeciliidae</i>		
Big Bend gambusia	<i>Gambusia gaigei</i>	E	TX
San Marcos gambusia	<i>Gambusia georgei</i>	E, CH	TX
Clear Creek gambusia	<i>Gambusia heterochir</i>	E	TX
Pecos gambusia	<i>Gambusia nobilis</i>	E	NM, TX
Gila topminnow	<i>Poeciliopsis occidentalis</i>	E	AZ, NM
	<i>Salmonidae</i>		
Bull trout	<i>Salvelinus confluentus</i>	T, CH	CA, ID, MT, NV, OR, WA
Little Kern Golden trout	<i>Oncorhynchus aquabonita whitei</i>	T, CH	CA
Apache trout	<i>O. apache</i>	T	AZ
Lahontan Cutthroat trout	<i>O. clarkii henshawi</i>	T	CA, NV, OR, UT
Paiute Cutthroat trout	<i>O. clarkii seleniris</i>	T	CA
Greenback Cutthroat Mountain trout	<i>O. clarkii stomias</i>	T	CO
Gila trout	<i>O. gilae</i>	T	AZ, NM
Atlantic salmon	<i>Salmo salar</i>	E	ME
	AMPHIPODS		
Illinois cave amphipod	<i>Gammarus acherondytes</i>	E	IL
Noel's amphipod	<i>G. desperatus</i>	E	NM
	MUSSELS		
Cumberland elktoe	<i>Alasmidonta atropurpurea</i>	E, CH	KY, TN
Dwarf wedgemussel	<i>A. heterodon</i>	E	CT, MA, MD, NC, NH, NJ, NY, PA, VA, VT
Appalachian elktoe	<i>A. raveneliana</i>	E, CH	NC, TN
Fat three-ridge	<i>Amblema neislerii</i>	E, CH	FL, GA
Ouachita rock pocketbook	<i>Arkansia wheeleri</i>	E	AR, OK
Birdwing pearlymussel	<i>Conradilla caelata</i>	E, XN	AL, TN, VA
Fanshell	<i>Cyprogenia stegaria</i>	E, XN	AL, IL, IN, KY, OH, TN, VA, WV
Dromedary pearlymussel	<i>Dromus dromas</i>	E, XN	AL, KY, TN, VA
Chipola slabshell	<i>Elliptio chipolaensis</i>	T, CH	AL, FL
Tar River spinymussel	<i>E. steinstansana</i>	E	NC
Purple bankclimber	<i>Elliptoideus sloatianus</i>	T, CH	FL, GA
Cumberlandian combshell	<i>Epioblasma brevidens</i>	E, XN, CH	AL, KY, TN, VA
Oyster mussel	<i>E. capsaeformis</i>	E, XN, CH	AL, GA, KY, NC, TN, VA

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Common Name	Scientific Name	Status	Distribution
Curtis pearlymussel	<i>(E. florentina curtisii)</i>	E	AR, MO
Yellow blossom	<i>E. florentina florentina</i>	E, XN	AL, TN
Tan riffleshell	<i>E. florentina walkeri</i>	E	KY, TN, VA
Upland combshell	<i>E. metastrata</i>	E, CH	AL, GA, TN
Catspaw	<i>E. obliquata obliquata</i>	E, XN	AL, IL, IN, KY, OH, TN
White catspaw	<i>E. o. perobliqua</i>	E	IN, OH
Southern acornshell	<i>E. othcaloogensis</i>	E, CH	AL, GA, TN
Southern combshell	<i>E. penita</i>	E	AL, MS
Green blossom	<i>E. torulosa gubernaculum</i>	E	TN, VA
Northern riffleshell	<i>E. t. rangiana</i>	E	IN, KY, MI, OH, PA, WV
Tubercled blossom	<i>E.a torulosa torulosa</i>	E, XN	AL, IL, IN, KY, TN, WV
Turgid blossom	<i>Epioblasma turgidula</i>	E, XN	AL, TN
Shiny pigtoe	<i>Fusconaia cor</i>	E, XN	AL, TN, VA
Finerayed pigtoe	<i>Fusconaia cuneolus</i>	E, XN	AL, TN, VA
Cracking pearlymussel	<i>Hemistena lata</i>	E, XN	AL, IN, KY, PA, TN, VA
Pink mucket	<i>Lampsilis abrupta</i>	E	AL, AR, IL, IN, KY, LA, MO, OH, PA, TN, WV
Fine-lined pocketbook	<i>Lampsilis altilis</i>	T, CH	AL, GA, TN
Higgins eye	<i>Lampsilis higginsii</i>	E	IA, IL, MN, MO, WI
Orangenacre mucket	<i>Lampsilis perovalis</i>	T, CH	AL, MS
Arkansas fatmucket	<i>Lampsilis powelli</i>	T	AR
Speckled pocketbook	<i>Lampsilis streckeri</i>	E	AR
Shinyrayed pocketbook	<i>Lampsilis subangulata</i>	E, CH	AL, FL, GA
Alabama lampmussel	<i>Lampsilis virescens</i>	E, XN	AL, TN
Carolina heelsplitter	<i>Lasmigona decorata</i>	E, CH	NC, SC
Scaleshell mussel	<i>Leptodea leptodon</i>	E	AR, MO, OK
Louisiana pearlshell	<i>Margaritifera hembeli</i>	T	LA
Alabama moccasinshell	<i>Medionidus acutissimus</i>	T, CH	AL, GA, MS
Coosa moccasinshell ¹	<i>Medionidus parvulus</i>	E, CH	GA, TN
Gulf moccasinshell	<i>Medionidus penicillatus</i>	E, CH	FL, GA
Ochlockonee moccasinshell	<i>Medionidus simpsonianus</i>	E, CH	FL, GA
Ring pink	<i>Obovaria retusa</i>	E, XN	AL, IN, KY, PA, TN
Littlewing pearlymussel	<i>Pegias fibula</i>	E	KY, NC, TN, VA
White wartyback pearlymussel	<i>Plethobasus cicatricosus</i>	E, XN	AL, IN, KY, TN
Orangefoot pimpleback	<i>Plethobasus cooperianus</i>	E, XN	AL, IL, IN, KY, PA, TN
Clubshell	<i>Pleurobema clava</i>	E, XN	AL, IN, KY, MI, OH, PA, WV
James spinymussel	<i>Pleurobema collina</i>	E	NC, VA, WV
Black clubshell	<i>Pleurobema curtum</i>	E	MS

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Common Name	Scientific Name	Status	Distribution
Southern clubshell	<i>Pleurobema decisum</i>	E, CH	AL, GA, MS
Dark pigtoe	<i>Pleurobema furvum</i>	E, CH	AL
Southern pigtoe	<i>Pleurobema georgianum</i>	E, CH	AL, GA, TN
Cumberland pigtoe	<i>Pleurobema gibberum</i>	E	TN
Flat pigtoe	<i>Pleurobema marshalli</i>	E	AL, MS
Ovate clubshell	<i>Pleurobema perovatatum</i>	E, CH	AL, MS
Rough pigtoe	<i>Pleurobema plenum</i>	E, XN	AL, IN, KY, PA, TN, VA
Oval pigtoe	<i>Pleurobema pyriforme</i>	E, CH	FL, GA
Heavy pigtoe	<i>Pleurobema taitianum</i>	E	AL
Fat pocketbook	<i>Potamilus capax</i>	E	AR, IL, IN, KY, LA, MO, MS
Alabama heelsplitter	<i>Potamilus inflatus</i>	T	AL, LA
Triangular kidneyshell	<i>Ptychobranchnus greeni</i>	E, CH	AL, GA, TN
Rough rabbitsfoot	<i>Quadrula cylindrica strigillata</i>	E, CH	TN, VA
Winged mapleleaf	<i>Quadrula fragosa</i>	E, XN	AR, MN, OK, WI
Cumberland monkeyface	<i>Quadrula intermedia</i>	E, XN	AL, TN, VA
Appalachian monkeyface	<i>Quadrula sparsa</i>	E, XN	TN, VA
Stirrupshell	<i>Quadrula stapes</i>	E	AL, MS
Pale lilliput	<i>Toxolasma cylindrellus</i>	E	AL, TN
Purple bean	<i>Villosa perpurpurea</i>	E, CH	TN, VA
Cumberland bean	<i>Villosa trabalis</i>	E, XN	AL, KY, TN, VA
	AMPHIBIANS		
	<i>Ambystomatidae</i>		
Reticulated flatwoods salamander	<i>Ambystoma bishop</i>	E, CH	AL, FL, GA
Frosted flatwoods Salamander	<i>A. cingulatum</i>	T, CH	FL, GA, SC
California tiger salamander	<i>A. californiense</i> Central California DPS	T, CH	CA
California tiger salamander	<i>A. californiense</i> Santa Barbara County DPS	E, CH	CA
California tiger salamander	<i>A. californiense</i> Sonoma County DPS	E	CA
Santa Cruz long-toed salamander	<i>A. macrodactylum croceum</i>	E	CA
Sonora tiger salamander	<i>A. tigrinum stebbinsi</i>	E	AZ
	<i>Plethodontidae</i>		
San Marcos salamander	<i>Eurycea nana</i>	T, CH	TX
Barton Springs salamander	<i>E. sosorum</i>	E	TX
Texas blind salamander	<i>Typhlomolge rathbuni</i>	E	TX
	<i>Bufo</i>		
Wyoming toad	<i>Bufo baxteri</i>	E	WY

Common Name	Scientific Name	Status	Distribution
Arroyo toad	<i>B. californicus</i>	E, CH	CA
Houston toad	<i>B. houstonensis</i>	E, CH	TX
	<i>Eleutherodactylidae</i>		
Guajon	<i>Eleutherodactylus cooki</i>	T, CH	PR
	<i>Ranidae</i>		
California red-legged frog	<i>Rana aurora draytonii</i>	T, CH	CA
Chiricahua leopard Frog	<i>R. chiricahuensis</i>	T	AZ, NM
Mountain yellow-legged frog	<i>R. muscosa</i>	E, CH	CA, NV

7.0 EFFECTS OF THE ACTION

Our analysis of the effects of the proposed action on listed species and critical habitats includes a discussion of the sources of cyanide, an explanation of our assumptions about cyanide exposure, an overview of cyanide toxicity, an analysis of effects to listed fish, followed by analyses of effects to listed amphibians and invertebrates including freshwater mussels. The taxon-specific analyses (i.e. fish, amphibians, invertebrates, mussels) considered information on cyanide toxicity, factors influencing toxicity, approaches for estimating effects on individuals, and discussions of population-level responses. Our analyses were dependent on the available information, which varied among taxa. For fish, the available chronic toxicity data enabled us to develop a method for estimating the magnitude of effect on fish species. For other taxa we exercised best professional judgement to estimate effects based on existing data. These sections provide the basis for our discussion of effects to individual listed species and their designated critical habitat.

General Sources of Cyanide

Cyanide is ubiquitous in the environment, and enters waterways as a point source and nonpoint source of pollution. Cyanide is produced synthetically to support industrial uses, it is found naturally in such foods as oil of bitter almonds, cassava, cherry pits, and various microorganisms produce cyanide (Leduc 1984, Eisler 2000, Dzombak et al. 2006). Cyanides are used widely in ore-extraction, steel and heavy metal industries (e.g., electroplating), the manufacture of synthetic fabrics and plastics, some chlorination treatment plants, as a pesticide and as an intermediate ingredient in herbicides, in road salts, and until recently some fire retardants (see Table 2). Anthropogenic sources contribute the vast majority of cyanide in the environment. Certain activities directly release cyanide into the environment, or cyanide may be produced as the byproduct of activities. Industrial activities that produce cyanide as a byproduct include municipal waste and sludge incineration and coking and gasification of coal, to name a few. Metal industries and organic chemical industries are major contributors of cyanide into the freshwater aquatic environment, whereas, atmospheric cyanide, a by-product of forests fires, may be the primary source of oceanic cyanide (Leduc 1984, EPA 2005, Dzombak et al. 2006).

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Compared to storm water runoff, urban roadway snow exposed to traffic and winter maintenance practices has a much greater capacity to accumulate and retain heavy metals and other pollutants. In a study of urban highway sites, concentrations of cyanide and metals were orders of magnitude higher than at the control sites and exceeded storm water runoff concentrations by one to two orders of magnitude. Cyanide levels, although demonstrating some variability, remained relatively constant at all sites, averaging 154 µg/L, or cyanide concentrations increased according to the increased application of deicing salts that contained cyanide compounds as anti-caking agents (Glenn and Sansalone 2002).

A study on the effect of cyanide on the anaerobic treatment of synthetic wastewater noted that cyanide is produced on an industrial scale of 2–3 million tons per year and, therefore is in many different industrial waste waters. The concentrations encountered in industrial waste generally are in the range 0.01–10,000 mg/L, most of it in complexed species of cyanide, which are less toxic than free cyanide. Cyanide contamination also occurs in the processing of agricultural crops containing high concentrations of this compound, such as cassava (Gijzen et al. 2000). Systematic surveys of large wastewater effluents in Southern California support a low estimate of free cyanide in wastewaters. In different years reported from 1992 – 2002, mean cyanide concentrations in effluents ranged from <2 to 30 µg/L (Steinberger and Stein 2003).

A more widespread risk of cyanide poisoning of aquatic life is likely from biomass burning (e.g., burning waste biomass for energy conversion, crop burning, prescribed forest fires and wildfires). Barber et al. (2003) examined releases of cyanides from biomass burning and their effect on surface runoff water. In laboratory test burns, available cyanide concentrations in leachate from residual ash were much higher than in leachate from partially burned and unburned fuel and were similar to or higher than a 96-h median lethal concentration (LC50) for rainbow trout (45 µg/L). Free cyanide concentrations in stormwater runoff collected after a wildfire in North Carolina averaged 49 µg/L, an order of magnitude higher than in samples from an adjacent unburned area (Barber et al. 2003).

According to the Toxics Release Inventory, cyanide compound releases to land and water totaled about 1.5 million lbs. between 1987 and 1993, of which about 65 percent was to water. The largest releases (combined land and water releases) occurred in California and Pennsylvania. Releases in California were land-based, while the releases in Pennsylvania occurred primarily in water (EPA 2005). A review of STORET data in 1981, reported the mean cyanide concentration in surface waters of the United States was less than 3.5 µg/L, with concentrations in some industrial areas exceeding 200 µg/L (Dzombak et al. 2006). Coincidentally, production of HCN, one of most toxic forms of cyanide in water, has more than doubled in the United States since the early 1980s (from 330,000 tons per year in 1983 to 750,000 tons per year in 2001).

Table 2. Forms of cyanides and their uses.

Form	Uses	Reference
Cyanide salts (potassium	Steel manufacturing & heat-treating facilities Metal cleaning, electroplating	IPCS; Leduc 1984; EPA 2005

cyanide, sodium cyanide)	Ore-extraction (gold-mining, coke extraction)	IPCS; Leduc 1984; EPA 2005
	Dyeing, printing of photographs	IPCS; EPA 2005
	Production of resin monomers (acrylates)	IPCS
	Fire retardants ¹	Little and Calfee 2002
	Anti-caking agent for road salts	Dzombak et al. 2006
	Pharmaceuticals (antibiotics, steroids, chemotherapy)	Dzombak et al. 2006
Hydrogen cyanide, organocyanides, metallo-cyanide compounds	Fumigant/pesticide	IPCS, Dzombak et al. 2006
	Herbicides (dichlobenil, bromoxynil, bantrol)	EPA 2005, Dzombak et al. 2006
	Road salts	EPA 2005
	Production of other cyanides (e.g., sodium cyanide for gold mining)	EPA 2005, Dzombak et al. 2006
	Pyrolysis of paper, wool, polyurethane	IPCS
	Chelating agents for water and wastewater treatment	EPA 2005, Dzombak et al. 2006
	Production of clear plastics	Dzombak et al. 2006
	Methionine ² for animal food supplement	Dzombak et al. 2006

¹ As of 2007 sodium ferrocyanide was no longer an accepted ingredient in fire retardants (Long Term Retardants) used in the U.S. (U.S. Forest Service. 2007. Specification 5100-304c Long Term Retardant, Wildland Firefighting)

² Hydrogen cyanide is used in the manufacturing of methionine

The risk to aquatic environments from cyanide releases depends on several factors including: the form and concentration of cyanide released, water pH, the presence of metallic trace elements like iron, degree of solar radiation, air and water temperatures, and the presence of natural cyanide sinks (Dzombak et al. 2006). Aqueous cyanide readily evolves from hydrogen cyanide, metalocyanide complexes, and organocyanides, and from metal-cyanide solids. Solid forms of cyanide may exist in the soil of sites for years, and once exposed to water may result in dissolved cyanide reaching ground water and eventually surface waters (see Dzombak et al.'s [2006] discussion about the industrial legacy of cyanide box wastes at thousands of former manufactured gas plants in the United States). Free cyanide readily biodegrades to carbon dioxide and the ammonia ion but its fate depends upon water temperatures, dissolved oxygen levels, mixing, and nutrients (Young et al. 2006).

While available free cyanide is the primary toxic agent in water and is the form expressed by the aquatic life criteria reported by EPA (1985; see Table 1), total cyanide is most commonly measured in discharges (EPA 1985, Dzombak et al. 2006). Measurements are frequently conducted via colorimetric, titrimetric, or electrochemical finish techniques (Dzombak et al. 2006). Measurements of total cyanide are limited to detection in a reagent water matrix of about 1 to 5 µg/L and do not measure: cyanates, thocyanates, most organic-cyanide compounds, and most cobalt and platinum cyanide complexes (Dzombak

et al. 2006). Problems with sample storage, regulatory criteria, and the methods for testing and their sensitivity are a concern (Eisler 2000, Dzombak et al. 2006). Eisler (2000) notes that due to the volatilization of cyanide, periodic monitoring is not informative (for example, monitoring once per quarter [see EPA 2006]). Consequently, Eisler (2000) and others recommend that continuous monitoring systems are necessary, with particular emphasis on industrial dischargers, to understand the fate and transport, critical exposures, and relative contributions of human and natural sources of cyanide in the aquatic environment.

It should be noted that EPA's request for ESA section 7 consultation on the aquatic life criteria for cyanide is the first consultation request on its recommended aquatic life criteria. Cyanide will co-occur in waters of the United States in mixture with numerous other pollutants. These other pollutants may or may not have criteria established under section 304(a) of the CWA.

Basis for Assuming Exposure to Cyanide at Criteria Concentrations

We based our effects analysis in this Biological Opinion on the premise that the proposed action could be implemented fully. Listed aquatic species and their critical habitats could be exposed to cyanide concentrations in the water column at concentrations consistent with the CMC and CCC. These criteria, as approved by EPA in State and Tribal water quality standards, become the "protection level" to which the water quality-based approach to pollution control is applied. In freshwater systems, this protection level is met if the one-hour average concentration in waters does not exceed 22 ug/L cyanide more than once every three years on average; AND, the four-day average concentration does not exceed 5.2 ug/L cyanide more than once every three years on average. For saltwater systems, the one-hour average concentration should not exceed 1.0 ug/L more than once every three years on the average.

Our rationale for assuming exposure to cyanide at the CMC and CCC includes the following:

1. Section 7 consultation is future-oriented, focusing on the potential effects of the *proposed* action.
2. The CMC and CCC represent the basis for administering water quality programs under the water quality-based approach to pollution control, including monitoring to determine whether waters are attaining designated uses, 303(d) listing of impaired waters, and the development and implementation of TMDLs.
3. We can make no reasonable assumption that any additional protections afforded by antidegradation policies will persist in the future, given EPA's conclusion that section 7 consultation does not apply to EPA approval of new or revised antidegradation policies.
4. There are no other elements of the action proposed that would limit the timing, duration, frequency, or magnitude of exposure of listed species and critical habitats to cyanide pollution other than those defined by the CMC and CCC. General Policies could have the effect of relaxing limits on exposure.

5. Approval of cyanide concentrations at the CMC and CCC can result in exposures that vary through time and space or occur uniformly throughout waterbodies. Exposure to cyanide pollution at the CMC and CCC could occur to portions of populations, whole populations, or the full range of a species.

Physiology of Cyanide Toxicity

The adverse effects of cyanide on fish and other aquatic species have been well documented (Leduc 1984, Eisler 2000, Gensemer et al. 2006, Lanno and Menzie 2006) and are discussed in subsequent sections of this document. Our knowledge of the basic mechanisms by which cyanide exerts its toxic effects, however, has been gained largely as a result of human health-related studies with rodents and other mammals. Recent reviews of the mammalian literature (ATSDR 2006, Borowitz et al. 2006) describe the physiological mechanisms for cyanide toxicity and are summarized below.

Cyanide is known to affect several organ systems including the central nervous system, heart, liver and kidney, although, the principal sites of action are the brain and heart. Cyanide is a potent respiratory toxin which inhibits mitochondrial respiration and can induce rapid lethal responses following acute exposures. Cyanide preferentially binds to the ferric iron atom of the metalloenzyme, cytochrome c oxidase, which is the terminal oxidase in the mitochondrial membrane respiratory chain (electron transport system). The binding of cyanide to the ferric iron atom inhibits the reaction by which electrons are transferred from reduced cytochrome to oxygen thereby blocking oxidative metabolism and ATP production. Blockage of oxidative metabolism can disrupt calcium homeostasis by stimulating calcium release from intracellular stores and enhance influx of extracellular calcium. Elevated cellular calcium activates numerous biochemical pathways including those that lead to nerve cell death via necrosis and apoptosis (programmed cell death). Cytotoxicity is common following acute lethal cyanide exposures. Impaired cellular oxygen utilization results in hypoxia, a shift from aerobic to anaerobic metabolism, and a build up of lactic acid. Because neural tissues have a high oxygen demand and cannot function under low oxygen, the brain is particularly sensitive. The effects of cellular hypoxia and lactate acidosis depress the central nervous system and, at lethal cyanide concentrations, lead to respiratory arrest and death.

Cyanide also binds to other metalloenzymes such as catalase, peroxidase, methemoglobin, hydroxocobalamin, phosphatase, tyrosinase, ascorbic acid oxidase, xanthine oxidase, and succinic dehydrogenase (ATSDR 2006). Modulation of these enzymes contributes to other manifestations of cyanide toxicity. For example, oxidative stress plays an important role in cyanide-induced neurotoxicity. Inhibition of antioxidant enzymes (e.g., catalase) promotes oxidative stress because these enzymes provide protection from the adverse effects (such as lipid peroxidation) that are caused by reactive oxygen species (ROS). In addition, elevated cellular calcium levels (described above) have been associated with increased concentrations of ROS (Borowitz et al. 2006). The effects of rising ROS and antioxidant enzyme inhibition can exacerbate nerve cell damage. Cyanide can also cause the release of excitatory neurotransmitters (e.g., dopamine) in the brain and can initiate the release of catecholamines from the adrenals and adrenergic nerve terminals (Smith 1996).

Modulation of catecholamine biosynthesis and neurotransmitter release may help explain some of the acute and chronic neurological effects of cyanide (Borowitz et al. 2006).

The major route for cyanide elimination from the body is via enzyme-mediated thiocyanate formation in the kidney and excretion in urine. Rhodanase is the enzyme that catalyzes the transformation of thiosulfate to thiocyanate and its activity is limited by the availability of sulfur. Although thiocyanate (SCN^-) is the principle form of cyanide that is eliminated, it can also accumulate in tissues and is known to have antithyroidal properties. SCN^- inhibits iodine uptake by thyroid tissues and disrupts thyroid hormone homeostasis which can result in the development of goiter.

In the aquatic environment, the primary route of exposure for fish, amphibians, and aquatic invertebrates is via water. Cyanide enters the body through semipermeable membranes such as gills, egg capsules and other sites where gas exchange and osmoregulatory processes occur (Eisler 2000). Cyanide does not tend to bioaccumulate in aquatic biota so dietary uptake (at criteria concentrations) is not considered to be an important exposure pathway (Lanno and Menzie 2006).

7.1 Effects to Fish

Acute Toxicity to Fish

Knowledge of the acute lethal effects of cyanide on fish has been gained through observations following accidental spills, intentional field applications for lake/stream management, and controlled laboratory studies. Massive kills of fish and other aquatic organisms have been observed following accidental spills of cyanide from storage reservoirs, overturned rail tank cars, and other sources (Leduc 1984). Some of the most catastrophic releases have been from gold mine heap leaching operations, where cyanide is used in the gold extraction process (Wong-Chong et al. 2006). Releases from waste heap leaching pads and tailing storage ponds have discharged large quantities of cyanide into surface waters. In 1997, 245,000 gallons of cyanide solution leaked from a heap leach pad at the Gold Quarry Mine, NV into two nearby creeks, and 7 million gallons of treated leach solution (0.2 ppm cyanide) was released from storage ponds at the USMX Mine, UT into the East Fork of Beaver Dam Wash (Wong-Chong et al. 2006). One of the largest spills occurred in Baia Mare, Romania where 26 million gallons of cyanide-bearing tailings were released due to a failure in the tailings dam, killing an estimated 1,240 tons of fish in the Hungarian portion of the Tisza River alone (Wong-Chong et al. 2006). Because of its high toxicity and relatively short half-life, cyanide has been used by fishery managers for lake restoration (Leduc 1984) and for collecting fish from ponds, lakes and streams (Lewis and Tarrant 1960). Following treatment, poisoned fish exhibit several symptoms including increased ventilation, surfacing, gulping for air, frantic swimming in circles, convulsions, tremors and finally death (Leduc 1984). At high levels of exposure the onset of acute toxicity occurs rapidly, however, live fish that are rescued and transferred to clean water may survive (Leduc 1984).

Laboratory tests under controlled conditions have revealed that not all life stages of fish are equally sensitive to acute cyanide exposure, that cyanide toxicity can be modulated by abiotic factors, and that there is a wide range in sensitivity among aquatic organisms. Smith et al. (1978) conducted 96-hr acute toxicity tests with fathead minnow, bluegill, brook trout, and yellow perch and found that juveniles and fry were more sensitive to the lethal effects of cyanide than eggs. Bluegill, yellow perch, and brook trout juveniles were more sensitive than newly-hatched fry, where as, swim-up fry were the most sensitive fathead minnow life stage.

Smith et al. (1978) also reported that dissolved oxygen (DO) concentration and water temperature affect the susceptibility of these species to cyanide toxicity. Cyanide was more toxic when fish were held in water with lower DO concentrations. Considering that cyanide is a respiratory toxin that inhibits oxidative metabolism, it is not surprising that the effects are exacerbated under conditions where oxygen availability is limited. The authors also tested the effect of temperature on cyanide toxicity. LC₅₀ values for juvenile brook trout, bluegill and yellow perch were lower at low temperatures than at higher temperatures, indicating a heightened sensitivity at lower temperatures. The combination of low DO and low temperature tended to produce conditions that rendered juveniles most susceptible to cyanide toxicity.

Kovacs (1979) found that the sensitivity of juvenile rainbow trout to cyanide was similarly influenced by temperature. He conducted 96-hr acute toxicity tests at three different temperatures (6, 12 and 18° C) and reported LC₅₀ values of 28, 42, and 68 ug HCN/L, respectively. Thus, trout held at 6° C were 2.4 times more sensitive to cyanide than trout held at 18° C. These studies demonstrate that life stage as well as abiotic factors (DO and temperature) can influence acute cyanide toxicity. There is also considerable variability among aquatic taxa in terms of their intrinsic sensitivity to cyanide.

Standardized acute toxicity tests with cyanide have been conducted with numerous aquatic species. EPA compiled toxicity data for 83 species of aquatic animals and plants (61 freshwater species and 22 saltwater species) as part of their cyanide BE (EPA 2007). Based on this compilation, there appears to be a large range in sensitivity between the most sensitive (rock crab LC₅₀ 4.89 ug CN/L) and the least sensitive species tested (river snail LC₅₀ 760,000 ug CN/L). Freshwater species represented 9 phyla, 15 classes, 29 orders, 36 families, and 52 genera. Fishes were among the most sensitive freshwater taxa although there was substantial variability in sensitivity. Among the 24 freshwater fish species included in the list, there was a 33-fold difference in sensitivity between the most sensitive (rainbow trout, *Oncorhynchus mykiss*, LC₅₀ 59 ug CN/L) and the least sensitive (bata, *Labeo bata*, LC₅₀ 1970 ug CN/L). The 8 most sensitive fish species belong to 3 different families: Salmonidae (3 species, 3 genera); Percidae (2 species, 1 genera); and Centrarchidae (3 species, 3 genera). Because of the relatively low number of species that have been tested within these families, it is difficult to get a sense of the amount of intra-family variability in species sensitivity on the low end of the species sensitivity distribution. By contrast, the family Cyprinidae was well represented with 10 different species representing 8 genera. Among those 10 species, there is an 18-fold difference in sensitivity between the most sensitive (roach LC₅₀ 108 ug CN/L) and the least sensitive

(bata, *Labeo bata*, LC₅₀ 1970 ug CN/L) species. Because of pronounced intra-family variation it is unlikely that the 8 species within the 3 most sensitive families represent the most sensitive species within those families.

Dwyer et al. (2005) conducted acute toxicity tests with 5 different water pollutants (carbaryl, copper, 4-nonylphenol, pentachlorophenol, and permethrin), 3 common test species (rainbow trout, fathead minnow, and sheepshead minnow) and 17 federally threatened, endangered or candidate fish species. They found that for some pollutants, threatened and endangered species in the following families were as or more sensitive than rainbow trout: Salmonidae (Apache trout, Greenback Cutthroat trout, and Lahontan Cutthroat trout), Percidae (Fountain darter and Greenthroat darter), Cyprinidae (Cape Fear shiner, Spotfin chub, and Colorado pikeminnow), and Acipenseridae (Atlantic sturgeon and Shortnose sturgeon). Based on these results, the authors recommended that for listed fish species which require greater protection (i.e., species that may be more sensitive than rainbow trout), a factor of 0.63 can be applied to the geometric mean of the rainbow trout LC₅₀, and that if even greater protection is desired a factor of 0.46 can be used (0.63 – 1 standard deviation). For cyanide, applying the adjustment factors of 0.63 and 0.46 to the rainbow trout LC₅₀ (59 ug CN/L) would result in LC₅₀ estimates of 27 to 37 ug CN/L for sensitive listed fish species.

Chronic Toxicity to Fish

Chronic cyanide toxicity tests have been conducted with relatively few fish species. However, available data indicate that cyanide not only reduces survival but also affects reproduction, growth, swimming performance, condition, and development (Table 3). Reproduction appears to be one of the most sensitive endpoints. Full and partial life cycle tests with fathead minnows and brook trout have shown that fish exposed to sublethal concentrations of cyanide spawned fewer eggs than non-exposed fish (Koenst et al. 1977, Lind et al. 1977). Fecundity was reduced by 57.8% and 46.9% (compared to controls) in female fathead minnows exposed to cyanide at 19.6 ug HCN/L (the LOEC) and 12.9 ug HCN/L (the NOEC), respectively. Similarly, the mean number of eggs spawned by brook trout was reduced by 53.3% at 11.2 ug HCN/L and by 17.7% at 5.7 ug HCN/L.

Kimball et al. (1978) exposed bluegill to cyanide (5.2 – 80 ug HCN/L) for 289 days and reported that no eggs were spawned in any of the cyanide treatments except for one spawning that occurred at the highest concentration. Although the single spawning is difficult to explain, the fact that spawning was completely inhibited in 42 of 43 cyanide-exposed females suggests that bluegill may be particularly sensitive. It is surprising, considering the overwhelming effects on reproduction, that a confirmatory study with bluegills has not been conducted over the past 30 years.

Cheng and Ruby (1981) studied the effects of pulsed exposures of cyanide on flagfish reproduction. Unlike the studies describe above, where fish were exposed over an extended period of time to a constant concentration, flagfish were exposed to sublethal concentrations of cyanide for 5-day pulses. Flagfish exposed to cyanide (65 ug/L) for 5 days following fertilization (i.e., as eggs) and then reared to maturity in clean water,

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spawned 25.6% fewer eggs than flagfish that had not been exposed. In another experiment by the same authors, flagfish that received a second 5-day pulse of cyanide as juveniles had an even greater reduction (39.3%) in the number of eggs spawned. These studies demonstrate that cyanide can affect an apical reproductive endpoint in fish.

The mechanism by which cyanide induces these reproductive effects is not fully understood. However, key physiological, biochemical, histological (morphological), and endocrine functions known to be involved in sexual maturation are affected by cyanide (Table 3).

Table 3. Chronic effects of cyanide on various fish species.

Species	Life stage	Exposure duration	Temp (C)	pH	HCN (ug/L)	CN (ug/L)	Effect	Reference
Atlantic Salmon <i>Salmo salar</i>	Adult females during late vitellogenesis	12 days	7	6.8	5	4.8	Inhibition of vitellogenin uptake by ovaries: observed increased plasma vitellogenin, decline in gonad vitellogenin, and no change in liver vitellogenin.	Ruby et al. 1987
Atlantic Salmon <i>Salmo salar</i>	Egg/fry	103-112 day incubation plus 58 days post hatch	3.5 – 8.3	7.6	10 – 100 (nominal)		Teratogenic effects observed in fry: malformation and/or absence of the eyes, defects in mouth and vertebral column and yolk-sac dropsy (note: CN concentrations not measured in exposure tanks)	Leduc 1977
Bluegill <i>Lepomis macrochirus</i>	Adult and early life stages	289 days ¹	24.9	8.1	5.2 - 80	5.4 – 82.7	No spawning occurred among fish exposed to CN except for one female in the 82.7 ug/l treatment. There were 5 and 8 spawnings in the two controls, respectively.	Kimball et al. 1978
Bluegill <i>Lepomis macrochirus</i>	Egg/larvae	57 days	25	8.02	4.8 – 82.1	4.9 – 84.4	NOEC for early life stage survival 9.4 ug CN/L. LOEC was 19.9 ug CN/L (88% reduction in survival compared to controls).	Kimball et al. 1978
Brook Trout <i>Salvelinus fontinalis</i>	19 month-old adults	144 days	12.5	7.9	5.7 – 75.3	5.6 – 74.4	NOEC for fecundity (number of eggs spawned) 5.6 ug CN/L	Koenst et al. 1977
Brook Trout <i>Salvelinus fontinalis</i>	Sac fry/juvenile	90 days (from hatch to 90 post hatch)	9	7.9	5.6 – 77.2	5.5 – 76	NOEC for growth (weight of juveniles at 90 days post hatch) 21.4 ug CN/L.	Koenst et al. 1977
Brook Trout <i>Salvelinus fontinalis</i>	Sac fry/juvenile	90 days (from hatch to 90 post hatch)	9	7.9	5.5 - 77	5.4 – 75.6	Survival reduced by 15%, 25% and 70%, at 55, 66 and 77 ug CN/L, respectively.	Koenst et al. 1977
Fathead Minnow <i>Pimephales promelas</i>	Larvae through adult	256 days	24.95	8.075	5.8 – 100.7	6.0 – 103.9	NOEC for fecundity (number of eggs spawned) 13.3 ug CN/L. LOEC 20.2 ug CN/L (58% reduction in spawning compared to controls).	Lind et al. 1977
Flagfish <i>Jordanella floridae</i>	Egg through adult	5 days during embryo/ larvae stage	25	8.05	65 – 87	66.8 – 89.5	Onset of spawning delayed, estrus cycle ² shortened, fecundity reduced in CN treatments 26% to 35% compared to controls	Cheng and Ruby 1981

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Species	Life stage	Exposure duration	Temp (C)	pH	HCN (ug/L)	CN (ug/L)	Effect	Reference
Flagfish <i>Jordanella floridae</i>	Egg through adult	5 days & 5 days ³	25	8.05	65 – 87	66.8 – 89.5	Onset of spawning delayed, estrus cycle ² shortened, fecundity reduced in CN treatments 39 to 47% compared to controls	Cheng and Ruby 1981
Flagfish <i>Jordanella floridae</i>	Egg/larvae	From fertilization through hatching (~5 – 9 days)	25	8.05	65 – 150	66.8 – 154.3	Hatching Success: 89% (control), 86%, - 3% in CN treatments. Hatching delayed and pituitary gland size reduced in all CN treatments Eye Malformations (microphthalmia - reduced eye size, and monophthalmia - disintegration of the eye), 30%, (66.8 ug/L), 40% (77 and 89.5 ug/L)	Cheng and Ruby 1981
Rainbow Trout <i>Oncorhynchus mykiss</i>	Juvenile males	18 days	12.5	7.9	10, 30	9.8, 29.5	Spermatogenesis: reduced number of dividing spermatogonia, 13% reduction at 10 ug/L, 50% reduction at 30 ug/L.	Ruby et al. 1979
Rainbow Trout <i>Oncorhynchus mykiss</i>	150 – 300 g vitellogenic females	12 days	12.5	7.2	10	9.7	Reduction in plasma vitellogenin and GSI compared to controls	Ruby et al. 1986
Rainbow Trout <i>Oncorhynchus mykiss</i>	2–3 year-old	12 days	12.5 ⁴ 10.7 ⁵	7.83 ⁴ 7.78 ⁵	10	9.8	Increase in brain dopamine compared to control fish, decrease in mean oocyte diameter (19%) of vitellogenic females, and higher numbers of spermatogonial cysts in males. Two identical experiments, one in July the other in August.	Szabo et al. 1991
Rainbow Trout <i>Oncorhynchus mykiss</i>	200 - 350g vitellogenic female rainbow trout	7 days	12	7.2	10, 20, 30	9.7, 19.3, 29	Decrease in serum calcium at 9.7 and 19.3 ug/L	Da Costa, H. and Ruby, S.M. 1984
Rainbow Trout <i>Oncorhynchus mykiss</i>	2.5 year old vitellogenic female rainbow trout	12 days	12	7.6	10	9.7	20% reduction in female GSI, 33% reduction in plasma vitellogenin, 55% reduction in plasma 17B-estradiol (E2), 48% reduction in oocyte diameter, and a 70% reduction in plasma T3 compared to controls. Plasma T4 levels were lower but difference was not statistically significant.	Ruby, S.M. et al. 1993a.
Rainbow Trout <i>Oncorhynchus mykiss</i>	juveniles (35 - 43g)	20 days	10	7.8	10, 20	9.8, 19.5	At 9.8: 65% reduction in the frequency of stage 5 (most mature) oocytes, 2 fold increase in the frequency of atretic follicles. At 19.5: 62% reduction in the frequency of stage 5 oocytes, 1.9 fold increase in the frequency of atretic follicles.	Lesniak, J.A. and Ruby, S.M. 1982.

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Species	Life stage	Exposure duration	Temp (C)	pH	HCN (ug/L)	CN (ug/L)	Effect	Reference
Rainbow Trout <i>Oncorhynchus mykiss</i>	2 year old sexually maturing males	12 days	11.5	7.9	10	9.8	Reduced # of spermatocytes, increased # of spermatogonial cysts, decreased # of basophils in pituitary, hypothesized effect of CN on hypothalamic-pituitary-gonadal axis	Ruby, S.M. et al. 1993b.
Rainbow Trout <i>Oncorhynchus mykiss</i>	Juveniles (3 g)	18 days	12.5	7.9	10	9.8	LOEC for growth measured as wet weight change over 18 days. NOEC < 9.8 ug CN/L	Dixon, D.G. and G. Leduc. 1981.
Rainbow Trout <i>Oncorhynchus mykiss</i>	Juveniles (12 g)	18 days	12.5	7.9	10	9.8	NOEC for growth measured as wet weight change over 18 days.	Dixon, D.G. and G. Leduc. 1981.
Rainbow Trout <i>Oncorhynchus mykiss</i>	Juveniles (3 g & 12 g)	18 days	12.5	7.9	10 – 30	9.8 – 29.5	Increased resting metabolic rate Liver damage: degenerative hepatic necrosis	Dixon, D.G. and G. Leduc. 1981.
Rainbow Trout <i>Oncorhynchus mykiss</i>	Juveniles (20g)	20 days	6 12 18	7.9	5 20 30	4.9 19.7 29.8	NOEC for mean specific growth rate (MSGR) based on dry weight. Author estimated thresholds at 6C, 12C and 18C to be < 4.9, 9.8 and 29.8 ug CN/L, respectively.	Kovacs, T.G. 1979.
Rainbow Trout <i>Oncorhynchus mykiss</i>	Juveniles (20g)	20 days	6,12, 18	7.9	5 – 45	4.9 – 44.7	Reduced swimming performance	Kovacs, T.G. 1979.
Sheepshead Minnow <i>Cyprinodon variegatus</i>	Embryo/larvae	28 days	22.4		29 - 462		Survival of 28 day-old larvae reduced from 22.5% (29 ug/L) to 54.5% (462 ug/L) compared to controls. Author states that MATC ⁶ lies between 29 and 45 ug /L	Schimmel et al. 1981

¹Exposure of first and second year spawners to HCN followed by 90-day exposure of eggs/larvae (second generation).

²Estrous cycle as defined by Cheng and Ruby 1981: “In this study, estrous cycle is defined as the duration of egg-laying starting from the first appearance of more than ten eggs through the occurrence of less than ten eggs on the mats.”

³ 5-day exposure during embryo/ larvae stage followed by a second 5 day exposure during juvenile stage.

⁴Experiment conducted in July.

⁵Experiment conducted in August.

⁶MATC – Maximum Acceptable Toxicant Concentration (the MATC is typically the geometric mean of the NOEC and LOEC).

Lesniak and Ruby (1982) reported abnormal oocyte development in sexually maturing female rainbow trout exposed to cyanide (10 and 20 ug HCN/L) for 20 days. Ovaries from cyanide-exposed fish contained fewer mature oocytes, exhibited altered patterns of secondary yolk deposition (in developing oocytes), had nearly twice the frequency of atresia (oocyte resorption), and had an overall reduction in the number of viable eggs.

Ruby et al. (1986) reported that vitellogenic female rainbow trout exposed for 12 days to 10 ug HCN/L had lower levels of plasma vitellogenin and a lower gonadosomatic index (GSI) compared to controls. In two similar studies, oocyte diameter (an indicator of gonadal growth and development) was reduced in sexually maturing female rainbow trout exposed for 12 days to 10 ug HCN/L (Ruby et al. 1993a, Szabo et al. 1991). Reduced oocyte diameter was accompanied by reductions in plasma vitellogenin, 17 β -estradiol (E2), and GSI (Ruby et al. 1993a), as well as increased whole brain dopamine levels (Szabo et al. 1991).

Dopamine has an inhibitory effect on gonadotropin-releasing hormone (GnRH) neurons in some fish species and it is GnRH that stimulates the release of gonadotropins (GtH I and GtH II) from the pituitary (Saligaut et al. 1999; Patino, R. 1997). GtH I and GtH II are believed to function similar to follicle-stimulating hormone and luteinizing hormone, respectively, in tetrapods (Patino, R. 1997). In female fish, GtH I acts on target cells in the gonad, stimulating E2 synthesis. E2 induces vitellogenin synthesis in the liver. Vitellogenin is the egg yolk precursor in fish that is produced by the liver, transported via blood, taken up by the ovaries, and incorporated into developing oocytes. GtH II also acts on the gonad by inducing the synthesis of maturation-inducing steroid (MIS). MIS induces oocyte maturational competence and ovulation (Park et al. 2007; Patino, R. 1997).

The control exerted by dopamine over gonadal maturation has been recognized by fish culturists, who have been successful in treating captive-reared fish with anti-dopaminergic drugs (which block dopamine receptors), such as pimozide and domperidone, to induce ovulation (Szabo et al. 2002, Park et al. 2007, Jensen 1993, Patino 1997). Thus, oocyte development, maturation and ovulation are under the control of gonadotropins and E2 which in turn, are modulated in part by GnRH and dopamine. This interaction between the neuroendocrine system and reproductive organs is referred to as the hypothalamus-pituitary-gonadal (HPG) axis (IPCS 2002).

Cyanide has also been shown to affect male reproductive processes. Exposure of male rainbow trout to cyanide concentrations of 10 and 30 ug HCN/L for 18 days disrupted spermatogenesis as evidenced by a reduction in the number of dividing spermatogonia and a blockage of mitotic progress (Ruby et al. 1979). Exposure of rainbow trout for 12 days to 10 ug HCN/L resulted in higher numbers of spermatogonial cysts in testes of male trout as well as higher levels of whole brain dopamine (Szabo et al. 1991). Ruby et al. (1993b) reported similar results where the number of spermatocytes decreased and the number of spermatocyte precursors (spermatogonial cysts) increased in two-year-old sexually maturing rainbow trout after a 12-day exposure to 10 ug HCN/L. There are indications that the transformation of spermatogonial cysts to spermatocytes is hormonally regulated through GtH along the HPG axis and that, within the pituitary, GtH is released from type I

granular basophils (Ruby et al. 1993b). Histological examination of pituitary glands from cyanide-exposed fish showed a reduction in the number of type I granular basophils. Ruby et al. (1993b) suggested that elevated levels of brain dopamine may be responsible for the selective loss of type I granular basophils and subsequent alteration of spermatocyte formation.

Ruby et al. (1979, 1993b) and Szabo et al. (1991) hypothesized that cyanide acts through the HPG axis to affect reproduction in fish. Their studies (described above) demonstrated (1) that cyanide caused an increase in brain dopamine levels, consistent with neuronal effects observed on mammals, (2) that levels of reproductive hormones (E2) and egg-yolk precursors (vitellogenin) were altered following exposure to cyanide, (3) the selective loss of putative GtH releasing pituitary cells (type I granular basophils) and (4) retarded gonad development in cyanide-exposed male and female rainbow trout. Taken together, these results appear to be consistent with HPG axis involvement. Ruby et al. (1979, 1993b) and Szabo et al. (1991) also reported that these effects occurred following relatively short (12 to 18 days), sublethal exposures to cyanide. Whether these effects would result in the same type of reduced fecundity and spawning, as was observed in cyanide-exposed female fathead minnows (Lind et al. 1977), bluegill (Kimball et al. 1978), and brook trout (Koenst et al. 1977), was not addressed in the rainbow trout studies because they were terminated before the fish reached full sexual maturity. However, it does seem likely that these effects would occur.

Results from Cheng and Ruby (1981) indicate that continuous exposure of fish to cyanide through the spawning period may not be necessary to affect fecundity. Short-term, pulsed exposures of cyanide to flagfish were sufficient to induce subsequent effects on the number of eggs spawned, and exposed fish did not appear to recover once the exposure had ceased. Even exposure of eggs, one of the most tolerant life stages in terms of acute toxicity (Smith et al. 1979), resulted in latent effects on fecundity once embryos hatched and survived to maturity. This finding is not unexpected given that it is during early developmental stages that the HPG endocrine axis is set up and feedback sensitivity of the hypothalamus and pituitary gonadotropes to gonadal steroids is established (IPCS 2002). Although Cheng and Ruby (1981) did not measure specific indicators of endocrine axis function, they did find that the pituitary gland of cyanide-exposed flagfish embryos was significantly smaller than the pituitaries from control fish. It appears that cyanide, like many EDCs (endocrine disrupting compounds, IPCS 2002), may affect the “set up” of the HPG axis and that these early developmental effects may have long-term consequences on reproduction.

Chronic exposure of eggs and larvae to cyanide can result in reduced embryo/larvae survival and altered development. Leduc (1978) exposed newly fertilized Atlantic salmon eggs to cyanide at concentrations of 10 to 100 ug HCN/L, and observed teratogenesis, as well as, delayed hatching and reduced hatching success at higher concentrations. There was a dose-dependent increase in the frequency of abnormal fry, ranging from 5.8% to 18.5%. Abnormalities included malformed and/or absence of eyes, defects in the mouth and vertebral column and yolk-sac dropsy (*Hydrocoele embryonalis*, also known as blue sac disease). Similar eye abnormalities were reported by Cheng and Ruby (1981) in flagfish larvae exposed, as eggs, to cyanide at concentrations of 65, 75, 87, and 150 ug

HCN/L. Egg hatching success was also reduced and time to hatch was delayed in all cyanide treatments. In a 28-day embryo/juvenile toxicity test, sheepshead minnow survival was significantly reduced in all treatments >29 ug HCN/L (Schimmel 1981). Schimmel (1981) noted there was considerable embryonic mortality and that there was no larval mortality during the last two weeks of exposure, indicating a greater sensitivity during early development. Kimball et al. (1978) exposed bluegill eggs and larvae to cyanide at concentrations of 4.8 to 82.1 ug/HCN/L, and reported that most deaths occurred within the first 30 days after hatching. Survival was reduced in all cyanide treatments and the effects were statistically significant at cyanide concentrations >9.1 ug HCN/L.

As previously mentioned, cyanide effects oxidative metabolism, energy production, and thyroid function; all are important for normal growth and performance. Therefore, it is not unexpected that sublethal exposure of fish to cyanide has been shown to impact growth, condition and swimming performance. There is also evidence that the effect of cyanide on these physiological endpoints can be modulated by other factors such as diet/ration and temperature. When cichlids (*Cichlasoma bimaculatum*) were fed unlimited rations and exposed to cyanide for 24 days, those fish exposed to lower concentrations of cyanide (< 0.06 ug HCN/L) were larger than controls, where as, at higher concentrations weight gain was depressed (Leduc 1984). The increased weight gain in the low-dose treatments was attributed to higher food consumption, which was allowed to occur because ration was not restricted. Low-dose stimulation is a common effect across a broad range of chemical and non-chemical stressors (Calabrese 2008).

Dixon and Leduc (1981) held juvenile rainbow trout on restricted rations and exposed them to cyanide at concentrations of 10, 20, and 30 ug HCN/L for 18 days and observed significantly reduced weight gain in all treatments compared to controls. The effect was characterized by an initial decrease in specific growth during the first 9 days followed by a significant increase from day 9 through day 18. The growth surge during the latter half of the exposure period was not sufficient to offset early reductions. Cyanide-affected juvenile rainbow trout were in poorer condition, as indicated by lower fat content, and had higher respiration rates for several days post-exposure. In addition, juvenile rainbow trout in all cyanide treatments exhibited degenerative necrosis of hepatocytes (i.e., liver tissue damage) that increased in severity with the level cyanide exposure.

Kovacs (1979) held juvenile rainbow trout on restricted rations and exposed them to cyanide for 20 days. The results were similar to those reported by Dixon and Leduc (1981). Cyanide reduced the mean specific growth rate (MSGR) and affected-fish gained less fat during the exposure period. Kovacs (1979) also examined the effects of temperature on rainbow trout growth and sensitivity to cyanide. He found that the growth rate of rainbow trout was inversely related to holding temperature (6, 12 and 18°C), as would be expected, and that trout held at colder temperatures were more sensitive to cyanide. The NOECs for MSGR were 5, 20, and 30 ug HCN/L for trout held at 6, 12, and 18°C, respectively. Based on the exposure response curves, Kovacs (1979) estimated thresholds for effects on rainbow trout growth to be <5 ug HCN/L at 6°C, 10 ug HCN/L at 12°C, and 30 ug HCN/L at 30°C.

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Kovacs (1979) also evaluated the effects of cyanide on fish swimming performance. His results indicated that swimming performance was affected by cyanide and that the effect was also temperature-sensitive. Fish from the growth study described above were placed in swimming chambers and tested for swimming stamina. Among non-exposed trout, swimming stamina, measured as distance travelled (in meters), decreased with decreasing temperature (i.e., fish held at 6°C travelled a shorter distance than fish held at 18°C). Cyanide-exposed fish had reduced swimming stamina compared to non-exposed fish and the effect was more severe at colder temperatures. Based on the exposure-response regression equations reported by Kovacs (1979), the predicted reduction in swimming stamina (compared to controls) for fish exposed to cyanide at the chronic water quality criterion (5.2 ug CN/L) would be 52% at 6°C, 20% at 12°C, and 3% at 18°C.

Cyanide has been shown to affect the swimming performance of other salmonid species, as well. Leduc (1984) calculated the cyanide concentration causing a 50% reduction in swimming ability based on original data collected for brook trout (Neil 1957) and coho salmon (*Oncorhynchus kisutch*) (Broderius 1970) to be 5 ug HCN/L and 7 ug HCN/L, respectively. Thus, chronic exposure of fish to cyanide at sublethal concentrations can affect growth, body condition, and swimming performance. Factors such as temperature and diet/ration can modulate cyanide toxicity, and, for some species-endpoint combinations, these effects may occur at or below the chronic cyanide criterion.

Effects of Pollutant Mixtures

Relatively few studies have been performed to measure the effects of free cyanide in combination with other contaminants. Concurrent exposure to cyanide and ammonia produced greater than additive effects to acute lethality in rainbow trout, salmon, and chub (Smith et al. 1979, Alabaster et al. 1983, and Douderoff 1976), and to chronic sublethal effects to growth in rainbow trout (Smith et al 1979). In rainbow trout and salmon, effects to acute lethality were 1.2 and 1.63 times greater, respectively, than would be expected by additivity. Concurrent exposure to cyanide and zinc also resulted in synergistic effects to acute lethality in fathead minnows, where toxicity was 1.4 times that predicted by additivity (Smith et al 1979). Although we are unable to quantify the effect of these synergistic mechanisms for this analysis, they should be considered when assessing effects of cyanide to aquatic organisms in waterways with elevated concentrations of ammonia and zinc.

Acute Effects Estimation for Listed Fish Evaluation Species

Of the 103 listed fish species that were evaluated for acute cyanide effects, only the 4 most sensitive (i.e. fountain darter, Apache trout, Lahontan cutthroat trout, and bull trout) were considered likely to be adversely affected (Appendix B). For these 4 species, the magnitude of effect resulting from acute exposure to cyanide at the CMC (22.4 ug CN/L) was estimated using regression analysis.

Estimated acute effects for Fountain Darter: Of the acute exposure-response regression equations listed in Appendix G there are six equations for juvenile mortality in a species of

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fish, Yellow Perch, from the family Percidae (Smith et al. 1978). Because toxicity is inversely related to water temperature (Eisler 2000), the equation for the lowest tested temperature, 15 C, would be the most appropriate equation for estimating acute effects in the Fountain Darter. That equation of:

$$\text{Probit (\% juv. Mortality)} = -17.790 + 11.650 (\log (\text{ug HCN /L}))$$

yields an estimated Yellow Perch acute LC₅₀ of 90.4 ug HCN /L, or 88.9 ug CN /L (at the test pH of 7.82). The estimated acute LC₅₀ for Fountain Darter from the ICE lower 95% confidence value is 21.5 ug CN/L (Appendix B). This yields an acute SSEC_x estimate of 92.6 ug CN/L, or 94.2 ug HCN /L (see chronic effects section and Appendix C for explanation of SSEC_xs). Entering that SSEC_x value on an HCN basis into our probit-log regression equation yields an estimated effect level for Fountain Darter of 58.2% juvenile mortality at the CMC criterion.

Estimated acute effects for Apache Trout: Of the acute exposure-response regression equations listed in Appendix G there is only one for an *Oncorhynchus* species of salmonid (Broderius and Smith 1979). That equation is for tests with juvenile Rainbow Trout. That equation of:

$$\text{Probit (\% juv. Mortality)} = 33.63 + 23.04 (\log (\text{mg CN /L}))$$

yields an estimated Rainbow Trout acute LC₅₀ of 57.2 ug CN /L. The estimated acute LC₅₀ for Apache Trout from the ICE lower 95% confidence value is 16.5 ug CN/L (Appendix B). This yields an acute SSEC_x estimate of 77.7 ug CN/L (see chronic effects section Appendix C for explanation of SSEC_xs). Entering that SSEC_x value into our probit-log regression equation yields an estimated effect level for Apache Trout of >99.9% juvenile mortality at the CMC criterion.

Estimated acute effects for Lahontan Cutthroat Trout: Of the acute exposure-response regression equations listed in Appendix G there is only one for an *Oncorhynchus* species of salmonid (Broderius and Smith 1979). That equation is for tests with juvenile Rainbow Trout. That equation of:

$$\text{Probit (\% juv. Mortality)} = 33.63 + 23.04 (\log (\text{mg CN /L}))$$

yields an estimated Rainbow Trout acute LC₅₀ of 57.2 ug CN /L. The estimated acute LC₅₀ for Lahontan Cutthroat Trout from the ICE lower 95% confidence value is 22.8 ug CN/L (Appendix B). This yields an acute SSEC_x estimate of 56.2 ug CN/L (see chronic effects section and Appendix C for explanation of SSEC_xs). Entering that SSEC_x value into our probit-log regression equation yields an estimated effect level for Lahontan Cutthroat Trout of 43% juvenile mortality at the CMC criterion.

Estimated acute effects for Bull Trout: Of the acute exposure-response regression equations listed in Appendix G there are nine equations for juvenile mortality in a species of fish, Brook Trout, from the genus *Salvelinus* (Smith et al. 1978). Because toxicity is

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inversely related to water temperature (Eisler 2000), the equation for the lowest tested temperature, 4 C, would be the most appropriate equation for estimating acute effects in the Bull Trout. That equation of:

$$\text{Probit (\% juv. Mortality)} = -28.849 + 19.626 (\log (\text{ug HCN /L}))$$

yields an estimated Brook Trout acute LC₅₀ of 53.1 ug HCN /L, or 51.2 ug CN /L (at the test pH of 7.19). The estimated acute LC₅₀ for Bull Trout from the ICE lower 95% confidence value is 15.7 ug CN/L (Appendix B). This yields an acute SSEC_x estimate of 73.0 ug CN/L, or 75.6 ug HCN /L (see chronic effects section and Appendix C for explanation of SSEC_xs). Entering that SSEC_x value on an HCN basis into our probit-log regression equation yields an estimated effect level for Bull Trout of 99.9% juvenile mortality at the CMC criterion.

Citing DeForest et al. (*in prep.*), Gensemer et al. (2007) noted that exposure-response curves for cyanide acute toxicity in fish are “quite steep”. Gensemer et al.’s (2007) observation is supported by the low LC₅₀/LC₁₀ ratios derived for this biological opinion from the exposure-response regression equations in Smith et al. (1978) and Broderius and Smith (1979). A combination of the steep response curves and the low estimated LC₅₀ values for the listed evaluation species relative to the surrogate toxicity test species results in substantive estimates of adverse effects at the acute (CMC) criterion of 22.4 ug CN /L even though the acute effects assessment EC_as (13.77-20.00) were not far below the CMC (22.4).

Chronic Effects Estimation for Listed Fish Evaluation Species

Ideally, concentration (dose)-response data suitable for predictive modeling would be available for sensitive chronic endpoints for each of the listed species evaluated in this analysis (hereafter referred to as “listed evaluation species”). Such data do not exist for cyanide for any of our listed evaluation species. As recently reviewed by Gensemer et al. (2007), the current inventory of concentration-response data from chronic toxicity testing with cyanide consists of four datasets: one each for reproductive endpoints among the fathead minnow (*Pimephales promelas*; Lind et al. 1977) and the brook trout (Koenst et al. 1977); one for juvenile survivorship among bluegill (Kimball et al. 1978); and one for the sheepshead minnow (Schimmel et al. 1981). Upon closer inspection, Gensemer et al. (2007) found the dataset for the sheepshead minnow to be insufficient for meaningful predictive modeling and we agree with that conclusion. Thus, we are left with three datasets as the best available scientific basis for estimating toxic effects (or the lack thereof) at the chronic criterion value of 5.2 ug CN/L. In addition to our three useable concentration-response datasets, we also possess estimates of LC₅₀ values for our listed evaluation species as per the procedures described in Appendix B.

Based on the above information, we took the following approach to evaluating the effects of the proposed action on the listed fish evaluation species:

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(1) The three concentration-response data sets were transformed into the most precise, predictive concentration-response models that the data can support; these models were used to predict the response of chronic toxicity test species to proposed chronic CN exposure levels.

(2) The predicted response of a listed fish evaluation species to chronic CN exposures was considered to be the same as the response observed for a chronic toxicity test species at an adjusted chronic CN exposure level based on the ratio of their respective LC₅₀ values (see example below).

Two assumptions were relied upon to predict the effect of proposed chronic CN exposure levels on the listed fish evaluation species:

- (1) The relative differences in sensitivity to chronic CN exposures between the listed evaluation species and the chronic toxicity test species (fathead minnow, brook trout, and bluegill) are approximated by the ratio of their respective LC₅₀ values; and
- (2) The slopes of the concentration-response curves are also approximately comparable between the listed evaluation species and the chronic toxicity test species.

These assumptions create a clearly defined basis for a default hypothesis that allows for an analysis of the effects of the proposed action on listed fish evaluation species to proceed within the constraints of minimal data until such time as more data become available. As more data become available appropriate modification (or validation) of our default approach will be facilitated.

An example of applying the above methodology: suppose that one of our chronic toxicity test species is predicted to exhibit a 20% adverse effect from being exposed to a concentration of 5.2 ug CN/L. If a listed evaluation species happens to have an estimated LC₅₀ value equal to that of the chronic toxicity test species, then a 20% adverse effect would also be predicted for the listed evaluation species. If the ratio of LC₅₀ values was 1.5 (rather than 1.0) in the direction of greater sensitivity for the listed evaluation species than the chronic toxicity test species, then the predicted response at our concentration of interest of 5.2 ug/L for our listed evaluation species would be the same as the response observed for the chronic toxicity test species at a CN concentration 1.5 times 5.2 ug/L, which equals 7.8 ug/L. We refer to such predicted response values as surrogate currency equivalents (or SSEC_x or SS_x values) for our listed evaluation species. In this example, the predicted adverse effect for our chronic toxicity test species at the SSEC_x of 7.8 ug/L would be our surrogate currency predicted effect for the listed evaluation species at 5.2 ug CN/L (for one of three prediction models) for the purposes of this Biological Opinion. A more detailed derivation and explanation of the SSEC_x/ SS_x concept is provided in Appendix C.

Because groups of taxonomically related listed evaluation species were assigned identical LC₅₀ values from the same ICE or SSD model, there are only 17 SSEC_x values that need to be evaluated for any given (chronic toxicity test species) prediction model, but they are

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different for each prediction model (3 models x 17 values = 51 total SSEC_x values of interest).

For the prediction model based on fathead minnow chronic toxicity data, the SSEC_x values range from 6.7 to 45.8 ug CN/L (Table 4). As indicated by the entire range of SSEC_x values being greater than 5.2 ug CN/L, all listed evaluation species have LC₅₀ values that are more sensitive to cyanide than the fathead minnow LC₅₀ value.

For the prediction model based on brook trout chronic toxicity data, the SSEC_x values range from 4.2 to 28.4 ug CN/L (Table 4).

For the prediction model based on bluegill chronic toxicity data, the SSEC_x values range from 6.1 to 41.7 ug CN/L (Table 4).

The SSEC_x ranges indicated above define for each prediction model the range of cyanide concentrations over which model fit will be of most relevance to the effects of the proposed action on the listed evaluation species considered in this Biological Opinion. Detailed SSEC_x-related results and the origins of the LC₅₀ values used to calculate the SSEC_x values are presented in Table 4 and Appendix D.

Table 4. Surrogate currency equivalents (SSEC_x) for each LC50 surrogate taxon/chronic toxicity test species combination. SSEC_x values were calculated using equation 5 in Appendix C. Surrogate taxa were used to estimate LC50 values for listed evaluation species except when measured values for the listed species were available (i.e. *Salmo salar*).

			Effects on Fecundity		Effects on Early Life Stage Survival
			Fathead Minnow SS LC ₅₀ =138.4 (ug CN/L)	Brook Trout SS LC ₅₀ =85.7 (ug CN/L)	Bluegill SS LC ₅₀ =126.1 (ug CN/L)
Surrogate taxa used to estimate listed species (LS) LC ₅₀	LSEC _x (ug CN/L)	LS LC ₅₀ (ug CN/L)	SSEC _x (ug CN/L)	SSEC _x (ug CN/L)	SSEC _x (ug CN/L)
Actinopterygii (class)	5.2	66.5 ¹	10.8	6.7	9.9
Cypriniformes (order)	5.2	84.55 ¹	8.5	5.3	7.8
Family Catostomidae					
<i>Xyrauchen texanus</i> (species)	5.2	83.8 ²	8.6	5.3	7.8
Cyprinidae (family)	5.2	101.7 ²	7.1	4.4	6.4
<i>Cyprinella monacha</i> (species)	5.2	36.81 ²	19.6	12.1	17.8
<i>Gila elegans</i> (species)	5.2	50.9 ²	14.1	8.8	12.9
<i>Notropis mekistocholas</i> (species)	5.2	48.5 ²	14.8	9.2	13.5
<i>Ptychocheilus lucius</i> (species)	5.2	43.5 ²	16.6	10.3	15.1
Perciformes (order)	5.2	90.8 ¹	7.9	4.9	7.2
Percidae (family)	5.2	42.3 ²	17.0	10.5	15.5
<i>Etheostoma</i> (genus)	5.2	40.0 ²	18.0	11.1	16.4
<i>Etheostoma fonticola</i> (species)	5.2	21.5 ²	33.4	20.7	30.5
Salmoniformes, Salmonidae					

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<i>Oncorhynchus</i> (genus)	5.2	47.0 ²	15.3	9.5	13.9
<i>O. apache</i> (species)	5.2	16.5 ²	43.6	27.0	39.7
<i>O. clarki henshawi</i> (species)	5.2	22.8 ²	31.5	19.5	28.7
<i>Salmo salar</i> (species)	5.2	90 ³	8	5	7.3
<i>Salvelinus</i> (genus)	5.2	15.7 ²	45.8	28.4	41.7

¹ LC₅₀ based on 5th percentile estimate from species sensitivity distribution (SSD), Table 2 – Cyanide BE.

² LC₅₀ estimate based on lower bound of the 95% CI from ICE model (Appendix D).

³ LC₅₀ based on measured value from the Cyanide BE (Table 1).

Prediction Models

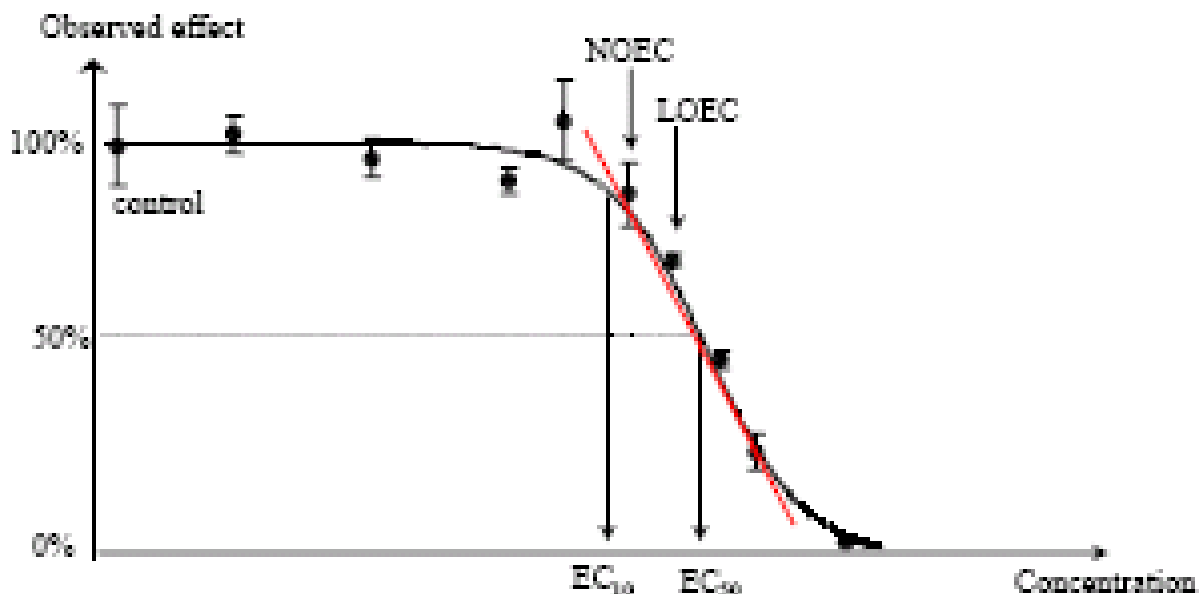
Statistical regression techniques were used to model or “fit” the relationship between cyanide concentrations and toxic effects to listed evaluation species based on data for the chronic toxicity test species. For nuances of statistical regression specific to toxicological applications, we relied substantively on two recent technical guidance documents: (1) Environment Canada (2005: “Guidance Document on Statistical Methods for Environmental Toxicity Tests”); and (2) OECD (2006: “Current Approaches in the Statistical Analysis of Ecotoxicity Data: A Guidance to Application”). We also reviewed other relevant guidance such as that provided in the documentation for EPA’s Toxicity Relationship Analysis Program (TRAP) (EPA 2002) and in discipline-specific statistical textbooks such as Gad and Weil (1988) and Sparks (2000).

As noted by Environment Canada (2005), an important principle of regression techniques is to keep the model simple, if that can reasonably be done. In completing this analysis, we have further incentive to follow that principle because we have a strong interest in evaluating the uncertainty (confidence) associated with point estimates and therefore an interest in avoiding what Environment Canada (2005) noted as the “...obstacle of calculating confidence intervals around nonlinear regression estimates...” Throughout this analysis we have been mindful of that because our models are not based on biological or chemical mechanisms of action, but are purely statistical constructs that have no mechanistic meaning. A statistical concentration-response model only serves to smooth the observed concentration-response to estimate effect concentrations by interpolating between treatment concentrations, and to provide a tool for assessing confidence intervals. Therefore, the choice of model is to some extent arbitrary (OECD 2006). That being noted, we constructed models that conformed to the non-arbitrary characteristics of the data we are working with and with statistical standard practices (such as data transformations). The degree of model fit achieved is an artifact of those specific decisions not the result of *post hoc* “model shopping” (EPA 2002).

Generic Concentration-response Relationship

Figure 1 illustrates a generic concentration-response relationship that typically takes on a sigmoidal form due to threshold effects on the low concentration end of the x-axis and to asymptotic effects at the high concentration end of the x-axis.

Figure 2. Generalized concentration-response relationship adapted from OECD (2006:Figure 3.2). Note that the illustrated curve is a plot fitted to a real dataset, thus the identification of NOEC and LOEC concentrations. For the purposes of this figure, consider the y-axis as a positive attribute that becomes diminished by toxicity, such as percent survivorship.



Note that the superimposed straight line in Figure 2 represents the region of concentrations that induce an intermediate toxic response that are well approximated by a linear fit. This “linear region” is strongest within one probit (also known as “normal equivalent deviate”) either side of the median response concentration (EC_{50}), or roughly for concentrations that induce 16 to 84% response (Environment Canada 2005). The narrow ranges of $SSEC_x$ values that we need to evaluate can be expected to overwhelmingly fall within those boundaries as a result of the methods EPA used to set the chronic criterion at 5.2 $\mu\text{g CN/L}$; see the next section titled “Derivation of the Criterion Continuous Concentration (CCC)”. Our approach is conceptually similar to the TRAP program’s Piecewise Linear regression option (EPA 2002). Even with regard to the nonlinear regression options in TRAP, EPA (2002) provides a recommendation for segmented analysis when there is a focal region (or subset) of test concentrations of particular concern:

Within the limitations of this program, one useful approach can be to exclude (censor) high effects data from the analysis if (a) only low levels of effect are of interest and (b) there are sufficient low-to-moderate effects data to support a good analysis.

Prediction Model based on the Fathead Minnow Dataset

Lind et al. (1977) examined fathead minnow fecundity (number of eggs per spawn) and egg hatchability in relation to a series of cyanide treatments (concentrations). The experimental structure, as well as the fecundity results, is summarized in Table 5. There

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were five control replicates, and two replicates each for ten exposure concentrations. The response data are reasonably monotonic, especially within the intermediate response range covered by the lowest six treatments. Those treatments ranged (on a free cyanide basis) from 6 to 45.6 ug/L CN; a span that closely corresponds to the SSEC_x range we want to evaluate (Table 4).

Table 5. Egg production of adult fathead minnows exposed for 256 days (from larvae through adult) to various concentrations of cyanide (from Lind et al. 1977; Table II).

Treatment HCN (ug/L)	Mean HCN (ug/L)	Free cyanide as CN (ug/L)	Mean eggs per female	Mean eggs per female per treatment	Reduction in the number of eggs per female - percent of control
Control			2530	3476	
Control			4483		
Control			3990		
Control			2718		
Control			3660		
5.7	5.8	6.0	1886	2512	27.7%
5.9			3138		
13.0	12.9	13.3 ^N	1701	1845	46.9%
12.7			1989		
19.6	19.6	20.2 ^L	1694	1468	57.8%
19.6			1241		
27.1	27.3	28.2	1093	1367	60.7%
27.5			1640		
36.0	35.8	36.9	678	1010	71.0%
35.6			1341		
43.7	44.2	45.6	2054	1124	67.7%
44.7			194		
62.5	63.5	65.6	74	72	97.9%
64.5			70		
73.1	72.8	75.1	573	319	90.8%
72.4			64		
81.5	80.7	83.3	266	243	93.0%
79.8			219		
96.1	100.8	103.9	0	0	100.0%
105.4			0		

^N NOEC

^L LOEC

To “build” our prediction model we transformed both the concentration data and the fecundity data for *a priori* reasons. We log-transformed the concentration data for two

reasons: (1) statistically, toxicological tolerance distributions have long been confirmed as log-normal (OECD 2006); and (2) biologically, organisms experience toxicants on a log scale. Toxicological custom is to use log base-10 for the log transformations of test concentrations (Environment Canada 2005). Count data, such as “number of eggs per spawn” typically conform to a Poisson distribution rather than a normal distribution. To normalize such data for regression analysis a square-root transformation is recommended (EPA 2002). We used the square-root transformed response data for statistical analysis and then back-transformed the data for reporting results. This transformation does not change the model, but affects what the best parameter estimates and confidence limits are (EPA 2002). Thus, our model of choice is a log-square root linear regression over our focal segment (subset) of test concentrations.

In agreement with Gensemer et al.’s (2007) treatment of the same dataset, we collapsed the fecundity and egg hatchability endpoints into a single endpoint: “eggs hatched per spawn” which is the product of (eggs per spawn) x (egg hatchability) at each treatment concentration. We went a step further than Gensemer et al. (2007) and additionally applied a data-smoothing procedure to meet the assumption of monotonicity of response inherent in a linear regression. We did that by calculating three-point moving averages for both the fecundity and hatchability endpoints. This is a standard statistical technique for separating the “signal” from the “noise” in epidemiological and earth sciences (e.g., Rothman et al. 2008, Borradaile 2003).

TABLE 6. Fathead minnow input data for effects modeling.

Treatment (free ug CN/L)	Pooled mean eggs/female	Pooled Proportion Hatch ^a	Unsmoothed Pooled mean hatch/female ^b	3-pt moving average of proportion hatch	Smoothed Pooled mean hatch/female ^b	SQRT transform
Control Mean	3476	0.842	2927	0.763 ^c	2652	51.5
6.00	2512	0.606	1522	0.754	1894	43.52
13.30	1845	0.813	1500	0.682	1258	35.47
20.20	1468	0.626	919	0.612	898	29.97
28.20	1367	0.396	541	0.527	720	26.83
36.90	1010	0.559	565	0.354	358	18.92
45.60	1124	0.108	121	0.271	305	17.46
65.60	72	0.147	11	0.149	11	3.31
75.10	319	0.192	61	0.181	58	7.62
83.30	243	0.204	50	0.132	32	5.66
103.90	0	0	0	0.068 ^c	0	0

^aMeans weighted by replicate sample sizes; excludes hatchability result for Control B as per recommendation by Lind et al. (1977:264-265).

^bRounded to the nearest whole number.

^cBased on double-weighted observed value; assuming any doses to the left of 0% response will be constant and any points to the right of 100% response will be constant.

^dFinal effects model based upon the shaded subset of data.

Although we didn’t use the control data in our focal segment linear regression, we estimated where the smoothed data would cross the y-axis by double-weighting the control

value, which then (along with its nearest neighboring data point) provided the basis of a three-point moving average for the “endpoint” of the concentration series. This double-weighting is justified conceptually because a treatment to the left of the controls on the concentration axis would be expected to respond the same as the controls (Environment Canada 2005). This enabled us to avoid comparing point estimates of eggs hatched per spawn from models fitted to smoothed data with “unsmoothed” control reference points. Note that our “smoothed” estimate of a control reference point was obtained using the actual data nearest to the y-axis and is not extrapolated from our estimated regression equation. Also note that we did not control-adjust the results prior to model fitting, a practice that leads to serious upward bias in EC_x point estimates (Environment Canada 2005, OECD 2006). A summary of response data smoothing and transformation is presented in Table 6.

The resulting log-square root focal segment linear regression model shows a very close fit to the data with an adjusted r-square of 0.964. The regression equation is:

$$\text{Square-root (hatched eggs per spawn)} = -30.19(\text{LOG CN}) + 68.36$$

The regression plot (Figure 3) and summary regression statistics (Table 7) are presented below. The regression was conducted using the multiple linear regression module of the *Statistica* software package (StatSoft 2006). Because we are dealing with small samples (six points in this case), we report the adjusted r-squared value which adjusts for the limited degrees of freedom in the model (StatSoft 2006).

Figure 3. Log- square root focal segment regression plot for fathead minnow fecundity x hatchability (= eggs hatched per spawn).

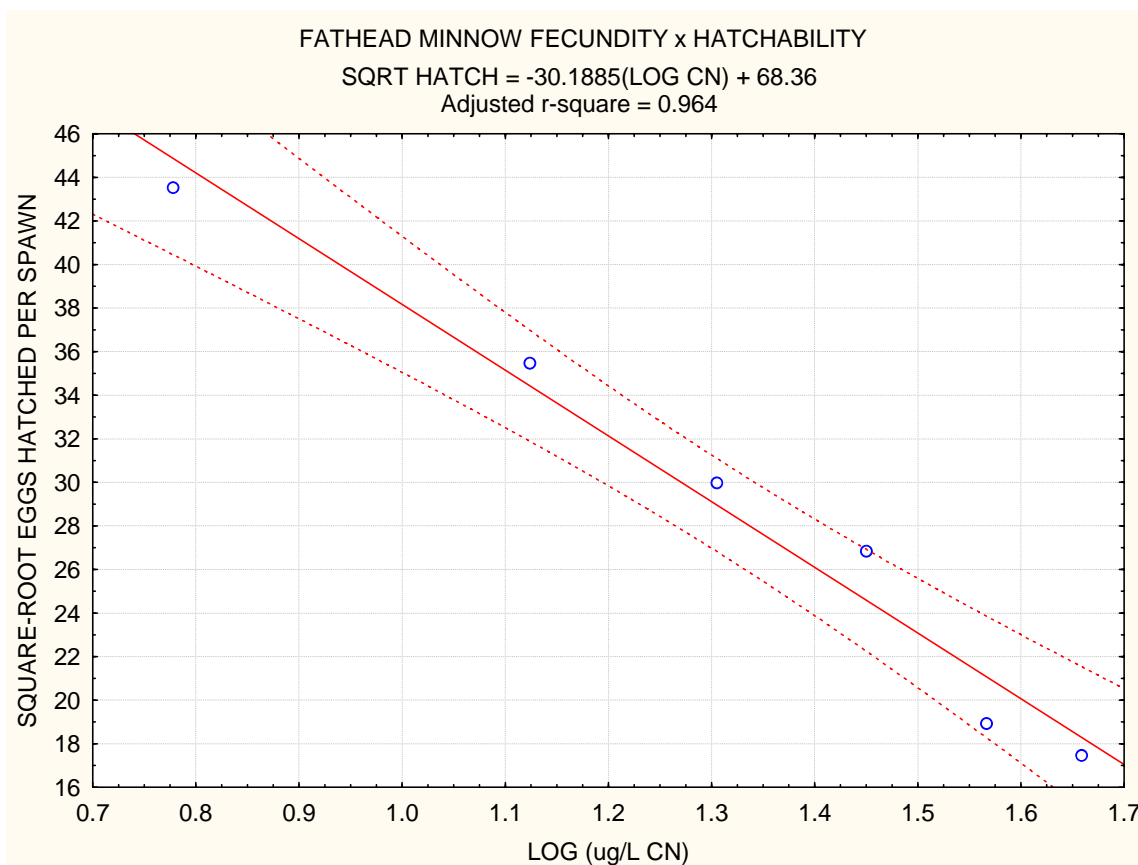


TABLE 7. Summary regression statistics.

Effects Surrogate	N	F value	p-level	Intercept	Std Err	p-level	Slope	Std Err	p-level
Fathead Minnow	6	134.6	<0.00032	68.36	3.505	0.000041	-30.19	2.602	0.00032
Brook Trout	5	12.34	<.039	24.85	2.595	0.0024	-6.594	1.877	0.039
Bluegill	5	11.75	<0.042	0.3514	0.9277	0.73	-2.533	0.7919	0.042

Prediction Model based on the Brook Trout Dataset

Koenst et al. (1977) examined brook trout fecundity (number of eggs per spawn) and egg viability in relation to a series of cyanide treatments (concentrations). The experimental structure, as well as the fecundity results, is summarized below (Table 8). There were two control replicates, and seven cyanide treatments. The lowest five treatments produced intermediate effects responses and covered a range of concentrations from 5.6 to 53.2 ug/L

CN; a span that closely corresponds to the SSEC_x range we want to evaluate (Table 4). There was substantive variability in the results for the two control replicates. This led to Koenst et al. (1977) excluding control replicate B, but noting that additional testing might indicate that the control results should be averaged. As noted in the footnote to Table 8, subsequent studies with brook trout (Holcombe et al. 2000) have confirmed that control replicate B should be averaged with control replicate A. For that reason, we used the control mean as our reference point for evaluating model predictions.

Table 8. Egg production of adult brook trout exposed to HCN for 144 days prior to the start of spawning (from Koenst et al., 1977).

HCN (ug/L)	Free cyanide as CN (ug/L)	Mean eggs spawned per female	Reduction in the number of eggs per female - percent of control*
Control A		502	
Control B		744	
Control Mean		623	
5.7	5.6	513	17.7%
11.2	11.1	291	53.3%
32.3	31.9	246	60.5%
43.6	43.1	442	29.1%
53.9	53.2	262	57.9%
64.9	64.1	124	80.1%
75.3	74.4	0	100.0%

* Reductions in the number of eggs spawned relative to controls were calculated using the Control mean (623 eggs per female). Koenst et al. (1977) performed the same calculation using only Control A (502 eggs per female) and reported that the MATC (Maximum Acceptable Toxicant Concentration) lies between 5.7 and 11.2 ug HCN/L. However, the authors went on to say that “When compared to the mean of the two controls, 5.7 ug/L HCN would appear to show a substantial reduction in eggs spawned per female, but due to the high variability in spawning in the two controls, further study would be required to reach this conclusion.” Since that time other studies with brook trout have been conducted (Holcombe et al. 2000). The mean number of eggs spawned per female observed by Koenst et al. (1977) is within the range reported for these other studies, which supports the use of data from both controls in estimating the effect of cyanide on brook trout fecundity.

Again, in agreement with Gensemer et al.’s (2007) treatment of the same dataset, we collapsed the fecundity and egg viability endpoints into a single endpoint: “viable eggs per spawn” which is the product of (eggs per spawn) x (egg viability) at each treatment concentration. In the five-point segment of the data that we focus on, there was a substantive deviation from monotonicity at the 43.1 ug/L CN concentration. Therefore, once again we employed data-smoothing with a 3-point moving average to restore a monotonic progression of responses. Because the endpoint here is virtually the same as the endpoint for the fathead minnow dataset, other aspects of our treatment of the data for “building” a prediction model are the same as previously presented for the fathead minnow model. A summary of response data-smoothing and transformation is presented in Table 9 below.

TABLE 9. Brook trout input data for effects modeling.

Treatment (free CN ug/L)	Mean eggs/female	3-pt moving average of mean eggs/spawn	Proportion Viable	3-pt moving average of proportion viable	Smoothed mean viable/female ^a	SQRT transform
Control Mean	623	586 ^b	0.935	0.923 ^b	541	23.26
5.60	513	476	0.899	0.872	415	20.37
11.10	291	350	0.781	0.803	281	16.76
31.90	246	326	0.729	0.792	258	16.06
43.10	442	317	0.866	0.745	236	15.36
53.20	262	276	0.641	0.502	139	11.79
64.10	124	129	0	0.214	28	5.29
74.40	0	41 ^b	0	0 ^b	0	0

^aRounded to the nearest whole number.

^bBased on double-weighted observed value; assuming any doses to the left of 0% response will be constant and any points to the right of 100% response will be constant.

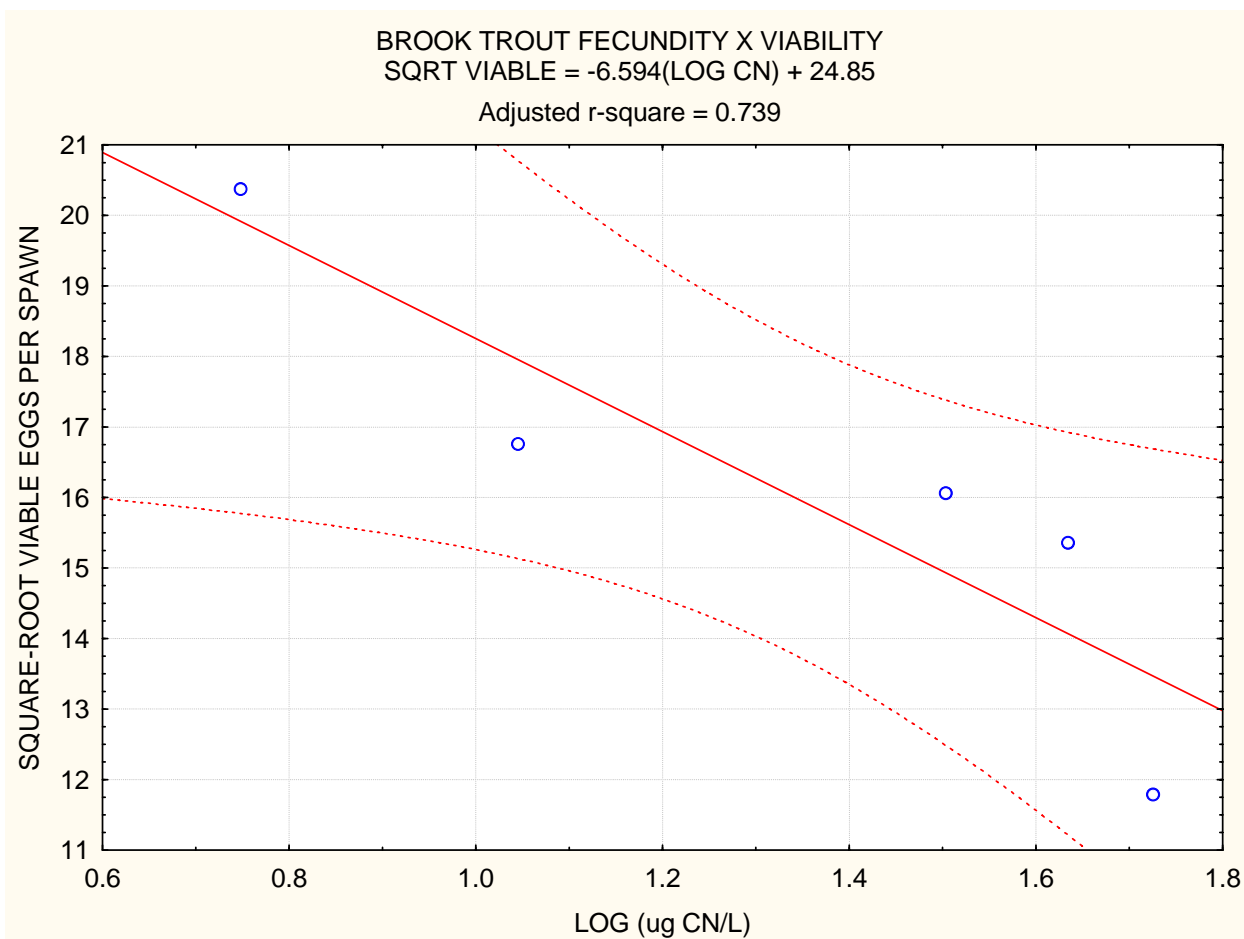
^cFinal effects model based upon the shaded subset of data.

The resulting log-square root focal segment linear regression model does not show as strong a fit to the data as the fathead minnow model does, but still shows a reasonably good fit with an adjusted r-square of 0.739. The regression equation is:

$$\text{Square-root (viable eggs per spawn)} = -6.594(\text{LOG CN}) + 24.85$$

The regression plot is presented in Figure 4 and summary regression statistics are presented in Table 7. The regression was conducted using the multiple linear regression module of the *Statistica* software package (StatSoft 2006). Because we are dealing with small samples, i.e., five points in this case, we report the adjusted r-squared value which adjusts for the limited degrees of freedom in the model (StatSoft 2006).

Figure 4. Log-square root focal segment regression plot for brook trout fecundity x viability (= viable eggs per spawn).



Prediction Model based on the Bluegill Dataset

Kimball et al. (1978) examined bluegill juvenile survivorship in relation to a series of cyanide treatments (concentrations). The experimental structure, as well as the survivorship results, is summarized in Table 10. There were four control replicates, and two replicates each for eight cyanide treatments. The lowest five treatments produced intermediate effects responses and covered a range of concentrations from 4.9 to 40.6 ug/L CN; a span that closely corresponds to the SSEC_x range we want to evaluate (Table 4).

Table 10. Survival of bluegills from fertilized egg to the 57-day juvenile state in various HCN concentrations (from Kimball et al. 1978; Table 3).

HCN (ug/L)	Mean HCN (ug/L)	Free cyanide as CN (ug/L)	Percent survival	Number of surviving juveniles *	Mean percent survival	Reduction in survival compared to controls
Control			37.5	75	23.3	
Control			20.0	40		
Control			10.0	20		
Control			25.5	51		
4.8	4.8	4.9	18.5	37	18.5	20.6%
5.2			lost			
8.9	9.1	9.4 ^N	25.0	50	16.3	30.0%
9.2			7.5	15		
19.2	19.4	19.9 ^L	3.0	6	2.8	88.0%
19.6			2.5	5		
28.5	29.1	29.9	2.5	5	2.5	89.3%
29.7			2.5	5		
38.7	39.5	40.6	3.0	6	3.8	83.7%
40.2			4.5	9		
49.3	49.3	50.7	13.5	27	13.5	42.1%
51.9			lost			
61.8	62.9	64.6	0.0	0	0.0	100.0%
64			0.0	0		
80.4	82.1	84.4	0.0	0	0.0	100.0%
83.8			0.0	0		

* Number of surviving juveniles was calculated by multiplying the reported percent survival times the starting number of fertilized eggs per treatment (200).

^N NOEC

^L LOEC

The bluegill dataset differs qualitatively from the fathead minnow and brook trout datasets because the response variable, juvenile survivorship, is a quantal (binary) rather than continuous variable. Quantal variables conform to a binomial distribution. Such data are typically analyzed via either probit transformation, as employed by Gensemer et al. (2007), or logit transformation of the proportions of responding and non-responding test subjects. Probits are normal equivalent deviates and logits are logistic equivalent deviates. These two transforms usually yield similar estimates of EC₅₀ values, but differ appreciably in their EC estimates in the tails of the distributions.

Environment Canada (2005) recommends logistic methods over probits for “... mathematical simplicity and other good reasons.” $\text{Logit} = \ln(p/1-p)$, where p is the proportion of effected test subjects (e.g., if juvenile survival were 30% for a particular treatment concentration, p would equal 0.3 and the logit transform would equal -0.8473).

The logit transform linearizes the sigmoidal logistic response curve (Environment Canada 2005, StatSoft 2006). Furthermore, in fitting the logit model, the control observations can be excluded, as they do not provide any information, unless a background parameter is included (OECD 2006).

Both Environment Canada (2005) and OECD (2006) note that it is common practice to correct the data for background response prior to analysis (for example via Abbott’s correction), but that such pre-treatment of the data is unsound statistical practice that can result in substantive overestimation of EC_x values. The bias increases as the control effect being adjusted for increases. We fit a focal segment of the bluegill dataset to a log-logit regression using results that were not control-adjusted prior to analysis. Thus, our prediction model yields unbiased estimates of proportion effect that can be control-adjusted for reporting purposes after-the-fact. The dataset is reasonably monotonic until the highly anomalous result for the treatment at a concentration of 50.7 ug/L CN. Gensemer et al. (2007) censored that point as an outlier. Because our $SSEC_x$ range extended up to only 41.7 ug/L CN (Table 4) the 50.7 ug/L CN treatment did not fall within our focal segment of concern. The last three treatments in our focal segment produced results of greater than 84% effect which would place them in the nonlinear upper tail of the sigmoidal curve (Figure 2), but unlike a log-square root regression the logit transform will linearize points in the tails relative to intermediate effect points. Thus, for log-logit regression points that fall in tails do not have to be avoided in order to apply linear regression. The minor deviation from monotonicity in the last two points of our focal segment did not warrant data-smoothing. A summary of the logit transformed response data is presented in Table 11.

TABLE 11. Bluegill input data for effects modeling.

Treatment (free CN ug/L)	Mean surviving juveniles	Proportion Survival	Logit Proportion Survival
Control Mean	46.5	0.2325	-1.1942
4.9	37	0.1850	-1.4828
9.4	32.5	0.1630	-1.6361
19.9	5.5	0.0280	-3.5472
29.9	5	0.0250	-3.6636
40.6	7.5	0.0380	-3.2314
50.7	27	0.1350	-1.8575
64.6	0	0.0000	
84.4	0	0.0000	

^aFinal effects model based upon the shaded subset of data.

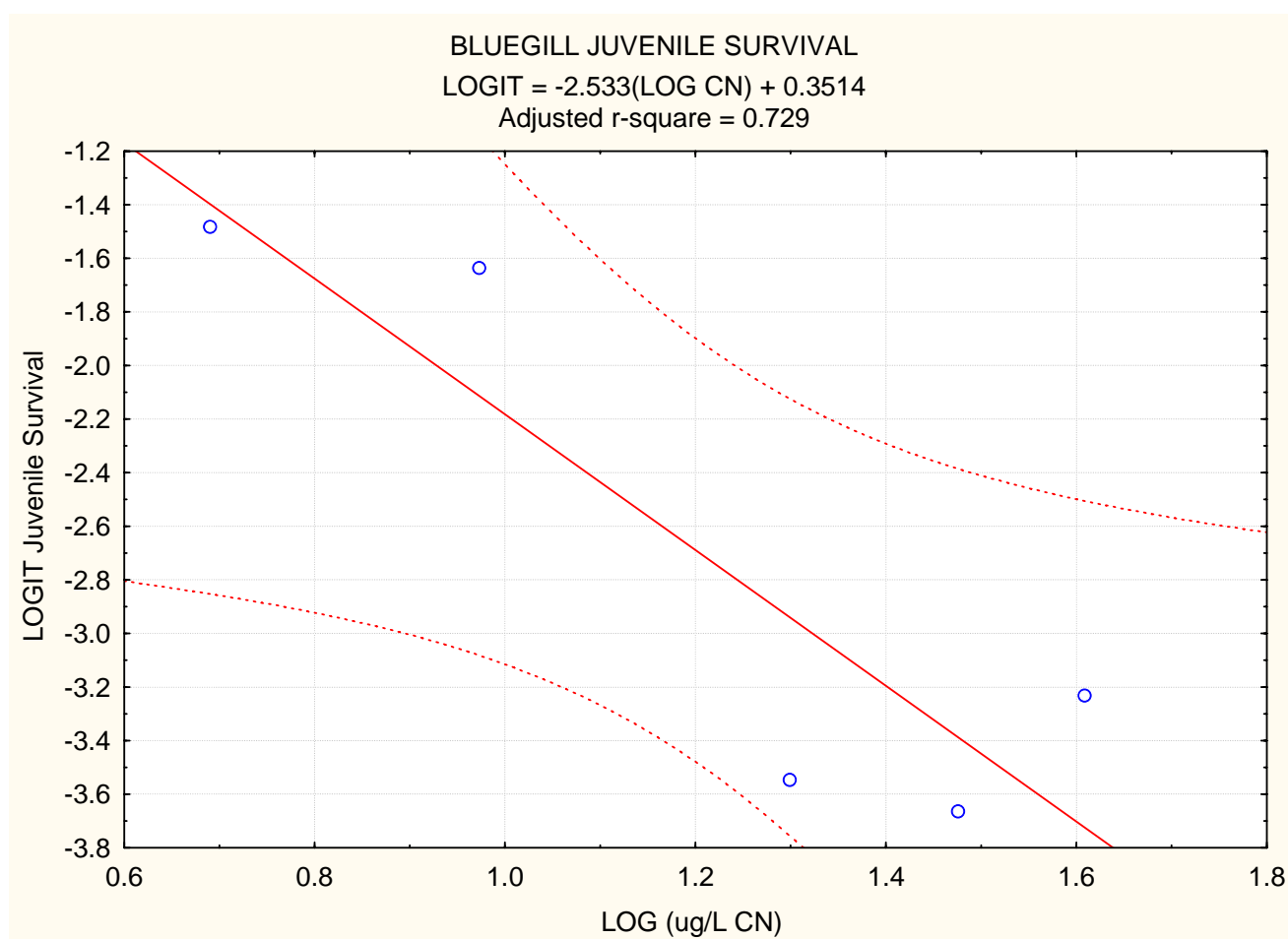
The resulting log-logit focal segment linear regression model does not show as strong a fit to the data as the fathead minnow model does, but with an adjusted r-square of 0.729

shows a reasonably good fit comparable to that achieved for the brook trout dataset. The regression equation is:

$$\text{Logit (proportion juvenile survival)} = -2.533 (\text{LOG CN}) + 0.3514$$

The regression plot is presented in Figure 5 and summary regression statistics are presented in Table 7. The regression was conducted using the multiple linear regression module of the *Statistica* software package (StatSoft 2006). Because we are dealing with small samples (five points in this case), we report the adjusted r-squared value which adjusts for the limited degrees of freedom in the model (StatSoft 2006).

FIGURE 5. Log-logit focal segment regression plot for bluegill juvenile survival.



Prediction Results

Effects predictions are generated by substituting LOG (SSEC_x) for LOG (CN) into the prediction regression equations. This was accomplished via the “predict dependent variable” algorithm in the multiple linear regression module of *Statistica* (StatSoft 2006). That algorithm also uses the estimated standard error of the regression coefficient to

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generate 95% confidence limits for the predicted point estimates (maximum likelihood estimates). For the fathead minnow and brook trout prediction regressions, the prediction and confidence limit output are in the form of square-roots of numbers of eggs. To convert those predictions to a percent effect, the predicted results were first squared and then scaled for percent change compared to the applicable smoothed control value according to the formula:

$$\% \text{ Effect} = [1 - (\text{predicted egg count} / \text{smoothed control value})] \times 100$$

Any predicted egg counts exceeding the smoothed control value were automatically converted to 0% effect. For the bluegill prediction regression, the prediction and confidence limit output are in the form of logit transforms for proportions of juvenile survivorship. The logit transforms are back-transformed to proportions by the formula:

$$\text{Proportion survival} = e^{(\text{logit})} / 1 + e^{(\text{logit})}$$

The predicted survival proportions are scaled for percent change compared to the reported control value according to the formula:

$$\% \text{ Effect} = [1 - (\text{predicted proportion survival} / \text{mean control proportion survival})] \times 100$$

Again, any predicted survivorship exceeding the observed mean control survivorship results in a percent effect prediction that is automatically converted to 0% effect. The raw input and output data for effects predictions are presented in Appendix E.

A summary of predicted effects and their estimated 95% confidence limits from each of the three prediction models for each of the 17 surrogate taxa from which listed evaluation species' LC₅₀ values were derived are presented in Table 12. The effects estimates are presented in Table 13 for the listed evaluation species based on matching up the effects estimates for surrogate taxa in Table 12 with the listed species linked to each surrogate taxon.

The EC₁₀ and EC₂₀ concentrations for each of our three regression models were also estimated.

The fathead minnow regression yielded an estimated EC₁₀ of 4.4 ug/L CN (95% CI = 2.6-6.2 ug/L CN) and an estimated EC₂₀ of 5.5 ug/L CN (95% CI = 3.5-7.4). By comparison, Gensemer et al. (2007) estimated an EC₂₀ of 6.0 ug/L CN from a log-probit analysis of the fathead minnow data, but did not report confidence limits for that estimate.

The brook trout regression yielded an estimated EC₁₀ of 2.6 ug/L CN (95% CI = 0.0-8.4 ug/L CN) and an estimated EC₂₀ of 4.1 ug/L CN (95% CI = 0.0-11.1). Gensemer et al. (2007) estimated an EC₂₀ of 7.7 ug/L by linear interpolation of the brook trout data, and again did not report confidence limits for that estimate. It is important to note that, for their analysis, Gensemer et al. (2007) did not average the controls, as we did, but used the

control which produced the lowest number of eggs (refer to the Table 8 footnote for more details).

The bluegill regression yielded an estimated EC₁₀ of 4.6ug/L CN (95% CI = 0.0-10.5 ug/L CN) and an estimated EC₂₀ of 5.3 ug/L CN (95% CI = 0.0-11.5). Gensemer et al. (2007) estimated an EC₂₀ of 5.6 ug/L CN from a log-probit analysis of the bluegill data, and also estimated an EC₂₀ of 8.9 ug/L CN for the bluegill data from EPA's TRAP program.

All of Gensemer et al.'s (2007) estimates fall within our 95% confidence limits, and in general show excellent agreement with our results even though Gensemer et al.'s methods differed from ours. This suggests that our results are not highly dependent on the particular statistical approach that we chose for our analysis.

Table 12. Estimated magnitude of effect of cyanide (at the CCC, 5.2 ug CN/L) on surrogate taxa for listed fish species (± 95% CL). The magnitude of effect was estimated using the regression model for each surrogate response species and SS EC_x value for each surrogate taxa (Table 1). For each surrogate taxa there were two estimates of effects on reproductive performance and one estimate of effects on early life stage survival.

Surrogate taxa used to estimate magnitude of effect on listed species	Surrogate species		
	Fathead Minnow	Brook Trout	Bluegill
	Reduction in the mean number of hatched eggs per spawn compared to controls	Reduction in the mean number of viable eggs per spawn compared to controls	Reduction in the number of surviving larvae/juveniles compared to controls
Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
Cypriniformes (order)	39% (28%, 49%)	26% (0%, 54%)	44% (0%, 80%)
Family Catostomidae			
<i>Xyrauchen texanus</i> (species)	39% (28%, 49%)	26% (0%, 54%)	44% (0%, 80%)
Cyprinidae (family)	31% (18%, 44%)	22% (0%, 53%)	33% (0%, 78%)
<i>Cyprinella monacha</i> (species)	68% (63%, 72%)	42% (23%, 58%)	76% (50%, 89%)
<i>Gila elegans</i> (species)	57% (51%, 63%)	36% (12%, 56%)	66% (30%, 84%)
<i>Notropis mekistocholas</i> (species)	59% (53%, 65%)	37% (14%, 56%)	68% (34%, 85%)
<i>Ptychocheilus lucius</i> (species)	63% (57%, 68%)	39% (18%, 57%)	71% (41%, 86%)
Perciformes (order)	36% (24%, 47%)	24% (0%, 53%)	40% (0%, 79%)
Percidae (family)	63% (58%, 68%)	39% (18%, 57%)	72% (43%, 87%)
<i>Etheostoma</i> (genus)	65% (60%, 70%)	40% (20%, 58%)	74% (46%, 88%)

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Surrogate taxa used to estimate magnitude of effect on listed species	Surrogate species		
	Fathead Minnow	Brook Trout	Bluegill
	Reduction in the mean number of hatched eggs per spawn compared to controls	Reduction in the mean number of viable eggs per spawn compared to controls	Reduction in the number of surviving larvae/juveniles compared to controls
<i>Etheostoma fonticola</i> (species)	81% (76%, 85%)	52% (37%, 64%)	86% (64%, 95%)
Order Salmoniformes, Family Salmonidae			
<i>Oncorhynchus</i> (genus)	60% (54%, 65%)	37% (15%, 57%)	69% (36%, 85)
<i>Oncorhynchus apache</i> (species)	87% (82%, 91%)	56% (42%, 68%)	90% (67%, 97%)
<i>Oncorhynchus clarki henshawi</i> (species)	80% (75%, 84%)	51% (36%, 63%)	85% (63%, 94%)
<i>Salmo salar</i> (species)	36% (24%, 47%)	24% (0%, 54%)	41% (0%, 79%)
<i>Salvelinus</i> (genus)	87% (83%, 92%)	57% (43%, 69%)	90% (68%, 97%)

Table 13. Estimated magnitude of effect of cyanide (at the CCC, 5.2 ug CN/L) on listed fish species (95% CL). There are two estimates for effects on fecundity and one estimate for effects on early life stage survival. Estimates are based on analyses using surrogate taxa (Table 12). Surrogate taxa were used to estimate LC₅₀'s for listed species Surrogate response species (fathead minnow, brook trout, bluegill) datasets were used to estimate magnitude of chronic effects.

Listed Species		Order/Family	Surrogate Taxa	Estimated reduction in fecundity and larvae/juvenile survival due to cyanide exposure (5.2 ug/L) based on surrogate species data sets.						
				Fathead Minnow (Reduction in the mean number of hatched eggs per spawn compared to controls)	Brook Trout (Reduction in the mean number of viable eggs per spawn compared to controls)	Bluegill (reduction in the number of surviving larvae/juveniles compared to controls)				
Gulf sturgeon	<i>Acipenser oxyrinchus desotoi</i>	Acipenseriformes Acipenseridae	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)				
Kootenai River white sturgeon	<i>Acipenser transmontanus</i>									
Pallid sturgeon	<i>Scaphirhynchus albus</i>									
Alabama sturgeon	<i>Scaphirhynchus suttkusi</i>									
Waccamaw silverside	<i>Menidia extensa</i>	Atheriniformes Atherinopsidae								
Modoc sucker	<i>Catostomus microps</i>	Cypriniformes Catostomidae	Cypriniformes (order)	39% (28%, 49%)	26% (0%, 54%)	44% (0%, 80%)				
Santa Ana sucker	<i>Catostomus santaanae</i>									
Warner sucker	<i>Catostomus warnerensis</i>									
Shortnose sucker	<i>Chasmistes brevirostris</i>									
Cui ui	<i>Chasmistes cujus</i>									
June sucker	<i>Chasmistes liorus</i>									
Lost River sucker	<i>Deltistes luxatus</i>									
Razorback sucker	<i>Xyrauchen texanus</i>						<i>Xyrauchen texanus</i>	39% (28%, 49%)	26% (0%, 54%)	44% (0%, 80%)
Spotfin chub	<i>Cyprinella monacha</i>						<i>Cyprinella monacha</i>	68% (63%, 72%)	42% (23%, 58%)	76% (50%, 89%)
Blue shiner	<i>Cyprinella caerulea</i>						Cypriniformes Cyprinidae	Cyprinidae (family)	31% (18%, 44%)	22% (0%, 53%)
Beautiful shiner	<i>Cyprinella formosa</i>									
Devils River minnow	<i>Dionda diaboli</i>									
Slender chub	<i>Erimystax cahni</i>									
Mohave tui chub	<i>Gila bicolor mohavensis</i>									
Owens tui chub	<i>Gila bicolor snyderi</i>									
Borax Lake chub	<i>Gila boraxobius</i>									
Humpback chub	<i>Gila cypha</i>									
Sonora chub	<i>Gila ditaenia</i>									
Gila chub	<i>Gila intermedia</i>									
Pahrnagat roundtail chub	<i>Gila robusta jordani</i>									
Virgin River chub	<i>Gila robusta seminuda</i>									

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Rio Grand silvery minnow	<i>Hybognathus amarus</i>					
Big Spring spinedace	<i>Lepidomeda mollispinis pratensis</i>					
Little Colorado spinedace	<i>Lepidomeda vittata</i>					
Spikedace	<i>Meda fulgida</i>					
Moapa dace	<i>Moapa coriacea</i>					
Palezone shiner	<i>Notropis albizonatus</i>					
Cahaba shiner	<i>Notropis cahabae</i>					
Arkansas River shiner	<i>Notropis girardi</i>					
Pecos bluntnose shiner	<i>Notropis simus pecosensis</i>					
Topeka shiner	<i>Notropis Topeka</i>					
Oregon chub	<i>Oregonichthys crameri</i>					
Blackside dace	<i>Phoxinus cumberlandensis</i>					
Woundfin	<i>Plagopterus agrentissimus</i>					
Ash Meadows speckled dace	<i>Rhinichthys osculus nevadensis</i>					
Kendall Warm Springs dace	<i>Rhinichthys osculus thermalis</i>					
Loach minnow	<i>Tiaroga cobitis</i>					
Bonytail chub	<i>Gila elegans</i>		<i>Gila elegans</i>	57% (51%, 63%)	36% (12%, 56%)	66% (30%, 84%)
Cape Fear shiner	<i>Notropis mekistocholas</i>		<i>Notropis mekistocholas</i>	59% (53%, 65%)	37% (14%, 56%)	68% (34%, 85%)
Colorado pikeminnow	<i>Ptychocheilus lucis</i>		<i>Ptychocheilus lucis</i>	63% (57%, 68%)	39% (18%, 57%)	71% (41%, 86%)
White River springfish	<i>Crenichthys baileyi baileyi</i>	Cyprinodontiformes Goodeidae				
Hiko White River springfish	<i>Crenichthys baileyi grandis</i>					
Railroad Valley springfish	<i>Crenichthys nevadae</i>					
Big Bend gambusia	<i>Gambusia gaigei</i>	Cyprinodontiformes Poeciliidae				
San Marcos gambusia	<i>Gambusia georgei</i>					
Clear Creek gambusia	<i>Gambusia heterochir</i>					
Pecos gambusia	<i>Gambusia nobilis</i>					
Gila topminnow (including Yaqui)	<i>Poeciliopsis occidentalis</i>					
Unarmored threespine stickleback	<i>Gasterosteus aculeatus williamsoni</i>	Gasterosteiformes Gasterosteidae				
Delta smelt	<i>Hypomesus transpacificus</i>	Osmeriformes Osmeridae				
Tidewater goby	<i>Eucyclogobius newberryi</i>	Perciformes Gobiidae	Perciformes	36% (24%, 47%)	24% (0%, 53%)	40% (0%, 79%)
Slackwater darter	<i>Etheostoma boschungii</i>	Perciformes Percidae	<i>Etheostoma</i> (genus)	65% (60%, 70%)	40% (20%, 58%)	74% (46%, 88%)
Vermilion darter	<i>Etheostoma chermocki</i>					

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Relict darter	<i>Etheostoma chienense</i>					
Etowah darter	<i>Etheostoma etowahae</i>					
Niangua darter	<i>Etheostoma nianguae</i>					
Watercress darter	<i>Etheostoma nuchale</i>					
Okaloosa darter	<i>Etheostoma okaloosae</i>					
Duskytail darter	<i>Etheostoma percnurum</i>					
Bayou darter	<i>Etheostoma rubrum</i>					
Cherokee darter	<i>Etheostoma scotti</i>					
Maryland darter	<i>Etheostoma sellare</i>					
Bluemask darter	<i>Etheostoma sp.</i>					
Boulder darter	<i>Etheostoma wapiti</i>					
Fountain darter	<i>Etheostoma fonticola</i>		<i>Etheostoma fonticola</i> (species)	81% (76%, 85%)	52% (37%, 64%)	86% (64%, 95%)
Amber darter	<i>Percina antesella</i>		Percidae (family)	63% (58%, 68%)	39% (18%, 57%)	72% (43%, 87%)
Goldline darter	<i>Percina aurolineata</i>					
Conasauga logperch	<i>Percina jenkinsi</i>					
Leopard darter	<i>Percina pantherina</i>					
Roanoke logperch	<i>Percina rex</i>					
Snail darter	<i>Percina tanasi</i>					
Ozark cavefish	<i>Amblyopsis rosae</i>	Percopsiformes Amblyopsidae	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
Alabama cavefish	<i>Spleoplatyrhinus poulsoni</i>					
Little Kern golden trout	<i>Oncorhynchus aguabonita whitei</i>	Salmoniformes Salmonidae	<i>Oncorhynchus</i> (genus)	60% (54%, 65%)	37% (15%, 57%)	69% (36%, 85)
Paiute cutthroat trout	<i>Oncorhynchus clarki seleniris</i>					
Greenback cutthroat trout	<i>Oncorhynchus clarki stomias</i>					
Gila trout	<i>Oncorhynchus gilae</i>					
Apache trout	<i>Oncorhynchus apache</i>					
Lahontan cutthroat trout	<i>Oncorhynchus clarki henshawi</i>					
Atlantic salmon	<i>Salmo salar</i>					
Atlantic salmon						
Bull trout	<i>Salvelinus confluentus</i>					
Pygmy sculpin	<i>Cottus paulus</i>	Scorpaeniformes Cottidae	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)

Other Effects Estimates

The estimates of effects presented in Table 13 are based largely on ICE LCL (lower confidence limit) LC_{50} values for listed fish evaluation species. Those are the LC_{50} values that we accept as sufficiently accounting for the uncertainties inherent in relying on surrogate data and numerous other untested assumptions to estimate the sensitivity of listed species to cyanide. The Service, NMFS, and EPA agreed that using ICE LCL values was preferable to the practice of applying arbitrary uncertainty factors.

However, EPA has, at various times, questioned whether the use of ICE LCL values might not be overly conservative. Therefore, we also estimated effect levels using ICE MLE (maximum likelihood estimates) LC_{50} values for listed fish evaluation species (via revised $SSEC_x$ estimates). Those results are presented in Appendix F. Based on the fathead minnow prediction model, which was the strongest model, the median levels of effect predicted for the 15 ICE surrogate taxa were 51% and 65%, respectively, for ICE MLE and ICE LCL. The number of surrogate taxa with a predicted effect of 35% or greater was 11 and 14, respectively, for ICE MLE and ICE LCL. Those differences indicate only modest conservatism conferred by ICE LCL-based effects estimates as compared to ICE MLE-based estimates. Such modest differences would not have a decision-making impact. For both sets of results, unacceptably high levels of effect would overwhelmingly be the predominant prediction.

Empirical Test of Method Performance

Because only three concentration-response datasets are available, there is almost no basis for testing our method performance (i.e., there are no known directly measured “true” values for effects to our listed fish evaluation species at a concentration of 5.2 ug/L CN). However, because the fathead minnow and brook trout datasets focused on essentially the same response variable (number of hatchable/viable eggs produced per spawn) we can perform two tests of method performance. For each species, we can directly estimate a predicted effect level at 5.2 ug/L CN using the species-specific regressions. Those would be our estimates of the “true” effect level. Next, we can use our surrogate method and estimate an $SSEC_x$ for each species on the other species’ response curve and evaluate the predicted effect level for that $SSEC_x$ value and compare the surrogate estimate to the estimated “true” value. The results are as follows:

The directly estimated fathead minnow effect level at 5.2 ug/L CN is 18% with a 95% CI of 0%-34%. The fathead minnow $SSEC_x$ value on the brook trout response curve would be 3.2 ug/L CN, which yields an effects estimate of 15%. That is nearly identical to estimated “true” value and easily within the 95% CI for the “true value”.

The directly estimated brook trout effect level at 5.2 ug/L CN is 25% with a 95% CI of 0%-54%. The brook trout $SSEC_x$ value on the fathead minnow response curve would be 8.4 ug/L CN, which yields an effects estimate of 38%. Again, that is within the 95% CI for the “true” value, although our estimate of the “true” value is not very precise and therefore the 95% CI is fairly wide.

In summary, in both test cases, the estimated effect level derived from our surrogate methodology is not significantly different from the estimated “true” value in a statistical sense, but the second comparison has low statistical power. Further validation testing of this sort should be done as concentration-response datasets become available for more species using a comparable response variable, but it is reassuring that in these test cases our method yielded results that were nearly identical to the “true” value in one case and reasonably close to the “true” value in the other case.

Derivation of the Criterion Continuous Concentration (CCC)

Our analysis predicts that the listed fish evaluation species considered in this Biological Opinion would be highly affected by exposure to cyanide at the CCC. These results prompted us to better understand the level of protection that aquatic life criteria in general and the cyanide criterion in particular were intended to provide.

The objective of the Clean Water Act (CWA) is to “*restore and maintain the chemical, physical and biological integrity of the Nation’s waters*” with an interim goal of “*water quality which provides for the protection and propagation of fish, shellfish and wildlife and provides for recreation in and on the water*”, where attainable.

Section 304(a) of the CWA requires the EPA Administrator to publish “*criteria for water quality accurately reflecting the latest scientific knowledge on the kind and extent of all identifiable effects on the health and welfare including, but not limited to, plankton, fish shellfish, wildlife, plant life....*” including information “*on the factors necessary for the protection and propagation of shellfish, fish, and wildlife for classes and categories of receiving waters...*”

Water quality criteria are developed to protect different attributes or uses of water bodies, referred to as designated uses. Aquatic life water quality criteria are intended to protect waters where the designated use includes the protection and propagation of fish, shellfish and wildlife.

EPA’s *Guidelines for Deriving Numeric National Water Quality Criteria for the Protection of Aquatic Organisms and Their Uses* (Stephan et al. 1985) has been used to derive aquatic life water quality criteria, including the cyanide criterion, since the mid 1980’s. This excerpt from the guidelines document describes, from an operational perspective, the intended purpose of national criteria and their limitations:

“Because aquatic ecosystems can tolerate some stress and occasional adverse effects, protection of all species at all times and places is not deemed necessary. If acceptable data are available for a large number of appropriate taxa from an appropriate variety of taxonomic and functional groups, a reasonable level of protection will probably be provided if all except a small fraction of the taxa are protected, unless a commercially or recreationally important species is very sensitive. The small fraction is set at 0.05 because other fractions resulted in criteria that seemed too high or too low in

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comparison with the sets of data from which they were calculated. Use of 0.05 to calculate the Final Acute Value does not imply that this percentage of adversely affected taxa should be used to decide in a field situation whether a criterion is too high or too low or just right.”

Based on this description it appears that National criteria are intended to protect ecosystems and ecosystem functions and not necessarily to protect all taxa or species within the ecosystem all of the time. It is expected that a small fraction (5%) of taxa or species may be adversely affected. The guidelines go on to say:

“To be acceptable to the public and useful in field situations, protection of aquatic organisms and their uses should be defined as prevention of unacceptable long-term and short-term effects on (1) commercially, recreationally, and other important species and (2) (a) fish and benthic invertebrates assemblages in rivers and streams, and (b) fish, benthic invertebrate, and zooplankton assemblages in lakes reservoirs, estuaries, and oceans.”

Thus, the level of protection afforded to aquatic organisms should prevent *unacceptable* long-term and short term effects. The threshold for unacceptable long-term or chronic effects is estimated by the CCC. The guidelines indicate that some adverse effects may occur at the CCC but they should not rise to a level that is unacceptable:

“However, it is important to note that this is a threshold of unacceptable effect, not a threshold of adverse effect. Some adverse effect, possibly even a small reduction in the survival, growth, or reproduction of a commercially or recreationally important species, will probably occur at, and possibly even below, the threshold. The Criterion Continuous Concentration (CCC) is intended to be a good estimate of this threshold of unacceptable effect.”

EPAs Water Quality Standards Handbook (EPA 1994) defines the CCC as *“the EPA national water quality criteria recommendation for the highest instream concentration of a toxicant or an effluent to which organisms can be exposed indefinitely without causing unacceptable effect”*.

The guidelines provide some guidance for determining what constitutes unacceptable levels of adverse effects. The following guidance is for monitoring programs designed to detect unacceptable levels of adverse effects in the field:

“The amount of decrease in the number of taxa or number of individuals in an assemblage that should be considered unacceptable should take into account appropriate features of the body of water and its aquatic community. Because most monitoring programs can only detect decreases of more than 20 percent, any statistically significant decrease should usually be considered unacceptable. The insensitivity of most monitoring programs greatly limits their usefulness for studying the validity of criteria because unacceptable changes can occur and not be detected. Therefore, although

limited field studies can sometimes demonstrate that criteria are underprotective, only high quality field studies can reliably demonstrate that criteria are not underprotective.”

Here, the guidelines appear to suggest that because of the insensitivity of monitoring programs only relatively high levels of effect (>20%) may be detected as statistically significant, and suggest that lower, yet unacceptable, levels of effects may go undetected.

However, the National criteria are typically derived using chronic toxicity data from laboratory tests rather than field studies. Chronic data from individual tests are analyzed and a chronic value is computed according to the following guidance:

“A chronic value may be obtained by calculating the geometric mean of the lower and upper chronic limits from chronic tests or by analyzing chronic data using regression analysis. A lower chronic limit is the highest tested concentration (a) in an acceptable chronic test, (b) which did not cause an unacceptable amount of adverse effect on any of the specified biological measurements, and (c) below which no tested concentration caused an unacceptable effect. An upper chronic limit is the lowest tested concentration (a) in an acceptable chronic test, (b) which did cause an unacceptable amount of adverse effect on one or more of the specified biological measurements, and (c) above which all tested concentrations also caused such an effect.”

For most aquatic life criteria that have been derived thus far, including the cyanide criterion, chronic values have been obtained by calculating the geometric mean of the lower and upper chronic limits. In practice, the upper and lower chronic limits are often statistically determined by hypothesis testing. The lower limit is typically the No Observable Effect Concentration (NOEC), which is defined as the highest test concentration where the effects are not statistically significantly different from controls. The upper limit is typically the Lowest Observable Effect Concentration (LOEC), which is defined as the lowest test concentration where the effects are statistically significantly different from controls. The guidelines recommend that the magnitude of effect associated with the upper and lower chronic limits should be considered when determining values that appropriately estimate acceptable and unacceptable levels of adverse effect:

“Because various authors have used a variety of terms and definitions to interpret and report results from chronic tests, reported results should be reviewed carefully. The amount of effect that is considered unacceptable is often based on a statistical hypothesis test, but might also be defined in terms of a specified percent reduction from the controls. A small percent reduction (e.g., 3%) might be considered acceptable even if it is statistically significantly different from the control, whereas, a large percent reduction (e.g., 30%) might be considered unacceptable even if it is not statistically significant.”

Based on this guidance, the threshold for unacceptable adverse effects would be estimated by the chronic value. The magnitude of effect at the threshold would then be equivalent to the magnitude of effect at the chronic value. For chronic criteria derived using hypothesis tests, this would be the magnitude of effect occurring at a concentration

equal to the geometric mean of the NOEC and LOEC, that is, somewhere between an acceptable and unacceptable level of adverse effect. The guidelines do not specify a level of adverse effect on which the threshold for unacceptability should be based. The only mention of a numeric value or range is provided in the guidance for selecting chronic limits (mentioned above) and suggests that this threshold may lie between 3% and 30%.

Thus, for a given species or test the magnitude of effect at the chronic value will depend on the magnitude of effect at the lower and upper chronic limits. We followed this approach for estimating the magnitude of effect occurring at the cyanide CCC. The freshwater cyanide CCC was derived based on chronic toxicity data for 4 species (Table 14): 3 fish (fathead minnow, brook trout, and bluegill) and 1 invertebrate (*Gammarus pseudolimnaeus*). Chronic values for each species were obtained by calculating the geometric mean of the lower and upper chronic limits. The magnitude of effect at the lower and upper chronic limits was calculated by comparing responses at the lower and upper limits to controls. For fathead minnow and brook trout these effects were expressed as reduction in the mean number of eggs spawned per female compared to controls; for the bluegill the effect was reduction in larvae/juvenile survival compared to controls; and for *G. pseudolimnaeus* the effect was a reduction in the mean number of eggs or young per gravid female relative to controls.

We then estimated the magnitude of effect at the chronic value by linear interpolation between lower and upper chronic limits (Table 14). Based on these calculations the magnitude of effect at the chronic values for the fathead minnow, brook trout, bluegill and *G. pseudolimnaeus* would be 52%, 32%, 54%, and 47%, respectively. According to the guidelines, if there were a sufficient number of chronic values (i.e., chronic values for species in 8 phylogenetic families) the chronic criterion could be computed directly from the distribution of chronic values. If there were fewer chronic values, as was the case for cyanide, the chronic criterion would be computed using Acute-Chronic Ratios (ACR). ACRs for the 4 freshwater species were reported in the cyanide criterion document and are shown in Table 14. The ACRs were calculated by dividing the species mean acute value (i.e., mean LC₅₀ for the species) by the chronic value. For example, the ACR for fathead minnow (7.633) was computed by dividing 125.1 ug CN/L (the mean LC₅₀ for the species) by 16.39 ug/ CN/L (the chronic value). Thus, the ACR is the ratio between the concentration of cyanide causing 50% lethality (following acute exposure) and the concentration following chronic exposure that causes a level of adverse effect that is at the threshold of unacceptability, i.e., 52% for fathead minnow. The guidelines require that, for criteria derivation, the geometric mean of individual species ACRs is used to obtain the Final ACR. For cyanide, the freshwater Final ACR was 8.562 (Table 14). We estimated the magnitude of chronic effects associated with the Final ACR to be about 45% (Table 14).

The Final ACR and the Final Acute Value (FAV) were then be used to derive the CCC. The guidelines describe how the FAV is computed. In short, the FAV is set equal to the 5th percentile estimate from the distribution of genus mean acute values. In other words, the FAV represents the genus with acute sensitivity (LC₅₀) in the *sensitive tail* of the distribution where, theoretically, approximately 5% of the genera would be more

sensitive and about 95% of the genera would be less sensitive. Based on this analysis, the FAV for cyanide was determined to be 62.68 ug CN /L. The guidelines also include provisions for adjusting the FAV to protect commercially and recreationally important species:

“However, in some cases, if the Species Mean Acute Value of a commercially or recreationally important species is lower than the calculated Final Acute Value, then that Species Mean Acute Value replaces the calculated Final Acute Value in order to provide protection for that important species.”

For cyanide, the FAV was lowered from 62.68 ug/L to 44.73 ug/L because the Species Mean Acute Value for rainbow trout (44.73 ug/L) was below the calculated FAV. The cyanide criterion (5.2 ug/L) was then derived by division of the FAV (44.73 ug/L) by the

Table 14. Chronic toxicity data used by EPA to derive the freshwater chronic criterion for cyanide. Effects levels were calculated using data from the original papers.

Species	Chronic Limits ¹				Chronic Value ²		LC ₅₀ ³ (ug CN/L)	ACR ³	
	Lower		Upper		(ug CN/L)	Effect			
	(ug CN/L)	Effect	(ug CN/L)	Effect					
Fathead Minnow	13.3	47%	20.2	58%	16.39	52%	125.1	7.633	
Brook Trout	5.6	18%	11.0	53%	7.849	32%	83.14	10.59	
Bluegill	9.3	30%	19.8	88%	13.57	54%	99.28	7.3	
Gammarus	16	0%	21	100%	18.33	47%	167	9.111	
Geometric mean							45%		8.562

¹ Lower and upper chronic limits were taken from the cyanide criteria document. For fathead minnow and bluegill these values were determined statistically (i.e., NOEC and LOEC identified via hypothesis tests). Effect levels were taken from Tables 5, 8 and 10 in the Effects section of the BO and from Oseid and Smith 1979.

² Chronic values were taken from the cyanide criteria document. Effect levels associated with the chronic values were estimated by linear interpolation between the effects at the lower and upper chronic limits.

³ Acute-Chronic Ratios were taken from the cyanide criteria document.

Final ACR (8.562). Thus the chronic criterion, 5.2 ug CN/L, was based on the concentration intended to protect rainbow trout from unacceptable adverse effects. Based on our estimate of the magnitude of effect associated with the Final ACR, we estimate the magnitude of adverse effects occurring to rainbow trout at the chronic criterion to be approximately 45%. This value is higher than we would have expected considering it is intended to represent the threshold for unacceptable adverse effects. However, the magnitude is in line with effects we predicted for the listed fish evaluation species, most of which were estimated to be as (or more) sensitive to cyanide as rainbow trout.

The same conclusion reached above, that NOEC/LOEC-based estimates of “chronic values” can correspond to $\geq 40\%$ adverse effect, has also been reached by others. Decades ago Suter et al. (1987) reported that MATC’s for fish fecundity, on average,

corresponded to a 42% level of adverse effect (MATC = Maximum Aceptable Toxicant Concentration; a term for the geometric mean of the NOEC and LOEC from a given toxicity test and often assigned by EPA as the estimated “chronic value” from a test). Other response endpoints were found to correspond to average adverse effect levels of 12-35%. More recently, SETAC (Society for Environmental Toxicology and Chemistry) convened a panel of experts (Reiley et al. 2003) who concluded that “...[toxicity] tests with high variability may result in an(sic) NOEC corresponding to a response greater than 40% different from the control.” Moore and Caux (1997) statistically examined nearly 200 toxicity data sets and found that most NOEC’s (76.9%) exceeded a 10% adverse effect level and most LOEC’s (62.4%) exceeded a 30% effect level. Various other researchers have noted a variety of adverse effect levels for NOEC’s, such that Crane and Newman (2000) were led in summary to conclude that “...[NOEC] effect levels from individual tests ranged from nearly 0% to nearly 100%.” For seven cyanide toxicity tests with sufficient data for comparison, Gensemer et al. (2007: Figure 3-7) found in all cases that the geometric mean of the NOEC and LOEC corresponded to an adverse effect level of $\geq 20\%$ (how much greater was not reported).

Because of the highly variable and often substantive levels of effect associated with NOEC’s, LOEC’s, MATC’s, and with the “chronic values” based on them, and for numerous other reasons, a strong professional consensus recommendation to avoid using NOEC/LOEC-based estimates for regulatory thresholds (when possible) has been expressed repeatedly. For example, there was an ISO (International Organization for Standardization) resolution (ISO TC147/SC5/WG10 Antalya 3) as well as an OECD (Organisation for Economic Co-operation and Development) workshop recommendation (OECD 1998) that the NOEC should be phased out from international standards (OECD 2006:14). Environment Canada (2005) notes, that there is a growing literature which points out many deficiencies of the NOEC approach (Suter et al. 1987, Miller et al. 1993, Pack 1993, Noppert et al. 1994, Chapman 1996, Chapman et al. 1996, Pack 1998, Suter 1996, Moore and Caux 1997, Bailer and Oris 1999, Andersen et al. 2000, Crane and Newman 2000, Crane and Godolphin 2000). Moving away from the NOEC/LOEC approach was also among the recommendations of the SETAC panel for improving the scientific basis of water-quality criteria (Reiley et al. 2003).

Accordingly, EPA has begun employing a regression approach for estimating “chronic values” whenever sufficient data are available to do so. For example, in the 1999 update for ammonia water quality criteria EPA used regression analyses to estimate 20% effect concentrations (EC₂₀’s) from individual toxicity tests and used those EC₂₀’s as estimates of “chronic values” (EPA 1999). Likewise, estimated EC₂₀’s have been the basis for estimating “chronic values” in recently proposed updates for copper and selenium water quality criteria (EPA 2003a, 2004). EPA’s choice of the EC₂₀ as a basis for estimating “chronic values” was justified from statistical considerations rather than from biological or demographic considerations:

“To make [chronic values] reflect a uniform level of effect, regression analysis was used here both to demonstrate that a significant concentration-effect relationship was present and to estimate [chronic values] with a consistent level of effect. Use of regression

analysis is provided for on page 39 of the 1985 Guidelines (U.S. EPA 1985b). The most precise estimates of effect concentrations can generally be made for 50 percent reduction (EC50); however, such a major reduction is not necessarily consistent with criteria providing adequate protection. In contrast, a concentration that caused a low level of reduction, such as an EC5 or EC10, is rarely statistically significantly different from the control treatment. As a compromise, the EC20 is used here as representing a low level of effect that is generally significantly different from the control treatment across the useful chronic datasets that are available for ammonia.”

Pack (1993) asserted that most ecotoxicologists consider effects in the range of 5-20% to be biologically acceptable depending on the species involved and the type of effect. However, EPA appears to have chosen the top end of that range based more on the expected statistical power of toxicity tests than on a serious examination of the typical demographic sensitivity of biotic populations to a 20% adverse effect on survival, growth, or reproduction. Furthermore, 95% statistical confidence limits for most EC₂₀ estimates are likely to extend well into adverse effect levels that would be of unquestionably serious demographic concern for most organisms. As evident from the above discussion, most chronic criteria derived by EPA, including for cyanide, are highly likely to be associated with $\geq 20\%$ adverse effect level for species at the vulnerable end of species sensitivity distributions (such as the subset of ESA-listed species we are evaluating). Therefore, it should be no surprise that our estimated effect levels for such species at the current cyanide CCC of 5.2 ug/L are almost always higher than 20% and in some cases substantively higher.

Population Responses to Reductions in Fecundity and Juvenile Survival

Laboratory experiments have demonstrated that even closely related fish species can demonstrate great differences in sensitivity when exposed to the same chemical, as measured by differences in acute or chronic toxicity values. This variability in sensitivity has been related to differences in species' physiology and life history strategies. Similarly, population modeling and experimental studies have shown that variation in population-level responses to environmental toxicity can also be expected among species as a consequence of factors such as life history strategies, life stage affected, and density dependence. Studies have also demonstrated that chronic toxicity can lead to population decline and extirpation.

Under the ESA, in determining whether a proposed Federal action is likely to jeopardize the continued existence of a listed species under the ESA, we assess whether the proposed activity reasonably would be expected to appreciably reduce the likelihood of survival and recovery of a listed species by reducing its reproduction, numbers, or distribution. Two common metrics used in population modeling to assess effects of perturbations on populations are population growth rate and time to or probability of extinction.

Population growth rate is the change in a population size over a unit time period. Long-term reductions in population growth rate as low as 5% has been shown to significantly

increase a population's likelihood of extinction (Snell and Serra 2000). Population growth rate can be positive when the population is increasing, negative when decreasing, or zero when the net difference between births, deaths, and migration is zero and the population is stable. For listed species, populations may exist in any of these states depending on its recovery status. Our analysis determines the relative predicted effects of the action to the population growth rate, regardless of its starting value.

Using known parameters of a species' life history, sensitivity analyses can be conducted to determine which parameters, when modified, will have the greatest impact on the species' population growth rate. Elasticity analysis is one type of sensitivity analysis that is commonly used in conservation biology to demonstrate the relative contributions to population growth rate made by life cycle transitions, based on vital rate statistics for survival, growth and fertility. While these types of analyses cannot predict absolute effects to population size, because they quantify the relative importance of an element to changes in population growth rate, they can help focus management decisions on those demographic parameters that exhibit the largest elasticity, and thus, the largest impact on population growth (de Kroon et al. 2000). However, elasticity analysis requires the development of a population model, for which adequate data are often scarce. Because this type of demographic data is often lacking for threatened and endangered species in particular, the need to develop generalized approaches for classifying population responses to perturbation for rare species has been recognized (Heppell et al. 2000, Dennis et al. 1991).

Several authors have examined the effect of life history strategies on the elasticities of various demographic measures. In evaluating demographic parameters of 50 mammal populations with different life history strategies, Heppell et al. (2000) found that phylogeny alone is often not a reliable indicator of which vital rates (survival, growth and fertility) will have the greatest impact on elasticity. Instead, the authors found that species that mature early and have high reproductive output had high fertility elasticities and low adult survival elasticities. Conversely, for those which mature late and have long lifespans, fecundity and early offspring survival are less important than survival of juveniles to maturity to changes in population growth rate. Calow et al. (1997) also found that the relative importance of juvenile fish survival can vary according to reproductive strategy. These authors concluded that reductions in juvenile survival would have the greatest impact on semelparous fish species, in which adults die after reproduction, a lesser impact on a moderately iteroparous population, in which adult postreproductive survival is intermediate, and the least impact on strongly iteroparous species, in which adult survival after reproduction is high. These assumptions held true for elasticity analysis of the green sturgeon, a fish species with life history patterns such as late-maturity and long-life that are common to other sturgeon (Heppell 2007). Juvenile survival had relatively lower elasticity values than adult and subadult survival, with compensation for the loss of adults requiring much larger increases in young-of-the-year survival than would be commensurate with the loss. However, other authors have found increased importance of juvenile survival for sturgeon, despite their lifespan (Gross et al. 2002, Paragamian and Hansen 2008). Gross et al. (2002) hypothesized that this

difference was due to the vastly larger fecundity of sturgeon as compared to other long-lived species.

Vélez-Espino et al. (2006) argue the need for a broadscale summary of species' population dynamics to help guide the conservation biology of freshwater fishes, for which information on life history is often limited. Using information, on adult survival, juvenile survival, and fecundity, the authors performed elasticity analyses on 88 species of freshwater fish and found that they could be classified into 4 functional groups with regard to the sensitivity of their population growth rates:

1. species most sensitive to perturbations in adult survival
2. species most sensitive to perturbations to adult and juvenile survival
3. species most sensitive to perturbations to juvenile survival
4. species most sensitive to perturbations to juvenile survival and fecundity

These groups are characterized by decreased age at maturity, longevity, and reproductive lifespan as one moves from group 1 to group 4. Age at maturity, reproductive lifespan, fecundity, juvenile survivorship, and longevity were all correlated with adult survival and fecundity. However, the best predictors of elasticity patterns were longevity, which explained 93% of the variability in the elasticity of adult survival, and age at maturity, which explained 92% of the variability in the elasticity of fecundity. The authors also found that elasticities are highly conserved among genera within the same taxonomic family

Spromberg and Birge (2005) also found that life history strategies influence effects to populations. The five life history strategies they modeled encompassed differences in stage-specific survival, fecundity and hatch success, number of spawning events, and life-span. The authors found that regardless of strategy, changes in the number of young-of-the-year stage individuals had the greatest impact on population growth rate. However, the relative contribution of this parameter was greatest for life history strategies with multiple spawnings, high fecundity, and short lifespans as opposed to those with longer lifespan, which had increased elasticity of adult survival.

Spromberg and Meador (2005) linked toxicant effects on immune suppression, reproductive development, and growth reduction to demographic traits in Chinook salmon (*Oncorhynchus tshawytscha*) and modeled their influence on population growth rate. Overall, effects to first- and second-year survival had the greatest elasticities, with constant reductions to first year survival as low as 10% achieving population declines ranging from 35-78% compared to controls. Other studies have demonstrated the importance of first year survival in this species (Kareiva et al. 2000). Spromberg and Meador (2005) also found that models which incorporated effects to both survival and reproduction were additive, indicating the importance of evaluating the overall impact of all potential impacts to population growth.

Many listed species populations are limited by the amount of adequate habitat or resources and experience some degree of density dependence. Density-dependence at any life stage must be considered in elasticity analysis in order to yield appropriate results (Grant and Benton 2000, Hayashi et al. 2008). In a review of toxicant impacts on

density-limited populations, Forbes et al. (2001) noted that the full range of interactions have been found between toxicant stress and density dependence, including less than additive, additive, and more than additive effects. Also, the type of effect may vary with increasing toxicant concentration from one that ameliorates density dependent effects at low toxicant concentrations to one that exacerbates density dependent effects at higher toxicant concentrations. Case studies which incorporate density-dependence into population modeling demonstrate this variability, with overall impacts to populations shown to be both lesser (Van Kirk and Hill 2007) and greater (Hayashi et al. 2008) than the level of effect that would be predicted from individual response depending on the situation. In time, density-dependant populations may rebound, stabilize at a lower absolute population number, or continue to decline until the population is extirpated (Forbes et al. 2001). Modeling exercises have demonstrated cases in which populations stabilize at new, lower equilibrium abundances in response to a constant impact (van Kirk and Hill 2007, Spromberg and Meador 2005).

A species' likelihood of persistence can also be estimated by modeling the species' time to extinction or probability of extinction. Population viability analysis (PVA) uses simulation modeling to identify threats to species and to assess the vulnerability of populations to these extinction risks. These models incorporate demographic parameters such as fecundity, survivorship, age structure, and population size, but can also incorporate effects to the environment such as habitat degradation and catastrophic events. As for the evaluation of population growth rate, sensitivity analysis is used to determine which factors have the greatest impact on population persistence, and many experts feel that parsing out these influential factors for management purposes is the best utilization of these models, as opposed to absolute predictions of population decline. Though PVA also requires a depth of demographic data that is often lacking for listed species, even PVAs with little data incorporated can be useful in comparative analyses of management considerations (Akçakaya and Sjögren-Gulve 2000).

Only a limited number of PVAs have been performed for listed aquatic species or other closely related species. A PVA for two darter species, the slackwater darter (*Etheostoma boschungii*) and the holiday darter (*E. brevirostrum*), found that fertility made the largest relative contribution to population growth, with juvenile survivability a more influential contributor to fertility than egg production (Hartup 2005). This conclusion held true regardless if the species was a single or multiple-batch spawner. In modeling the contributions of population size, age structure, and migration rate on the leopard darter (*Percina pantherina*), migration had the greatest influence on persistence (Williams et al. 1999). Catastrophe also played a significant role in persistence of the species, especially considering that the species is short-lived, has only one reproductive opportunity, and is restricted to few isolated populations. A similar analysis of the freshwater rotifer *Brachionus calyciflorus* also found that reductions in growth rate resulted in substantially increased probability of extinction when coupled with catastrophic population reductions (Snell and Serra 2000). A PVA analysis for the gila trout (*Oncorhynchus gilae*) revealed that the number of populations was the most influential of several life history parameters, and that the model was relatively insensitive to changes in population size and proportional abundance of age classes (Brown et al. 2001). The model was also sensitive

to large change in fecundity, producing significant changes in the probability of extinction when halved.

There are few field studies of pollutant effects on populations. Kidd et al. (2007) studied the effects of 17 α -ethynlestradiol (EE2) on fathead minnows in Canadian experimental lakes over a 7-year period. EE2, a synthetic estrogen found in birth control pills, was introduced at concentrations found in the receiving waters downstream from municipal wastewater dischargers. The fathead minnow, a short-lived species, was the first to show population collapse, but recovered once the pollutant stress was removed.

Summary of Population Responses to Reductions in Fecundity and Juvenile Survival

Modeling and experimental studies have shown that chronic toxicity to pollutants can lead to population decline and extirpation. Variation in population-level responses to environmental toxicity can be expected among species as a consequence of factors like species life history strategies, life stage affected, density dependence, and magnitude of toxicant stress. Although the degree varied among different life history strategies, fecundity and juvenile survival remained a highly influential demographic parameter throughout modeled scenarios, with adult survival taking on greater importance in long-lived species. These results must be coupled with other influences on the population status, such as the degree of density dependence and additional environmental perturbations such as catastrophes. Although population modeling often requires more demographic information than is available for threatened and endangered species, careful selection of surrogates and use of their data may allow for extrapolation from models for species with similar life histories.

Individual Species and Critical Habitat Accounts

Acipenseridae

GULF STURGEON

Acipenser oxyrinchus desotoi

Gulf sturgeon exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. Because no data for cyanide toxicity to sturgeon exist, LC50 values for sturgeon were derived from the 5% SSD concentration for the class Actinopterygii, which encompasses all known cyanide toxicity data for fish. From this data, we developed quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13).

Compared to control populations, we estimate Gulf sturgeon exposed to cyanide at the CCC could experience an approximate 48% reduction in the number of hatched eggs. We estimate that Gulf sturgeon exposed to cyanide at the CCC could experience an approximate 56% reduction in survival of young fish through the first year. Though no cyanide-specific data exist for this species, there are data from other chemicals that support the relative sensitivity of sturgeon to contaminants. Dwyer et al. (2005) tested the relative sensitivity of 18 fish and 1 amphibian species to five chemicals. Of these, the two sturgeon tested, the Atlantic sturgeon (*A. oxyrinchus*) and the shortnose sturgeon (*A. brevirostrum*) ranked first and second, respectively, in overall sensitivity. For all five chemicals, sturgeon were as or more sensitive than rainbow trout, for which reductions in viable eggs spawned and juvenile survival were estimated at 52% and 61%, respectively (Table 12).

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation).

Sturgeon have naturally high adult survival. Several authors have suggested that the rate of survival may be so high that management at the levels of these age classes is unlikely to improve their survival or increase population growth rate (Heppell 2007, Gross et al 2002). As such, recovery efforts are often based upon increasing survival in juvenile age classes. A population viability analysis of the Suwannee River population of the Gulf sturgeon found that slight changes in egg-to-age-1 mortality would strongly influence the recruitment and subsequent population (Pine et al. 2001). Decreasing the estimated 99.96% mortality for this life stage just 0.05 percentage points resulted in a 10-fold increase in population size, with a 5-fold increase in the number of recruits.

Gross et al (2002) modeled population growth rates for three species of sturgeon that varied in life history traits such as size, lifespan, age to maturity, and migration. All three sturgeons showed similar elasticity profiles, and thus the authors concluded that general interpretation could be applied to sturgeon across species. In contrast to other elasticity profiles for long-lived species, elasticity in sturgeon was highest in individual young-of-the-year and juvenile age classes, dropped at the onset of maturity, and continued to decline for each successive adult age class. Fecundity had relatively low elasticity, as the effects of changes in fecundity are shared among all adult age classes of these long-lived species, and the value of changes to egg numbers is lessened by the high mortality of the young-of-the-year age class. The authors concluded that population growth rate will show little response to improvements in fecundity, but greater responses in survival at either the young-of-the-year or juvenile age classes. However, since survival of the

juvenile and adult age classes is naturally high, improvements at these stages will have smaller effects to improving population growth rate than increases to survival of young-of-the-year, when natural mortality is greater. The authors note that among biologists and managers involved in sturgeon conservation, habitat improvement was regarded as the most important conservation undertaking for sturgeon. Results from this study indicate that restoration efforts should target the survival of age classes with high elasticity, specifically young-of-year and juvenile.

Paragamian and Hansen (2008) drew similar conclusions in modeling effects on population growth of the Kootenai River white sturgeon. The authors found that subadult and adult survival (>90%) was much higher than that of juveniles (40% in the first year), and recovery was most dependant on increasing first-year survival. The authors suggested that to have the largest effect on recovery, the managers should increase the current targeted recruitment rate.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Gulf sturgeon reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survival of young fish through the first year. Gulf sturgeon may also experience effects on growth, swimming performance, condition, and development. While sturgeon have developed a life history that allows them to cope with low survivorship to maturity and occasional hits to recruitment, these adaptation are unlikely to compensate for a constant reduction in both fecundity and early life stage survival. The reductions we estimate in survival of young fish through the first year in particular would substantially decrease recovery of this species. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. We would anticipate a consequent reduction in numbers of Gulf sturgeon. An effected Gulf sturgeon population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Gulf sturgeon are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion likely reduces the reproduction, numbers, and distribution of the gulf sturgeon.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the gulf sturgeon include water of sufficient quality for species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience individual and population growth. Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to these levels. This approval could adversely affect the quality of water to the degree that it would impair individual reproduction and survival of gulf sturgeon, and cause sturgeon to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high 48% and the reduction in the survival of young fish through the first year as high as 56%. These effects are estimated to be of a magnitude great enough to reduce numbers of gulf sturgeon. Approval of the CCC would adversely affect the quality of water to the degree that normal population

growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the gulf sturgeon.

KOOTENAI RIVER WHITE STURGEON

Acipenser transmontanus

Kootenai sturgeon exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience adverse effects on growth, swimming performance, condition, and development, as described above in the *Overview* section. The effects of chronic toxicity could also be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies are available for estimating the magnitude of adverse effects that could occur following Kootenai River white sturgeon exposure to cyanide at criterion concentrations. Because no data for cyanide toxicity to sturgeon exist, LC₅₀ values for sturgeon were derived from the 5% SSD concentration for the class Actinopterygii, which encompasses all known cyanide toxicity data for fish. From these data, we developed quantitative estimates of the effects on sturgeon fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate that Kootenai River white sturgeon that are exposed to cyanide at the CCC are likely to be subject to an approximately 48% reduction in the number of hatched eggs. We estimate that Kootenai River white sturgeon exposed to cyanide at the CCC are likely to experience an approximately 56% reduction in the survival of young fish through the first year.

Although no cyanide-specific data exist for this species, there are data from other chemicals that support the relative sensitivity of sturgeon to contaminants. Dwyer et al. (2005) tested the relative sensitivity of 18 fish and 1 amphibian species to five chemicals. Of these, the two sturgeon tested, the Atlantic sturgeon (*A. oxyrinchus*) and the shortnose sturgeon (*A. brevirostrum*) ranked first and second, respectively, in overall sensitivity. For all five chemicals, sturgeon were as or more sensitive than rainbow trout, for which reductions in viable eggs spawned and juvenile survival were estimated at 52% and 61%, respectively (Table 12).

As noted previously, the young first-year fish that do survive are likely to experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a

whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation).

Sturgeon have naturally high adult survival. Several authors have suggested that the rate of survival may be so high that management at the levels of these age classes is unlikely to improve their survival or increase the population growth rate (Heppell 2007, Gross et al. 2002). For that reason, recovery efforts are often based upon increasing survival in juvenile age classes. *The Recovery Plan for the Kootenai River White Sturgeon* lists as a Priority 1 task (tasks that must be taken to prevent extinction or to prevent the species from declining irreversibly in the foreseeable future) restoration of ecosystem functions “to ensure habitat conditions necessary for successful white sturgeon reproduction and recruitment, i.e. survival of juveniles during their first year of life and beyond” (Service 1999). The reestablishment of natural recruitment to the Kootenai River population of the white sturgeon is listed in the recovery plan as a recovery objective.

Gross et al. (2002) modeled population growth rates for three species of sturgeon that varied in life history traits such as size, lifespan, age to maturity, and migration. All three sturgeon species showed similar elasticity profiles, and thus the authors concluded that general interpretation could be applied to sturgeon across species. In contrast to other elasticity profiles for long-lived species, elasticity in sturgeon was highest in individual young-of-the-year and juvenile age classes, dropped at the onset of maturity, and continued to decline for each successive adult age class. Fecundity had relatively low elasticity, as the effects of changes in fecundity are shared among all adult age classes of these long-lived species, and the value of changes to egg numbers is lessened by the high mortality of the young-of-the-year age class. The authors concluded that population growth rate will show little response to improvements in fecundity, but greater responses in survival at either the young-of-the-year or juvenile age classes. However, since survival of the juvenile and adult age classes is naturally high, improvements at these stages will have smaller effects to improving population growth rate than increases to survival of young-of-the-year, when natural mortality is greater. The authors note that among biologists and managers involved in sturgeon conservation, habitat improvement was regarded as the most important conservation undertaking for sturgeon. Results from this study indicate that restoration efforts should target the survival of age classes with high elasticity: the young-of-the-year and juveniles.

Paragamian and Hansen (2008) drew similar conclusions in modeling effects on population growth of the Kootenai River white sturgeon. They reported that subadult and adult survival (>90%) was much higher than that of juveniles (40% in the first year), and recovery was most dependant on increasing first-year survival. Paragamian and Hansen (2008) suggested that to have the largest effect on recovery, the recruitment rate should increase.

In summary, exposure to cyanide concentrations at the chronic criterion are likely to substantially reduce Kootenai River white sturgeon reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survival of young fish through the first year. Such exposure may also subject Kootenai

River white sturgeon to adverse effects on growth, swimming performance, condition, and development. Although sturgeon have developed a life history that allows them to cope with low survivorship to maturity and occasional hits to recruitment, these adaptations are not sufficient to cope with a constant reduction in both fecundity and early life stage survival likely to be caused by exposure to cyanide concentrations at the chronic criterion. The estimated reductions in the survival of young fish through the first year in particular would significantly decrease recovery of this species in the wild. Because of the high magnitude of adverse effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed by sturgeon exposure to cyanide concentrations at the chronic criterion, which is likely to cause a consequent reduction in the numbers of Kootenai River white sturgeon. Based upon the magnitude of adverse effects that are likely to occur and given the extremely endangered status of this species, we conclude that ultimately Kootenai River white sturgeon are likely to be extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion throughout the range of this species is likely to reduce the reproduction, numbers, and distribution of the Kootenai River white sturgeon at the rangewide scale.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the Kootenai River white sturgeon include water of sufficient quality for the species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience individual and population growth. Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to these levels. This approval and cyanide in waters to these levels is likely to adversely affect the quality of water in sturgeon critical habitat to the degree that it would impair individual reproduction and survival of Kootenai River white sturgeon, and cause sturgeon to experience adverse effects to growth, swimming performance, condition, and development. Cyanide concentrations at the chronic criterion within critical habitat are likely to create habitat conditions that reduce the number of hatched sturgeon eggs by as much as 48% and reduce the survival of young sturgeon through the first year by as much as 56%. Approval of the CCC and cyanide in waters to these levels is likely to adversely affect the quality of water in sturgeon critical habitat to the degree that it is likely to preclude the intended conservation function of that habitat.

PALLID STURGEON
Scaphirhynchus albus

Pallid sturgeon exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. Because no data for

cyanide toxicity to sturgeon exist, LC50 values for sturgeon were derived from the 5% SSD concentration for the class Actinopterygii, which encompasses all known cyanide toxicity data for fish. From this data, we developed quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13).

Compared to control populations, we estimate pallid sturgeon exposed to cyanide at the CCC could experience an approximate 48% reduction in the number of hatched eggs. We estimate that pallid sturgeon exposed to cyanide at the CCC could experience an approximate 56% reduction in survival of young fish through the first year. Though no cyanide-specific data exist for this species, there are data from other chemicals that support the relative sensitivity of sturgeon to contaminants. Dwyer et al. (2005) tested the relative sensitivity of 18 fish and 1 amphibian species to five chemicals. Of these, the two sturgeon tested, the Atlantic sturgeon (*A. oxyrinchus*) and the shortnose sturgeon (*A. brevirostrum*) ranked first and second, respectively, in overall sensitivity. For all five chemicals, sturgeon were as or more sensitive than rainbow trout, for which reductions in viable eggs spawned and juvenile survival were estimated at 52% and 61%, respectively (Table 12).

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation).

Sturgeon have naturally high adult survival. Several authors have suggested that the rate of survival may be so high that management at the levels of these age classes is unlikely to improve their survival or increase population growth rate (Heppell 2007, Gross et al 2002). As such, recovery efforts are often based upon increasing survival in juvenile age classes. The 2007 Five-Year Review for the pallid sturgeon found that natural recruitment is limited throughout the species' range (USFWS 2007). Wild populations in two of six Recovery Priority Management Areas (RPMAs) are comprised of old aged individuals, and three of six RPMAs are dependent on hatchery augmentation programs for recruitment. Addressing recruitment bottlenecks in the three upper Missouri River RPMAs was deemed critically important for the species to become self sustaining and be recovered in those reaches.

Gross et al (2002) modeled population growth rates for three species of sturgeon that varied in life history traits such as size, lifespan, age to maturity, and migration. All three sturgeons showed similar elasticity profiles, and thus the authors concluded that general interpretation could be applied to sturgeon across species. In contrast to other elasticity profiles for long-lived species, elasticity in sturgeon was highest in individual young-of-the-year and juvenile age classes, dropped at the onset of maturity, and continued to

decline for each successive adult age class. Fecundity had relatively low elasticity, as the effects of changes in fecundity are shared among all adult age classes of these long-lived species, and the value of changes to egg numbers is lessened by the high mortality of the young-of-the-year age class. The authors concluded that population growth rate will show little response to improvements in fecundity, but greater responses in survival at either the young-of-the-year or juvenile age classes. However, since survival of the juvenile and adult age classes is naturally high, improvements at these stages will have smaller effects to improving population growth rate than increases to survival of young-of-the-year, when natural mortality is greater. The authors note that among biologists and managers involved in sturgeon conservation, habitat improvement was regarded as the most important conservation undertaking for sturgeon. Results from this study indicate that restoration efforts should target the survival of age classes with high elasticity, young-of-year and juvenile.

Paragamian and Hansen (2008) drew similar conclusions in modeling effects on population growth of the Kootenai River white sturgeon. The authors found that subadult and adult survival (>90%) was much higher than that of juveniles (40% in the first year), and recovery was most dependant on increasing first-year survival. The authors suggested that to have the largest effect on recovery, the managers should increase the current targeted recruitment rate.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the pallid sturgeon reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survival of young fish through the first year. Pallid sturgeon may also experience effects on growth, swimming performance, condition, and development. While sturgeon have developed a life history that allows them to cope with low survivorship to maturity and occasional hits to recruitment, these adaptation are not designed to withstand a constant reduction in both fecundity and early life stage survival. The reductions we estimate in survival of young fish through the first year in particular would significantly decrease recovery of this species. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. We would anticipate a consequent reduction in numbers of pallid sturgeon. An effected Pallid sturgeon population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Pallid sturgeon are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion likely reduces the reproduction, numbers, and distribution of the pallid sturgeon.

ALABAMA STURGEON
Scaphirhynchus suttkusi

Alabama sturgeon exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects

Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. Because no data for cyanide toxicity to sturgeon exist, LC50 values for sturgeon were derived from the 5% SSD concentration for the class Actinopterygii, which encompasses all known cyanide toxicity data for fish. From this data, we developed quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Alabama sturgeon exposed to cyanide at the CCC could experience an approximate 48% reduction in the number of hatched eggs. We estimate that Alabama sturgeon exposed to cyanide at the CCC could experience an approximate 56% reduction in survival of young fish through the first year. Though no cyanide-specific data exist for this species, there are data from other chemicals that support the relative sensitivity of sturgeon to contaminants. Dwyer et al. (2005) tested the relative sensitivity of 18 fish and 1 amphibian species to five chemicals. Of these, the two sturgeon tested, the Atlantic sturgeon (*A. oxyrinchus*) and the shortnose sturgeon (*A. brevirostrum*) ranked first and second, respectively, in overall sensitivity. For all five chemicals, sturgeon were as or more sensitive than rainbow trout, for which reductions in viable eggs spawned and juvenile survival were estimated at 52% and 61%, respectively (Table 12).

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation).

Sturgeon have naturally high adult survival. Several authors have suggested that the rate of survival may be so high that management at the levels of these age classes is unlikely to improve their survival or increase population growth rate (Heppell 2007, Gross et al 2002). As such, recovery efforts are often based upon increasing survival in juvenile age classes. The May 5, 2000 listing rule for the Alabama sturgeon (65 FR 26438) designated the primary threat to the immediate survival of the species to be its small population size and apparent inability to offset mortality rates with current reproduction and/or recruitment rates. Its small population also makes it vulnerable to natural or human-induced events (*e.g.*, droughts, floods, competition, variations in prey abundance, toxic spills), which may further depress recruitment.

Gross et al (2002) modeled population growth rates for three species of sturgeon that varied in life history traits such as size, lifespan, age to maturity, and migration. All three sturgeons showed similar elasticity profiles, and thus the authors concluded that general interpretation could be applied to sturgeon across species. In contrast to other elasticity profiles for long-lived species, elasticity in sturgeon was highest in individual young-of-the-year and juvenile age classes, dropped at the onset of maturity, and continued to decline for each successive adult age class. Fecundity had relatively low elasticity, as the effects of changes in fecundity are shared among all adult age classes of these long-lived species, and the value of changes to egg numbers is lessened by the high mortality of the young-of-the-year age class. The authors concluded that population growth rate will show little response to improvements in fecundity, but greater responses in survival at either the young-of-the-year or juvenile age classes. However, since survival of the juvenile and adult age classes is naturally high, improvements at these stages will have smaller effects to improving population growth rate than increases to survival of young-of-the-year, when natural mortality is greater. The authors note that among biologists and managers involved in sturgeon conservation, habitat improvement was regarded as the most important conservation undertaking for sturgeon. Results from this study indicate that restoration efforts should target the survival of age classes with high elasticity, young-of-year and juvenile.

Paragamian and Hansen (2008) drew similar conclusions in modeling effects on population growth of the Kootenai River white sturgeon. The authors found that subadult and adult survival (>90%) was much higher than that of juveniles (40% in the first year), and recovery was most dependant on increasing first-year survival. The authors suggested that to have the largest effect on recovery, the managers should increase the current targeted recruitment rate.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Alabama sturgeon reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survival of young fish through the first year. Alabama sturgeon may also experience effects on growth, swimming performance, condition, and development. While sturgeon have developed a life history that allows them to cope with low survivorship to maturity and occasional hits to recruitment, these adaptation are not designed to withstand a constant reduction in both fecundity and early life stage survival. The reductions we estimate in survival of young fish through the first year in particular would significantly decrease recovery of this species. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. We would anticipate a consequent reduction in numbers of Alabama sturgeon. An effected Alabama sturgeon population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Alabama sturgeon are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion likely reduces the reproduction, numbers, and distribution of the Alabama sturgeon.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the Alabama sturgeon include a flow regime and river system which allows for all life stages and processes of the species, a river channel with a stable sand and gravel bottom, as well as a rock wall and associated mussel beds, limestone outcrops and cut limestone banks, riverline spawning sites with substance suitable for embryo deposition and development, large sections of free flowing water for spawning migrations and the development of young, water of 32 degrees Celsius (90 degrees Fahrenheit) or less, dissolved oxygen at 5 milligrams per liter or more, and pH from 6.0 to 8.5.

Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect Alabama sturgeon critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of Alabama sturgeon, and cause Alabama sturgeon to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 48% and the reduction in the survival of young fish through the first year as high as 56%. These effects are estimated to be of a magnitude great enough to reduce numbers of Alabama sturgeon. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the Alabama sturgeon's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the Alabama sturgeon.

Amblyopsidae

OZARK CAVEFISH

Amblyopsis rosae

Ozark cavefish exposed to cyanide at the criterion continuous concentration (CCC) are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Ozark cavefish exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 48%. We estimate that Ozark cavefish exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 56%.

As noted previously, the first-year fish that do survive could experience reduced growth rates that would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, such as Ozark cavefish, than on species with greater adult survival. Ozark cavefish can live up to 7-10 years, but do not reach adulthood until 4 or more years of age. Furthermore, only about 20% of the female population breeds each year, producing 20-25 eggs. We anticipate the effects of cyanide on fecundity and juvenile survival could have a substantial population-level effect on Ozark cavefish. Individual Ozark cavefish females reproduce infrequently and produce few offspring. A reduction in potential fecundity and juvenile survival in the wild at the magnitude we estimate will add to the challenges to recovering the Ozark cavefish.

The reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the Ozark cavefish's potential recruitment substantially. The Ozark cavefish's potential recruitment would be diminished because of (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction and survival could be exacerbated further if reduced growth rates diminish survival through to adulthood.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Ozark cavefish's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Ozark cavefish may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. These potential effect could add substantially to the difficulties in reintroducing reproductively successful populations and attaining recovery. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Ozark cavefish are not likely to overcome the effects to their recruitment in waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could limit the reproduction, numbers, and distribution of the Ozark cavefish .

ALABAMA CAVEFISH

Speoplatyrhinus poulsoni

Alabama cavefish exposed to cyanide at the criterion continuous concentration (CCC) are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Alabama cavefish exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 48%. We estimate that Alabama cavefish exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 56%.

As noted previously, the first-year fish that do survive could experience reduced growth rates that would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Alabama cavefish may live 5-10 years. Cavefish do not reproduce every year, numbers of reproductive females are few, and those that do spawn lay very few eggs. We anticipate the potential effects of cyanide on fecundity and juvenile survival would have a substantial population-level effect on Alabama cavefish. A reduction in potential fecundity and juvenile survival in the wild at the magnitude we estimate will add to the significant challenges to recovering the Alabama cavefish.

The reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the Alabama cavefish's potential recruitment substantially. The Alabama cavefish's potential recruitment would be diminished because of (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent

individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates diminish survival beyond the early life stages analyzed.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Alabama cavefish's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Alabama cavefish may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be inadequate to avoid population decline. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Alabama cavefish are not likely to overcome the effects to their recruitment where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could limit the reproduction, numbers, and distribution of the Alabama cavefish.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the Alabama cavefish include water of sufficient quality for species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience individual and population growth. Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to these levels. This approval could adversely affect the quality of water to the degree that it would impair individual reproduction and survival of Alabama cavefish, and cause fish to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high 48% and the reduction in the survival of young fish as high as 56%. These effects are estimated to be of a magnitude great enough to reduce numbers of Alabama cavefish. Approval of the CCC would adversely affect the quality of water to the degree that normal individual and population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the Alabama cavefish.

Atherinidae

WACCAMAW SILVERSIDE

Menidia extensa

Waccamaw silverside exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were

available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Waccamaw silversides exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 48%. We estimate that Waccamaw silversides exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 56%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than species with greater adult survival. The Waccamaw silverside has a 1-year life cycle and depends upon successful reproduction each year for its survival. Almost all adults die soon after spawning. A comparison of 88 freshwater fish species, found that longevity and age at maturity were the best predictors of elasticity patterns (Vélez-Espino et al., 2006). Elasticity analyses performed on freshwater fish with similar longevity and reproductive lifespans to the Waccamaw silverside found that population growth rates for these species were highly susceptible to perturbations in juvenile survival and fecundity (Vélez-Espino et al., 2006). In combination, these two stages accounted for over 90% of the total elasticity of the population growth rate in all species with these life-history traits. These reproductive parameters, as defined in our analysis, include both egg production and larval / juvenile survival rate to the next year's spawn.

We anticipate the Waccamaw silverside's reproductive performance would be reduced substantially. The Waccamaw silverside's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Waccamaw silverside's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Waccamaw silversides may also experience

effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Waccamaw silverside population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Waccamaw silversides are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Waccamaw silverside.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the Waccamaw silverside include high quality water which is clear, open, and has a neutral pH, and a clean sand substrate. Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect Waccamaw silverside critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of Waccamaw silversides, and cause Waccamaw silversides to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 48% and the reduction in the survival of young fish through the first year as high as 56%. These effects are estimated to be of a magnitude great enough to reduce numbers of Waccamaw silverside. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the Waccamaw silverside's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the Waccamaw silverside.

Catostomidae

MODOC SUCKER

Catostomus microps

Modoc suckers exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Modoc suckers exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is

not likely to be greater than, 39%. We estimate that Modoc suckers exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 44%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Modoc suckers typically lives up to 5 years, achieving sexual maturation in its third year, resulting in a reproductive life span of only one or two years. A comparison of 88 freshwater fish species found that longevity and age at maturity were the best predictors of elasticity patterns (Vélez-Espino et al., 2006). Elasticity analyses performed on suckers with similar life-history traits to the Modoc sucker found population growth rate to be highly susceptible to perturbations in juvenile survival, accounting for about 60% of the total elasticity (Vélez-Espino et al., 2006). In combination, juvenile survival and fecundity in these species accounted for about 80% of the total elasticity of the population growth rate. These reproductive parameters, as defined in our analysis, include both egg production and larval / juvenile survival rate to the next year's spawn.

We anticipate the Modoc sucker's reproductive performance would be reduced substantially. The reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Modoc sucker's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Modoc suckers may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Modoc sucker's population's decline could stabilize at a reduced absolute population number or could continue to decline until it is

extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Modoc suckers are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Modoc sucker.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the Modoc sucker include intermittent and permanent-water creeks, and adjacent land areas that provide vegetation for cover and protection from soil erosion. Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect Modoc sucker critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of Modoc suckers, and cause Modoc sucker to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 39% and the reduction in the survival of young fish through the first year as high as 344%. These effects are estimated to be of a magnitude great enough to reduce numbers of Modoc sucker. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the Modoc sucker's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the Modoc sucker.

SANTA ANA SUCKER

Catostomus santaanae

Santa Ana suckers exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Santa Ana suckers to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 39%. We estimate that Santa Ana suckers exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 44%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival, such as sturgeon. Santa Ana sucker females are short-lived (up to 3 years), mature early, and may have prolonged spawning periods. Fecundity is considered to be exceptionally high for a species of its small size. In a comparison of 88 freshwater fish species, longevity and age at maturity were found to be the best predictors of elasticity patterns (Vélez-Espino et al., 2006). Elasticity analyses performed on freshwater fish with similar longevity, age at maturity, and reproductive lifespan as the Santa Ana sucker found that population growth rates for these species were highly susceptible to perturbations in juvenile survival and fecundity (Vélez-Espino et al., 2006). In combination, these two stages accounted for over 90% of the total elasticity of the population growth rate in all species with these life-history traits. These reproductive parameters, as defined in our analysis, include both egg production and larval / juvenile survival rate to the next year's spawn

We anticipate the Santa Ana sucker's reproductive performance would be reduced substantially. The Santa Ana sucker's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Santa Ana sucker's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Santa Ana suckers may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Santa Ana sucker population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Santa Ana suckers are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion

concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Santa Ana sucker.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the Santa Ana sucker include the following: A functioning hydrological system that experiences peaks and ebbs in the water volume that reflects seasonal variation in precipitation throughout the year; A mosaic of loose sand, gravel, cobble, and boulder substrates in a series of riffles, runs, pools, and shallow sandy stream margins; Water depths greater than 3 cm (1.2 in) and bottom water velocities greater than 0.03 meter per second (0.01 feet per second); Non-turbid water or only seasonally turbid water; Water temperatures less than 30 [deg]C (86 [deg]F); and stream habitat that includes algae, aquatic emergent vegetation, macroinvertebrates, and riparian vegetation.

Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect Santa Ana sucker critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of Santa Ana suckers, and cause Santa Ana suckers to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 39% and the reduction in the survival of young fish through the first year as high as 44%. These effects are estimated to be of a magnitude great enough to reduce numbers of Santa Ana sucker. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the Santa Ana sucker's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the Santa Ana sucker.

WARNER SUCKER

Catostomus warnerensis

Warner suckers exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Warner suckers exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 39%. We estimate that Warner suckers exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through

the first year and that reduction could be as much as, but is not likely to be greater than, 44%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). Warner suckers have been documented to live up to 17 years, though individuals residing in lakes are presumed to live longer than those found in rivers. Sexual maturity occurs at an age of 3 to 4 years. Spawning usually occurs in April and May in streams, in silt-free, gravel-bottomed, slow flowing sections of creeks. In years when access to stream spawning is limited by low flow or by physical in-stream blockages (such as beaver dams or diversion structures), suckers may attempt to spawn on gravel beds along the lake shorelines.

For species like the Warner sucker that exhibit intermediate longevity, moderately delayed reproduction, and multiple opportunities to spawn, population growth rate tends to be most sensitive to perturbation in juvenile survival (Vélez-Espino et al 2006). Elasticity analyses performed on suckers within the genus *Catostomus* that exhibit similar life-history traits to the Warner sucker found this pattern to hold true, with juvenile survival accounting for 50-60% of the total elasticity (Vélez-Espino et al., 2006). In combination, juvenile survival and fecundity accounted for 70 to 80% of the total elasticity of the population growth rate. A comparison of elasticities among closely related species found that elasticity values are highly conserved among genera within the same taxonomic family (Vélez-Espino et al., 2006). These reproductive parameters, as defined in our analysis, include both egg production and larval / juvenile survival rate to the next year's spawn.

We anticipate the Warner sucker's reproductive performance would be reduced substantially. The Warner sucker's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Warner sucker's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Warner suckers may also experience effects

on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Warner sucker population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Warner suckers are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Warner sucker.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the Warner sucker include the following: Streams should have clean, unpolluted flowing water and a stable riparian zone. The streams should support a variety of aquatic insects, crustaceans, and other small invertebrates for food.

Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect Warner sucker critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of Warner suckers, and cause Warner suckers to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 39% and the reduction in the survival of young fish through the first year as high as 44%. These effects are estimated to be of a magnitude great enough to reduce numbers of Warner sucker. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the Warner sucker's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the Warner sucker.

SHORTNOSE SUCKER

Chasmistes brevirostris

Shortnose sucker exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate shortnose suckers exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is

not likely to be greater than, 39%. We estimate that shortnose suckers exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 44%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). The shortnose sucker can live more than 30 years, though a 2007 study from the Upper Klamath Lake indicated that the average life expectancy after entering the spawning population was only 3.6 years, suggesting that in some populations, adults may be dying before reproducing often enough for population replacement. Sexual maturity for shortnose suckers appears to occur between the ages of 4 and 6 years. Freshwater fish species with longevity and reproductive lifespan characteristics similar to those of the shortnose sucker were found to have population growth rates that were particularly susceptible to perturbations in juvenile survival, accounting for 40% to 50% of the total elasticity (Vélez-Espino et al., 2006). Recruitment for this species is historically low and continues to be below levels to sustain population growth. The 2007 5-Year Review for the shortnose sucker identified recruitment of young fish to the breeding population as a high priority goal for recovery.

We anticipate the shortnose sucker's reproductive performance and recruitment would be reduced substantially. The shortnose sucker's reproductive performance rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the shortnose sucker's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Shortnose suckers may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected shortnose sucker population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could

occur, we conclude ultimately shortnose suckers are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the shortnose sucker.

CUI-UI

Chasmistes cujus

Cui-ui exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate cui-uies exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 39%. We estimate that cui-uies exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 44%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). The cui-ui can live more than 40 years, reaching sexual maturity in 6 to 12 years. Females can produce from 25,000 to 186,000 eggs and larval survival is presumed to be extremely small. Freshwater fish species with longevity and reproductive lifespan characteristics similar to those of the Cui-ui were found to have population growth rates that were particularly susceptible to perturbations in juvenile survival, accounting for 40% to 50% of the total elasticity (Vélez-Espino et al., 2006). The Recovery Plan for the cui-ui stresses the need for increased survival, and consequent recruitment, of young.

We anticipate the cui-ui's reproductive performance and recruitment would be reduced substantially. The cui-ui's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the

first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the cui-ui's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Cui-uies may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected cui-ui population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately cui-uies are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the cui-ui.

JUNE SUCKER

Chasmistes lioris

June suckers exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate June suckers exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 39%. We estimate that June suckers exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 44%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). Freshwater fish species with longevity and reproductive lifespan characteristics similar to those of the June sucker were found to have population growth rates that were particularly susceptible to perturbations in juvenile survival, accounting for 40% to 50% of the total elasticity (Vélez-Espino et al., 2006). June suckers can live to be over 40 years old and reach sexual maturity at age 9. Survival rates for Red Butte refuge population were estimated as follows: age 1, 0.4225; age 2, 0.4625; age 3 0.8020, and adults, 0.9576 (Billman and Crowl, 2007). However, the wild population endemic to Utah Lake has continued to experience wide recruitment failure leaving it dominated by adult fish (Billman and Crowl, 2007). The June sucker Recovery Plan identifies the minimization of factors limiting recruitment as a priority in the recovery of the species.

We anticipate the June sucker's reproductive performance and subsequent recruitment would be further reduced. The June sucker's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the June sucker's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. June suckers may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected June sucker population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately June suckers are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the June sucker.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the June sucker include the following: One to three feet of high quality water constantly flowing over a clean, unsilted gravel substrate. Larval June suckers require shallow areas with low velocities connected to the main channel of the river.

Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This

approval could adversely affect June sucker critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of June suckers, and cause June suckers to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 39% and the reduction in the survival of young fish through the first year as high as 44%. These effects are estimated to be of a magnitude great enough to reduce numbers of June sucker. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the June sucker's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the June sucker.

LOST RIVER SUCKER

Deltistes luxatus

Lost River sucker exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Lost River suckers exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 39%. We estimate that Lost River suckers exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 44%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). The Lost River sucker can live more than 40 years, though a 2007 study from the Upper Klamath Lake indicated that the average life expectancy after entering the spawning population was only 9 years,

suggesting that in some populations, adults may be dying before reproducing often enough for population replacement. Sexual maturity for Lost River suckers occurs between the ages of 6 to 14 years, with most maturing at age 9. Freshwater fish species with longevity and reproductive lifespan characteristics similar to those of the Lost River sucker were found to have population growth rates that were particularly susceptible to perturbations in juvenile survival, accounting for 40% to 50% of the total elasticity (Vélez-Espino et al., 2006). A lack of recruitment has been reported for the river spawning population of the Lost River sucker, and the 2007 5-Year Review identified recruitment of young fish to the breeding population as a whole as a high priority goal for recovery due to large-scale die-offs of adults in the mid-1990's.

We anticipate the Lost River sucker's reproductive performance and recruitment would be reduced substantially. The Lost River sucker's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Lost River sucker's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Lost River suckers may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An affected Lost River sucker population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Lost River suckers are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Lost River sucker.

RAZORBACK SUCKER

Xyrauchen texanus

Razorback suckers exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were

available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate razorback suckers exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 39%. We estimate that razorback suckers exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 44%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). The razorback sucker can live more than 40 years, and begin spawning at 3 to 4 years of age. Freshwater fish species with longevity and reproductive lifespan characteristics similar to those of the razorback sucker were found to have population growth rates that were particularly susceptible to perturbations in juvenile survival, accounting for 40% to 50% of the total elasticity (Vélez-Espino et al., 2006). Though the razorback sucker is a long-lived species, current populations have continued to experience persistent recruitment failure, resulting in the depletion and extirpation of numerous populations. Although razorback sucker in certain locations number in the thousands, low reproductive success, low survival of young, and little or no recruitment have contributed to high demographic uncertainty. Wild razorback sucker populations in many locations of the Colorado River Basin have become aged, senile, and perished from inadequate recruitment.

We anticipate the razorback sucker's reproductive performance and subsequent recruitment would be further reduced substantially. The razorback sucker's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the razorback sucker's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Razorback suckers may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory

mechanisms, if they exist, to be overwhelmed. An effected razorback sucker population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately razorback suckers are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the razorback sucker.

Cotidae

PYGMY SCULPIN

Cottus paulus

Pygmy sculpins exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate pygmy sculpins exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 48%. We estimate that pygmy sculpins exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 56%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Pygmy sculpin females are probably short-lived (1-3 years), mature early, and spawn year-round with a peaks in August and late winter. A comparison of 88 freshwater fish species found that longevity and age at maturity were

the best predictors of elasticity patterns (Vélez-Espino et al., 2006). Elasticity analyses performed on cyprinids with similar longevity, age at maturity, and reproductive lifespan as the pygmy sculpin found that population growth rates for these species were highly susceptible to perturbations in juvenile survival and fecundity (Vélez-Espino et al., 2006). In combination, these two stages accounted for over 90% of the total elasticity of the population growth rate in all species with these life-history traits. These reproductive parameters, as defined in our analysis, include both egg production and larval / juvenile survival rate to the next year's spawn.

We anticipate the pygmy sculpin's reproductive performance would be reduced substantially. The pygmy sculpin's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the pygmy sculpin's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Pygmy sculpins may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected pygmy sculpin population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately pygmy sculpins are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the pygmy sculpin.

Cyprinidae

BLUE SHINER

Cyprinella caerulea

Blue shiner exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were

available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate blue shiners exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that blue shiners exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Blue shiner females are short-lived (about 3 years). Most spawning adults are aged 2 years and spawn from early May through late August.

We anticipate the blue shiner's reproductive performance would be reduced substantially. The blue shiner's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the blue shiner's reproductive performance by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Blue shiners may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An affected blue shiner population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately blue shiners are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the blue shiner.

BEAUTIFUL SHINER
Cyprinella formosa

Beautiful shiner exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated beautiful shiner exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that beautiful shiners exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like beautiful shiners, than on species with greater adult survival, such as sturgeon. Beautiful shiner females are short-lived (less than 3 years).

We anticipate the beautiful shiner's fertility would be reduced substantially. The beautiful shiner's fertility rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the beautiful shiner's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Beautiful shiners may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected beautiful shiner population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately beautiful shiners are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the beautiful shiner.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the beautiful shiner include permanently flowing small streams with riffles, or intermittent creeks with pools and riffles in the Rio Yaqui drainage. The waters must be clear and unpolluted, and free of exotic fishes. The beautiful shiner needs water of sufficient quality for the species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience adequate individual and population growth.

Approval of the CCC in State and Tribal water quality standards would authorized States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect beautiful shiner critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of delta smelt, and cause delta smelt to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 31% and the reduction in the survival of young fish through the first year as high as 33%. These effects are estimated to be of a magnitude great enough to reduce numbers of delta smelt. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the beautiful shiner's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the beautiful shiner.

DEVILS RIVER MINNOW

Dionda diaboli

Devils River minnow exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated Devils River minnow exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that Devils River minnows exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Devils River minnow females are short-lived (1-2 years), mature early, and probably spawn from January through August.

The Devils River minnow probably has a reproductive strategy similar to the smalleye shiner (*Notropis buccula*). Durham and Wilde (2009) studied the population dynamics of the smalleye shiner in the Brazos River, Texas. Smalleye shiner are members of a reproductive guild of cyprinids that broadcast spawn multiple batches of nonadhesive, semibuoyant ova throughout an extended reproductive season, and experience extremely high post-spawning mortality. Elasticity analysis and sensitivity simulations of the projection matrix indicated that age-0 survival and age-1 fecundity were the most influential parameters in the population dynamics of smalleye shiners. In combination, these two stages accounted for the majority (70%) of the total elasticity of the population growth rate.

We anticipate the Devils River minnow's reproductive performance would be reduced substantially. The Devils River minnow's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Devils River minnow's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Devils River minnows may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Devils River minnow population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Devils River minnows are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Devils River minnow.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the Devils River minnow include the following: streams with slow to moderate water velocities (10 to 40 cm per second or 4 to 16 inches per second) and shallow to moderate water depths (10 cm to 1.5 m or 4 inches to 4.9 feet). These streams must be near vegetative structure, such as emergent or submerged vegetation or stream bank riparian vegetation that overhangs the water column. Gravel and cobble substrates with low or moderate amounts of fine sediment and low or moderate amounts of substrate embeddedness. Pool, riffle, run, and backwater components free of structures that would prevent fish movement up or downstream. High quality water from groundwater springs and seeps which is: between 17 and 29 degrees Celsius, has dissolved oxygen levels greater than 5 mg/l, a pH between 7 and 8.2, has less than 0.7 mS/cm conductivity and salinity of less than 1 part per thousand, has ammonia levels of less than 0.4 mg/l, and finally has no (or minimal amounts of) pollutants such as copper, arsenic, mercury, cadmium, human and animal waste, pesticides, fertilizers, suspended sediments, petroleum compounds, gasoline, and diesel fuel. Finally, the habitat must possess an adequate algae food base, and no nonnative aquatic species. By high quality water, we mean water of sufficient quality for the species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience individual and population growth sufficient for the critical habitat to serve its intended conservation function.

Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect Devils River minnow critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of Devils River minnows, and cause Devils River minnows to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 31% and the reduction in the survival of young fish through the first year as high as 33%. These effects are estimated to be of a magnitude great enough to reduce numbers of Devils River minnow. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the Devils River minnow's

extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the Devils River minnow.

SPOTFIN CHUB
Cyprinella monarcha

Spotfin chub exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate spotfin chub exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 68%. We estimate that spotfin chub exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 76%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like spotfin chub, than on species with greater adult survival, such as sturgeon. Most spotfin chub start spawning after their second year and may not live beyond the age of three. Counts of mature ova numbered 157-791, but may greatly underestimate fecundity if this species spawns fractionally.

The spotfin chub shares a relatively similar life history with darters. Hartup (2005) conducted field research on the slackwater darter and holiday darter (*E. brevirostrum*) and developed species-specific estimates for fecundity, adult survival, and population sizes, then used these data to conduct population viability analyses for the two species.

Average slackwater darter fecundity was estimated as 92 and 197, respectively, for one-batch and two-batch fecundity. Based on estimates of adult survival, Hartup (2005) calculated the adult fertility rate would need to average 0.896 for the slackwater darter population to be stable and 0.818 for the holiday darter population to be stable.

Assuming the slackwater darter population was stable, Hartup estimated the average proportion of spawned eggs needing to survive through the first year as 0.019 or 0.009, for one- and two-batch fecundity, respectively. For the holiday darter, the two-batch estimate was also 0.009. An elasticity analysis for the slackwater darter and holiday darter identified fertilities (defined as fecundity plus juvenile survival) as having the largest relative contribution to population growth rates for each species, as opposed to adult survival (Hartup 2005).

The reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the spotfin chub's potential recruitment substantially. The spotfin chub's potential recruitment would be diminished because of (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates diminish survival through the first winter. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the spotfin chub's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Spotfin chub may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected spotfin chub population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately spotfin chub are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the spotfin chub.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the spotfin chub include water of sufficient quality for species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience individual and population growth. Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to these levels. This approval could adversely affect the quality of water to the degree that it would impair individual reproduction and survival of spotfin chubs, and cause chubs to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high 68% and the reduction in the

survival of young fish through the first year as high as 76%. These effects are estimated to be of a magnitude great enough to reduce numbers of spotfin chub. Approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the spotfin chub.

SLENDER CHUB

Erimystax cahni

Slender chub exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated slender chub exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that slender chubs exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Slender chub females are short-lived (up to 3 years). A comparison of 88 freshwater fish species, found that longevity and age at maturity were the best predictors of elasticity patterns (Vélez-Espino et al., 2006). Elasticity analyses performed on cyprinids with similar longevity, age at maturity, and reproductive lifespan as the slender chub found that population growth rates for these species were highly

susceptible to perturbations in juvenile survival and fecundity (Vélez-Espino et al., 2006). In combination, these two stages accounted for over 90% of the total elasticity of the population growth rate in all species with these life-history traits. These reproductive parameters, as defined in our analysis, include both egg production and larval / juvenile survival rate to the next year's spawn.

We anticipate the slender chub's reproductive performance would be reduced substantially. The slender chub's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the slender chub's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Slender chubs may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected slender chub population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately slender chubs are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the slender chub.

Critical Habitat: Continued approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect slender chub critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of slender chub, and cause slender chub to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 31% and the reduction in the survival of young fish through the first year as high as 33%. These effects are estimated to be of a magnitude great enough to reduce numbers of slender chub. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the slender chub's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the slender chub.

MOJAVE TUI CHUB
Gila bicolor mohavensis

Mojave tui chub exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated Mojave tui chub exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that Mojave tui chubs exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Mojave tui chub females are short-lived (1-4 years) and highly fecund. A comparison of 88 freshwater fish species, found that longevity and age at maturity were the best predictors of elasticity patterns (Vélez-Espino et al., 2006). Elasticity analyses performed on cyprinids with similar longevity, age at maturity, and reproductive lifespan as the slender chub found that population growth rates for these species were highly susceptible to perturbations in juvenile survival and fecundity (Vélez-Espino et al., 2006). In combination, these two stages accounted for over 90% of the total elasticity of the population growth rate in all species with these life-history traits. These reproductive parameters, as defined in our analysis, include both egg production and larval / juvenile survival rate to the next year's spawn.

We anticipate the Mojave tui chub's reproductive performance would be reduced substantially. The Mojave tui chub's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The

combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Mojave tui chub's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Mojave tui chubs may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Mojave tui chub population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Mojave tui chubs are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Mojave tui chub.

OWENS TUI CHUB

Gila bicolor snyderi

Owens tui chub exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated Owens tui chub exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that Owens tui chubs exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. A comparison of 88 freshwater fish species, found that longevity and age at maturity were the best predictors of elasticity patterns (Vélez-Espino et al., 2006). Elasticity analyses performed on cyprinids with similar longevity, age at maturity, and reproductive lifespan as the Owens tui chub found that population growth rates for these species were most susceptible to perturbations in juvenile survival (Vélez-Espino et al., 2006). Juvenile survival accounted for about 58% of the total elasticity of the population growth rate in all species with these life-history traits. A comparison of elasticities among closely related species found that elasticity values are highly conserved among genera within the same taxonomic family. Juvenile survival accounted for about 59% of the total elasticity of the population growth rate for the Utah chub (*Gila atraria*).

We anticipate the Owens tui chub's reproductive performance would be reduced substantially. The Owens tui chub's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Owens tui chub's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Owens tui chubs may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Owens tui chub population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Owens tui chubs are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Owens tui chub.

Critical Habitat: Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect Owens tui chub critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of Owens tui chub, and cause Owens tui chub to experience

adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 31% and the reduction in the survival of young fish through the first year as high as 33%. These effects are estimated to be of a magnitude great enough to reduce numbers of Owens tui chub. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the Owens tui chub's extirpation from critical habitat containing cyanide at the CCC.

Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the Owens tui chub.

BORAX LAKE CHUB

Gila boraxobius.

Borax Lake chub exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated Borax Lake chub exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that Borax Lake chubs exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Borax Lake chub females are short-lived (mostly 1-2 years), mature early, and probably spawn twice per year. A comparison of 88 freshwater fish

species, found that longevity and age at maturity were the best predictors of elasticity patterns (Vélez-Espino et al., 2006). Elasticity analyses performed on cyprinids with similar longevity, age at maturity, and reproductive lifespan as the Moapa dace found that population growth rates for these species were highly susceptible to perturbations in juvenile survival and fecundity (Vélez-Espino et al., 2006). In combination, these two stages accounted for over 90% of the total elasticity of the population growth rate in all species with these life-history traits. These reproductive parameters, as defined in our analysis, include both egg production and larval / juvenile survival rate to the next year's spawn. A comparison of elasticities among closely related species found that elasticity values are highly conserved among genera within the same taxonomic family. Juvenile survival accounted for about 59% of the total elasticity of the population growth rate for the Utah chub (*Gila atraria*).

We anticipate the Borax Lake chub's reproductive performance would be reduced substantially. The Borax Lake chub's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Borax Lake chub's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Borax Lake chubs may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Borax Lake chub population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Borax Lake chubs are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Borax Lake chub.

Critical Habitat: Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect Borax Lake chub critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of Borax Lake chub, and cause Borax Lake chub to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 31% and the reduction in the survival of young fish through the first year as high as 33%. These effects are estimated to be of a magnitude great enough to reduce numbers of Borax Lake chub. Continued approval of the CCC could adversely affect the quality of water to the

degree that normal population growth is likely to be impacted and could result in the Borax Lake chub's extirpation from critical habitat containing cyanide at the CCC.

Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the Borax Lake chub.

HUMPBACK CHUB

Gila boraxobius.

Humpback chub exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated humpback chub exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that humpback chubs exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Little information is available for humpback chub. A comparison of 88 freshwater fish species, found that elasticities among closely related species are highly conserved among genera within the same taxonomic family (Vélez-Espino et al., 2006). Juvenile survival accounted for about 59% of the total elasticity of the population growth rate for the Utah chub (*Gila atraria*) and fecundity accounted for another 19% (Vélez-Espino et al., 2006).

We anticipate the humpback chub's reproductive performance would be reduced substantially. The humpback chub's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the humpback chub's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Humpback chubs may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected humpback chub population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately humpback chubs are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the humpback chub.

Critical Habitat: Continued approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect humpback chub critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of humpback chub, and cause humpback chub to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 31% and the reduction in the survival of young fish through the first year as high as 33%. These effects are estimated to be of a magnitude great enough to reduce numbers of humpback chub. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the humpback chub's extirpation from critical habitat containing cyanide at the CCC.

Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the humpback chub.

SONORA CHUB

Gila ditaenia.

Sonora chub exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as

dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated Sonora chub exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that Sonora chubs exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Little information is available for Sonora chub. A comparison of 88 freshwater fish species, found that elasticities among closely related species are highly conserved among genera within the same taxonomic family (Vélez-Espino et al., 2006). Juvenile survival accounted for about 59% of the total elasticity of the population growth rate for the Utah chub (*Gila atraria*) and fecundity accounted for another 19% (Vélez-Espino et al., 2006).

We anticipate the Sonora chub's reproductive performance would be reduced substantially. The Sonora chub's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Sonora chub's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Sonora chubs may also experience effects

on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Sonora chub population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Sonora chubs are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Sonora chub.

Critical Habitat: Continued approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect Sonora chub critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of Sonora chub, and cause Sonora chub to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 31% and the reduction in the survival of young fish through the first year as high as 33%. These effects are estimated to be of a magnitude great enough to reduce numbers of Sonora chub. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the Sonora chub's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the Sonora chub.

BONYTAIL CHUB

Gila elegans

Bonytail chub exposed to cyanide at the criterion continuous concentration (CCC) are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate bonytail chub exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 57%. We estimate that bonytail chub exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 66%.

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As noted previously, the first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival, such as bonytail. Bonytail may live for as many as 50 years and are very fecund. In hatcheries, females produce between 1,000 and 17,000 eggs and the survival rate of juveniles is 17-38%. Despite this reproductive potential, we anticipate the effects of cyanide on fecundity and juvenile survival would have a substantial population-level effect on bonytail. Although bonytail spawn many eggs, they do not guard their eggs and predation by non-native fish in the Colorado River basin is substantial, probably contributing to reproductive failure in the wild. A reduction in potential fecundity and juvenile survival in the wild at the magnitude we estimate will add to the significant challenges to recovering the bonytail.

The reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the bonytail chub's potential recruitment substantially. The bonytail chub's potential recruitment would be diminished because of (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates diminish survival through the first winter.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the bonytail chub's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Bonytail chub may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. These potential effects could add substantially to the difficulties in reintroducing reproductively successful populations and attaining recovery. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately bonytail chub are not likely to overcome the effects to their recruitment in waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could limit the reproduction, numbers, and distribution of the bonytail chub.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the bonytail chub include water of sufficient quality for species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience individual and population growth. Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to these levels. This approval could adversely affect the quality of water to the degree that it would impair individual reproduction and survival of bonytail chubs, and cause chubs to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high 57% and the reduction in the survival of young fish through the first year as high as 66%. These effects are estimated to be of a magnitude great enough to reduce numbers of bonytail chubs. Approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the bonytail chub.

GILA CHUB
Gila intermedia

Gila chub exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated Gila chub exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that Gila chubs exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth

rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Little information is available for Gila chub. A comparison of 88 freshwater fish species, found that elasticities among closely related species are highly conserved among genera within the same taxonomic family (Vélez -Espino et al., 2006). Juvenile survival accounted for about 59% of the total elasticity of the population growth rate for the Utah chub (*Gila atraria*) and fecundity accounted for another 19% (Vélez -Espino et al., 2006).

We anticipate the Gila chub's reproductive performance would be reduced substantially. The Gila chub's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Gila chub's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Gila chubs may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Gila chub population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Gila chubs are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Gila chub.

Critical Habitat: Continued approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect Gila chub critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of Gila chub, and cause Gila chub to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 31% and the reduction in the survival of young fish through the first year as high as 33%. These effects are estimated to be of a magnitude great enough to reduce numbers of Gila chub. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the Gila chub's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting

from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the Gila chub.

YAQUI CHUB

Gila purpurea.

Yaqui chub exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated Yaqui chub exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that Yaqui chubs exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Little information is available for Yaqui chub. A comparison of 88 freshwater fish species, found that elasticities among closely related species are highly conserved among genera within the same taxonomic family (Vélez-Espino et al., 2006). Juvenile survival accounted for about 59% of the total elasticity of the population growth rate for the Utah chub (*Gila atraria*) and fecundity accounted for another 19% (Vélez-Espino et al., 2006).

We anticipate the Yaqui chub's reproductive performance would be reduced substantially. The Yaqui chub's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through

the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Yaqui chub's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Yaqui chubs may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Yaqui chub population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Yaqui chubs are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Yaqui chub.

Critical Habitat: Continued approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect Yaqui chub critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of Yaqui chub, and cause Yaqui chub to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 31% and the reduction in the survival of young fish through the first year as high as 33%. These effects are estimated to be of a magnitude great enough to reduce numbers of Yaqui chub. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the Yaqui chub's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the Yaqui chub.

PAHRANAGAT ROUNDTAIL CHUB

Gila robusta jordani.

Pahranagat roundtail chub exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Formal Draft Biological Opinion.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated Pahrnagat roundtail chub exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that Pahrnagat roundtail chubs exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Little information is available for Pahrnagat roundtail chub. A comparison of 88 freshwater fish species, found that elasticities among closely related species are highly conserved among genera within the same taxonomic family (Vélez-Espino et al., 2006). Juvenile survival accounted for about 59% of the total elasticity of the population growth rate for the Utah chub (*Gila atraria*) and fecundity accounted for another 19% (Vélez-Espino et al., 2006).

We anticipate the Pahrnagat roundtail chub's reproductive performance would be reduced substantially. The Pahrnagat roundtail chub's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Pahrnagat roundtail chub's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Pahrnagat roundtail chubs may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected

Pahranagat roundtail chub population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Pahranagat roundtail chubs are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Pahranagat roundtail chub.

Critical Habitat: Continued approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect Pahranagat roundtail chub critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of Pahranagat roundtail chub, and cause Pahranagat roundtail chub to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 31% and the reduction in the survival of young fish through the first year as high as 33%. These effects are estimated to be of a magnitude great enough to reduce numbers of Pahranagat roundtail chub. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the Pahranagat roundtail chub's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the Pahranagat roundtail chub.

VIRGIN RIVER CHUB

Gila robusta seminuda

Virgin River chub exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated Virgin River chub exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that Virgin River chubs exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Little information is available for Virgin River chub. A comparison of 88 freshwater fish species, found that elasticities among closely related species are highly conserved among genera within the same taxonomic family (Vélez-Espino et al., 2006). Juvenile survival accounted for about 59% of the total elasticity of the population growth rate for the Utah chub (*Gila atraria*) and fecundity accounted for another 19% (Vélez-Espino et al., 2006).

We anticipate the Virgin River chub's reproductive performance would be reduced substantially. The Virgin River chub's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Virgin River chub's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Virgin River chubs may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Virgin River chub population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Virgin River chubs are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Virgin River chub.

Critical Habitat: Continued approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect Virgin River chub critical habitat by diminishing the quality of water to the degree that it would impair individual

reproduction and survival of Virgin River chub, and cause Virgin River chub to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 31% and the reduction in the survival of young fish through the first year as high as 33%. These effects are estimated to be of a magnitude great enough to reduce numbers of Virgin River chub. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the Virgin River chub's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the Virgin River chub.

RIO GRANDE SILVERY MINNOW

Hybognathus amarus

Rio Grande silvery minnow exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated Rio Grande silvery minnow exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that Rio Grande silvery minnows exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Rio Grande silvery minnow females are short-lived (less than

2 years). Norris et al. (2008) conducted a population viability analysis for the Rio Grande silvery minnow. The simulations indicated that reproductive output of those individuals just shy of one year old entering their first spawning season – hereafter referred to as Age 0 fish – is a primary factor that determines the extent of population growth from year to year. This reproductive rate, as defined in our analysis, includes both egg production and larval / juvenile survival rate to the next year's spawn.

We anticipate the reproductive output of Age 0 fish would be reduced substantially. The Rio Grande silvery minnow's reproductive output would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Rio Grande silvery minnow's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Rio Grande silvery minnows may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Rio Grande silvery minnow population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Rio Grande silvery minnows are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Rio Grande silvery minnow.

The physical and biological features of critical habitat essential to the conservation of the Rio Grande silvery minnow include a hydrologic regime which provides sufficient flowing water with low to moderate currents that form the aquatic habitats which the minnow prefers, such as backwaters, shallow side channels, pools, eddies, and runs of varying depth and velocity. The minnow also requires substrates of sand or silt, and sufficient water quality (water of proper temperatures and conditions such as dissolved oxygen content and pH).

Critical Habitat: Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect Rio Grande silvery minnow critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of Rio Grande silvery minnows, and cause Rio Grande silvery minnows to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 31% and the reduction in the survival of young fish through the first year as high as 33%. These effects are estimated to be of a magnitude great enough to reduce numbers

of Rio Grande silvery minnows. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the Rio Grande silvery minnow's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the Rio Grande silvery minnows.

BIG SPRING SPINEDACE

Lepidomeda mollispinis pratensis

Big Spring spinedace exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 31). Compared to control populations, we estimated Big Spring spinedace exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that Big Spring spinedaces exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Big Spring spinedace are sexually mature after about a year and live up to 4 years. Most spawning of the Big Spring spinedace occurs from March through June, with some continuing sporadically into July. The big spring spinedace is believed to broadcast spawn over gravel substrate. Females generally produce from 380-640 eggs during spawning, with older females sometimes producing two complements of ova in one breeding season. (BRRC 2001).

A comparison of 88 freshwater fish species, found that longevity and age at maturity were the best predictors of elasticity patterns (Vélez-Espino et al., 2006). Elasticity analyses performed on cyprinids with similar longevity, age at maturity, and reproductive lifespan as the Big Spring spinedace found that population growth rates for these species were highly susceptible to perturbations in juvenile survival and fecundity (Vélez-Espino et al., 2006). In combination, these two stages accounted for over 90% of the total elasticity of the population growth rate in all species with these life-history traits. These reproductive parameters, as defined in our analysis, include both egg production and larval / juvenile survival rate to the next year's spawn

We anticipate the Big Spring spinedace's reproductive performance would be reduced substantially. The Big Spring spinedace's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Big Spring spinedace's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Big Spring spinedaces may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Big Spring spinedace population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Big Spring spinedaces are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Big Spring spinedace.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the Big Spring spinedace include the following: Clean, permanent, flowing, spring-fed stream habitat with deep pool areas and shallow marshy areas along the shore; and the absence of nonnative fishes.

Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect Big Spring spinedace critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of Big Spring spinedaces, and cause Big Spring spinedaces to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 31% and the reduction in the survival

of young fish through the first year as high as 33%. These effects are estimated to be of a magnitude great enough to reduce numbers of Big Spring spinedace. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the Big Spring spinedace's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the Big Spring spinedace.

LITTLE COLORADO SPINEDACE

Lepidomeda vitatta

Little Colorado spinedace exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated Little Colorado spinedace exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that Little Colorado spinedaces exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Little Colorado spinedace are believed to live 3 to 4 years and mature early. Spinedace are late-spring to early-summer spawners although some females have been found to contain mature eggs as late as October. Spawning occurs in slow current over cobbles. Females may spawn more than once per year, and fecundity estimates range from 650-1000 total eggs per female.

A comparison of 88 freshwater fish species, found that longevity and age at maturity were the best predictors of elasticity patterns (Vélez-Espino et al., 2006). Elasticity analyses performed on cyprinids with similar longevity, age at maturity, and reproductive lifespan as the Little Colorado spinedace found that population growth rates for these species were highly susceptible to perturbations in juvenile survival and fecundity (Vélez-Espino et al., 2006). In combination, these two stages accounted for over 90% of the total elasticity of the population growth rate in all species with these life-history traits. These reproductive parameters, as defined in our analysis, include both egg production and larval / juvenile survival rate to the next year's spawn

We anticipate the Little Colorado spinedace's reproductive performance would be reduced substantially. The Little Colorado spinedace's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Little Colorado spinedace's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Little Colorado spinedaces may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Little Colorado spinedace population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Little Colorado spinedaces are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Little Colorado spinedace.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the Little Colorado spinedace include clean, permanently flowing water with pools and a fine gravel or silt-mud substrate. Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect Little Colorado spinedace critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of Little Colorado spinedace, and cause Little Colorado spinedace to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 31% and the reduction in the survival of young fish through the first year as high as 33%. These effects are estimated to be of a magnitude great enough to reduce numbers of Little Colorado spinedace. Continued

approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the Little Colorado spinedace's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the Little Colorado spinedace.

SPIKEDACE

Meda fulgida

Spikedace exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated spikedace exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that spikedace exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Spikedace females are short-lived (1-2 years) and mature early. Spawning occurs in the spring between April and June and seems to be triggered by a combination of stream discharge and water temperature. Females may be fractional spawners, with elapsed periods of days to weeks between spawning. Fecundity is correlated to age and length, and has been found to be between 90 to 250 ova.

A comparison of 88 freshwater fish species, found that life history parameters such as age at maturity, reproductive lifespan, fecundity, juvenile survivorship, and longevity could

be predictors of elasticity patterns (Vélez-Espino et al., 2006). The spikedace has a reproductive strategy similar to the smalleye shiner (*Notropis buccula*), a member of a reproductive guild of cyprinids that broadcast spawn multiple batches of nonadhesive, semibuoyant ova throughout an extended reproductive season, and experience extremely high post-spawning mortality. Durham and Wilde (2009) studied the population dynamics of the smalleye shiner in the Brazos River, Texas. Elasticity analysis and sensitivity simulations of the projection matrix indicated that age-0 survival and age-1 fecundity were the most influential parameters in the population dynamics of smalleye shiners. In combination, these two stages accounted for the majority (70%) of the total elasticity of the population growth rate. These reproductive parameters, as defined in our analysis, include both egg production and larval / juvenile survival rate to the next year's spawn

We anticipate the spikedace's reproductive performance would be reduced substantially. The spikedace's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the spikedace's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Spikedace may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected spikedace population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately spikedace are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the spikedace.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the spikedace include the following: living areas for each stage of the spikedace, a proper food base, a lack of non-native species, and high quality water, with the proper amount of dissolved oxygen, and no or minimal pollutant levels for pollutants such as copper, arsenic, mercury, and cadmium; human and animal waste products; pesticides; suspended sediments; and gasoline or diesel fuels.

Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect spikedace critical habitat by diminishing the quality of

water to the degree that it would impair individual reproduction and survival of spikedace, and cause spikedace to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 31% and the reduction in the survival of young fish through the first year as high as 33%. These effects are estimated to be of a magnitude great enough to reduce numbers of spikedace. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the spikedace's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the spikedace.

MOAPA DACE

Moapa coriacea

Moapa dace exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated Moapa dace exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that Moapa daces exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Moapa dace reach sexual maturity at 1 year of age and can

live up to 4 years. Moapa dace apparently reproduce year-round, peaking in the spring, in water temperatures ranging from 28°C-32°C. Fecundity is related to fish size and egg counts have range from 60 to 772 depending on size. A comparison of 88 freshwater fish species, found that longevity and age at maturity were the best predictors of elasticity patterns (Vélez-Espino et al., 2006). Elasticity analyses performed on cyprinids with similar longevity, age at maturity, and reproductive lifespan as the Moapa dace found that population growth rates for these species were highly susceptible to perturbations in juvenile survival and fecundity (Vélez-Espino et al., 2006). In combination, these two stages accounted for over 90% of the total elasticity of the population growth rate in all species with these life-history traits. These reproductive parameters, as defined in our analysis, include both egg production and larval / juvenile survival rate to the next year's spawn

We anticipate the Moapa dace's reproductive performance would be reduced substantially. The Moapa dace's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Moapa dace's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Moapa daces may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Moapa dace population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Moapa daces are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Moapa dace.

PALEZONE SHINER

Notropis albizonatus

Palezone shiner exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated palezone shiner exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that palezone shiners exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Palezone shiner females are short-lived (3-4 years).

We anticipate the palezone shiner's reproductive performance would be reduced substantially. The palezone shiner's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the palezone shiner's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Palezone shiners may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected palezone shiner population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately palezone shiners are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration.

Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the palezone shiner.

CAHABA SHINER

Notropis cahabae

Cahaba shiner exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated Cahaba shiner exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that Cahaba shiners exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Cahaba shiner females are short-lived (probably 3- 4 years), mature early, and spawn from late May through June.

We anticipate the Cahaba shiner's reproductive performance would be reduced substantially. The Cahaba shiner's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction

could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Cahaba shiner's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Cahaba shiners may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Cahaba shiner population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Cahaba shiners are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Cahaba shiner.

ARKANSAS RIVER SHINER

Notropis girardi

Arkansas River shiner exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated Arkansas River shiner exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that Arkansas River shiners exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a

whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Arkansas River shiner females are short-lived (less than 3 years) and appear to experience extremely high mortality after spawning.

The Arkansas River minnow has a reproductive strategy similar to the smalleye shiner (*Notropis buccula*). Durham and Wilde (2009) studied the population dynamics of the smalleye shiner in the Brazos River, Texas. Smalleye shiner are members of a reproductive guild of cyprinids that broadcast spawn multiple batches of nonadhesive, semibuoyant ova throughout an extended reproductive season, and post-spawning mortality is extremely high. Elasticity analysis and sensitivity simulations of the projection matrix indicated that age-0 survival and age-1 fecundity were the most influential parameters in the population dynamics of smalleye shiners. In combination, these two stages accounted for the majority (70%) of the total elasticity of the population growth rate.

We anticipate the Arkansas River shiner's reproductive performance would be reduced substantially. The Arkansas River shiner's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Arkansas River shiner's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Arkansas River shiners may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Arkansas River shiner population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Arkansas River shiners are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Arkansas River shiner.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the Arkansas River shiner include the following: —(i) a natural, unregulated hydrologic regime complete with episodes of flood and drought or, if flows are modified or regulated, a hydrologic regime characterized by the duration, magnitude, and frequency of flow events capable of forming and maintaining channel and instream

habitat necessary for particular Arkansas River shiner life-stages in appropriate seasons; (ii) a complex, braided channel with pool, riffle (shallow area in a streambed causing ripples), run, and backwater components that provide a suitable variety of depths and current velocities in appropriate seasons; (iii) a suitable unimpounded stretch of flowing water of sufficient length to allow hatching and development of the larvae; (iv) a river bed of predominantly sand, with some patches of gravel and cobble; (v) water quality characterized by low concentrations of contaminants and natural, daily and seasonally variable temperature, turbidity, conductivity, dissolved oxygen, and pH; (vi) suitable reaches of aquatic habitat, as defined by primary constituent elements (i) through (v) above, and adjacent riparian habitat sufficient to support an abundant terrestrial, semiaquatic, and aquatic invertebrate food base; and (vii) few or no predatory or competitive non-native fish species present.

Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect Arkansas River shiner critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of Arkansas River shiners, and cause Arkansas River shiners to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 31% and the reduction in the survival of young fish through the first year as high as 33%. These effects are estimated to be of a magnitude great enough to reduce numbers of Arkansas River shiner. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the Arkansas River shiner's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the Arkansas River shiner.

CAPE FEAR SHINER

Notropis mekistocholas

Cape Fear shiners exposed to cyanide at the criterion continuous concentration (CCC) are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Cape Fear shiners exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 59%. We estimate that Cape Fear shiners exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish

through the first year and that reduction could be as much as, but is not likely to be greater than, 68%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like Cape Fear shiners, than on species with greater adult survival, such as sturgeon. Cape Fear shiners live about two to three years. No information is presently available about this species' reproductive characteristics.

Hartup (2005) conducted field research on the slackwater darter and holiday darter (*E. brevirostrum*) and developed species-specific estimates for fecundity, adult survival, and population sizes, then used these data to perform population viability analyses for the two species. Average slackwater darter annual fecundity was estimated as 92 and 197 eggs spawned, respectively, for one-batch and two-batch fecundity. Two-batch fecundity for the holiday darter was averaged 175 eggs spawned. Based on estimates of adult survival, Hartup (2005) estimated the adult fertility rate (number of female offspring surviving to year 1) would need to average 0.896 for the slackwater darter population to be stable and 0.818 for the holiday darter population to be stable. Assuming the slackwater darter population was stable, Hartup estimated the average proportion of spawned eggs needing to survive through the first year as 0.019 or 0.009, for one- and two-batch fecundity, respectively. For the holiday darter, the two-batch estimate was also 0.009. An elasticity analysis for the slackwater darter and holiday darter (*E. brevirostrum*) identified fertilities (defined as fecundity plus juvenile survival) as having the largest relative contribution to population growth rates for each species, as opposed to adult survival (Hartup 2005).

Since there are no data on Cape Fear shiner, we infer the Cape Fear shiner's fecundity and survival are no greater than those estimated for the slackwater darter and holiday darter. Consequently, the reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the Cape Fear shiner's fertility substantially. The Cape Fear shiner's fertility rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Cape Fear shiner's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Cape Fear shiners may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Cape Fear shiner population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Cape Fear shiners are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Cape Fear shiner.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the Cape Fear shiner include water of sufficient quality for species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience individual and population growth. Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to these levels. This approval could adversely affect the quality of water to the degree that it would impair individual reproduction and survival of Cape Fear shiners, and cause shiners to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high 59% and the reduction in the survival of young fish through the first year as high as 68%. These effects are estimated to be of a magnitude great enough to reduce numbers of Cape Fear shiners. Approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the Cape Fear shiner.

PECOS BLUNTNOSE SHINER

Notropis simus pecosensis

Pecos bluntnose shiner exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated Pecos bluntnose shiner exposed to cyanide at the CCC could experience a

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substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that Pecos bluntnose shiners exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Pecos bluntnose shiner females are short-lived (up to 3 years).

The Pecos bluntnose shiner has a reproductive strategy similar to the smalleye shiner (*Notropis buccula*). Durham and Wilde (2009) studied the population dynamics of the smalleye shiner in the Brazos River, Texas. Smalleye shiner are members of a reproductive guild of cyprinids that broadcast spawn multiple batches of nonadhesive, semibuoyant ova throughout an extended reproductive season, and post-spawning mortality is extremely high. Elasticity analysis and sensitivity simulations of the projection matrix indicated that age-0 survival and age-1 fecundity were the most influential parameters in the population dynamics of smalleye shiners. In combination, these two stages accounted for the majority (70%) of the total elasticity of the population growth rate.

We anticipate the Pecos bluntnose shiner's reproductive performance would be reduced substantially. The Pecos bluntnose shiner's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Pecos bluntnose shiner's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Pecos bluntnose shiners may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent

compensatory mechanisms, if they exist, to be overwhelmed. An effected Pecos bluntnose shiner population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Pecos bluntnose shiners are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Pecos bluntnose shiner.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the Pecos bluntnose shiner include clean permanent water, a main river channel habitat with a sandy substrate, and a low velocity flow. Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect Pecos bluntnose shiner critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of Pecos bluntnose shiners, and cause Pecos bluntnose shiners to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 31% and the reduction in the survival of young fish through the first year as high as 33%. These effects are estimated to be of a magnitude great enough to reduce numbers of Pecos bluntnose shiner. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the Pecos bluntnose shiner's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the Pecos bluntnose shiner.

TOPEKA SHINER
Notropis Topeka

Topeka shiner exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated Topeka shiner exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that Topeka shiners exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like Topeka shiners, than on species with greater adult survival, such as sturgeon. Topeka shiner females are short-lived (1-2 years), mature early, and spawn year-round with two peaks in August and late winter.

The Topeka shiner has a reproductive strategy similar to the smalleye shiner (*Notropis buccula*). Durham and Wilde (2009) studied the population dynamics of the smalleye shiner in the Brazos River, Texas. Smalleye shiner are members of a reproductive guild of cyprinids that broadcast spawn multiple batches of nonadhesive, semibuoyant ova throughout an extended reproductive season, and post-spawning mortality is extremely high. Elasticity analysis and sensitivity simulations of the projection matrix indicated that age-0 survival and age-1 fecundity were the most influential parameters in the population dynamics of smalleye shiners. In combination, these two stages accounted for the majority (70%) of the total elasticity of the population growth rate. Although Topeka shiners utilized a different spawning strategy than smalleye shiners, they are similar in that survival of post-spawning adults is extremely low. Consequently, we anticipate that age-0 survival and age-1 fecundity will similarly be most important to the Topeka shiner's growth rate.

We anticipate the Topeka shiner's reproductive performance would be reduced substantially. The Topeka shiner's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Topeka shiner's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Topeka shiners may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if

they exist, to be overwhelmed. An effected Topeka shiner population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Topeka shiners are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Topeka shiner.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the Topeka shiner include the following: stream most often with permanent flow, but which become dry during intermittent periods. Side channel pools and oxbows which are either seasonally connected to a stream or maintained by groundwater inputs are also required. Streams and side channel pools with water quality (temperature, turbidity, conductivity, salinity, dissolved oxygen, pH, chemical contaminants, and other chemical characteristics) necessary for unimpaired behavior, growth, and viability of all life stages is essential. Living and spawning areas for adults are required, which must have water velocities of less than a half meter per second, and depths from 0.1 to 2 meters. Similarly, living areas for juveniles are required. These must have the same flow rate as areas for adults, but require greatly decreased depths, that of less than a quarter of a meter. Proper substrate for the Topeka shiner is sand, gravel, cobble, or silt, with proper amounts of fine sediment and substrate embeddedness. A proper food base of terrestrial, semiaquatic and aquatic invertebrates is necessary. A hydrologic regime capable of forming, maintaining, or restoring flow periodicity, channel morphology, fish community composition, off channel habitats, and habitat components essential for the fish. Finally, this fish requires few or no nonnative predatory or competitive species to be present in their habitat.

Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect Topeka shiner critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of Topeka shiners, and cause Topeka shiners to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 31% and the reduction in the survival of young fish through the first year as high as 33%. These effects are estimated to be of a magnitude great enough to reduce numbers of Topeka shiner. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the Topeka shiner's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the Topeka shiner.

OREGON CHUB
Oregonichthys crameri

Oregon chub exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated Oregon chub exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that Oregon chubs exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Oregon chub females exhibit an intermediate life span, mature in their second year, and spawn from mid-May through August with peak activity in July. A comparison of 88 freshwater fish species, found that longevity and age at maturity were the best predictors of elasticity patterns (Vélez-Espino et al., 2006). Elasticity analyses performed on cyprinids with similar longevity, age at maturity, and reproductive lifespan as the Oregon chub found that population growth rates for these species were highly susceptible to perturbations in juvenile survival. Juvenile survival accounted for about 58% of the total elasticity of the population growth rate in all species with these life-history traits. A comparison of elasticities among closely related species found that elasticity values are highly conserved among genera within the same taxonomic family.

We anticipate the Oregon chub's reproductive performance would be reduced substantially. The Oregon chub's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The

combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Oregon chub's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Oregon chubs may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Oregon chub population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Oregon chubs are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Oregon chub.

BLACKSIDE DACE

Phoxinus cumberlandensis

Blackside dace exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated blackside dace exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that blackside daces exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival. Blackside dace generally have a lifespan of 3 years, reach sexual maturity at age 1, and are broadcast spawners, spawning over clean, gravel-sized substrate from April to July. Average fecundity is about 1500. A comparison of 88 freshwater fish species, found that longevity and age at maturity were the best predictors of elasticity patterns, and elasticity values are highly conserved among genera within the same taxonomic family (Vélez-Espino et al., 2006). Sensitivity and elasticity analyses were performed for the southern redbelly dace, (*Phoxinus erythrogaster*), a closely-related species with similar longevity, age at maturity, and reproductive lifespan (Stasiak 2007). Results indicated that population growth rate was most sensitive (>96%) to changes in first-year survival and most elastic to changes in first-year reproduction, followed by first-year survival. Overall fertility accounted for 70% of total elasticity. These reproductive parameters, as defined in our analysis, include both egg production and larval / juvenile survival rate to the next year's spawn

We anticipate the blackside dace's fertility would be reduced substantially. The blackside dace's fertility rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the blackside dace's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Blackside daces may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected blackside dace population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately blackside daces are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the blackside dace.

WOUNDFIN
Plagopterus argentissimus

Woundfin exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated Woundfin exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that Woundfin exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, than on species with greater adult survival. The generation time for the woundfin is predominately limited to 1 year, with most individuals reaching sexual maturity in the second year of a 3-year lifespan. Individuals must achieve sufficient growth prior to the spring spawning period in order to contribute to the next generation. In a comparison of 88 freshwater fish species, longevity and age at maturity were found to be the best predictors of elasticity patterns (Vélez-Espino et al., 2006). Elasticity analyses performed on cyprinids with similar longevity, age at maturity, and reproductive lifespan to the woundfin found that population growth rates were most susceptible to perturbations in juvenile survival, followed by fecundity (Vélez-Espino et al., 2006). In combination, these two stages accounted for over 90% of the total elasticity of the population growth rate. These reproductive parameters, as defined in our analysis, include both egg production and larval / juvenile survival rate to the next year's spawn.

We anticipate the woundfin's reproductive performance fertility would be reduced substantially. The woundfin's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through

the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the woundfin's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Woundfins may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected woundfin population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately woundfins are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the woundfin.

Critical Habitat: Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect woundfin critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of woundfins, and cause woundfins to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 31% and the reduction in the survival of young fish through the first year as high as 33%. These effects are estimated to be of a magnitude great enough to reduce numbers of woundfin. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the woundfin's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the woundfin.

COLORADO PIKEMINNOW

Ptychocheilus lucius

Colorado pikeminnows exposed to cyanide at the criterion continuous concentration (CCC) are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were

available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Colorado pikeminnows exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 63%. We estimate that Colorado pikeminnows exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 71%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation).

The Colorado pikeminnow is a long-lived fish (40+ years) that evolved in a variable system, with high adaptability to natural environmental variability and resilience to natural catastrophes. This evolution has become manifest as pulsed recruitment from periodic strong year classes, great longevity of adults, and low vulnerability of adults to environmental influences. Great longevity and stability of adults provides a “storage effect” for populations, into which periodic recruitment from strong year classes allows fish to become stored (Gilpin 1993). This is seen as a way that Colorado pikeminnow maintain long-term population viability and stability under environmental variation.

As stated in the recovery plan for this species, a critical aspect of recovery is increased frequency of strong year classes. The reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the Colorado pikeminnow’s potential recruitment substantially. The Colorado pikeminnow’s potential recruitment would be diminished because of (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates diminish survival through the first winter. The reductions in the Colorado pikeminnow’s recruitment could diminish the frequency and could even preclude the occurrence of strong year classes.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Colorado pikeminnow’s reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Colorado pikeminnows may also

experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Colorado pikeminnow population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Colorado pikeminnows are likely to become extirpated from waters where they are exposed to cyanide toxicity at the CCC. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Colorado pikeminnow.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the Colorado pikeminnow include water of sufficient quality for species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience individual and population growth. Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to these levels. This approval could adversely affect the quality of water to the degree that it would impair individual reproduction and survival of Colorado pikeminnow, and cause pikeminnows to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 683 and the reduction in the survival of young fish through the first year as high as 71%. These effects are estimated to be of a magnitude great enough to reduce numbers of Colorado pikeminnows. Approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the Colorado pikeminnow.

ASH MEADOWS SPECKLED DACE

Rhinichthys osculus spp

Ash Meadows speckled dace exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated Ash Meadows speckled dace exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that Ash Meadows speckled daces exposed to cyanide at the CCC could experience a substantial reduction in

the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, than on species with greater adult survival. Ash Meadows speckled dace females reach sexual maturity at 2 years, spawn primarily over the spring and summer, and are believed to live up to 4 years. Elasticity analyses performed on cyprinids with similar life-history traits, including the blacknose dace, *Rhinichthys atratulus*, found that population growth rates for these species were most susceptible to perturbations in juvenile survival, followed by fecundity (Vélez-Espino et al., 2006). In combination, these two stages accounted for over 90% of the total elasticity of the population growth rate. A comparison of elasticities among closely related species found that elasticity values are highly conserved among genera within the same taxonomic family (Vélez-Espino et al., 2006). These reproductive parameters, as defined in our analysis, include both egg production and larval / juvenile survival rate to the next year's spawn

We anticipate the Ash Meadows speckled dace's fertility would be reduced substantially. The Ash Meadows speckled dace's fertility rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Ash Meadows speckled dace's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Ash Meadows speckled daces may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Ash Meadows speckled dace population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Ash Meadows

speckled daces are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Ash Meadows speckled dace.

Critical Habitat: Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect Ash Meadows speckled dace critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of Ash Meadows speckled daces, and cause Ash Meadows speckled daces to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 31% and the reduction in the survival of young fish through the first year as high as 33%. These effects are estimated to be of a magnitude great enough to reduce numbers of Ash Meadows speckled dace. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the Ash Meadows speckled dace's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the Ash Meadows speckled dace.

KENDALL WARM SPRINGS DACE

Rhinichthys osculus thermalis

Kendall Warm Springs dace exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated Kendall Warm Springs dace exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that Kendall Warm Springs daces exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn. Kendall Warm Springs dace females reach sexual maturity at 2 years, are thought to spawn several times a year, and are likely to live between 3 and 5 years. Elasticity analyses performed on cyprinids with similar life-history traits, including the blacknose dace, *Rhinichthys atratulus*, found that population growth rates for these species were most susceptible to perturbations in juvenile survival, followed by fecundity (Vélez-Espino et al., 2006). In combination, these two stages accounted for over 90% of the total elasticity of the population growth rate. A comparison of elasticities among closely related species found that elasticity values are highly conserved among genera within the same taxonomic family (Vélez-Espino et al., 2006). These reproductive parameters, as defined in our analysis, include both egg production and larval / juvenile survival rate to the next year's spawn

We anticipate the Kendall Warm Springs dace's reproductive performance would be reduced substantially. The Kendall Warm Springs dace's reproductive performance rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Kendall Warm Springs dace's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Kendall Warm Springs daces may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Kendall Warm Springs dace population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Kendall Warm Springs daces are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Kendall Warm Springs dace.

LOACH MINNOW
Tiaroga cobitis

Loach minnow exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated Loach minnow exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 31%. We estimate that Loach minnows exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 33%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than species with greater adult survival. Loach minnow females are short-lived (1-2 years), and mature early. Elasticity analyses performed on cyprinids with similar life-history traits found that population growth rates for these species were highly susceptible to perturbations in juvenile survival and fecundity (Vélez-Espino et al., 2006). In combination, these two stages accounted for over 90% of the total elasticity of the population growth rate in all species with these life-history traits. These reproductive parameters, as defined in our analysis, include both egg production and larval / juvenile survival rate to the next year's spawn.

We anticipate the Loach minnow's reproductive performance would be reduced substantially. The Loach minnow's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction

could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Loach minnow's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Loach minnows may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Loach minnow population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Loach minnows are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Loach minnow.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the loach minnow include the following: permanent flowing water with no or low levels of pollutants, which must include living areas for adult loach minnows with moderate to swift flow velocities (9 to 32 inches per second) in shallow water between approximately 1 and 30 inches (3 to 75 cm) in depth. The substrate of these areas should consist of gravel, cobble, or rubble. Furthermore, there must be living areas for juveniles as well. To support juveniles, areas must have moderate to swift flow velocities, from 1 to 34 inches per second (3 and 85 cm/second). Juveniles require the same depth and substrates as adults. It is the larval loach minnow which requires greatly different habitat. Loach minnow larvae require slow to moderate flow velocities, from 3 to 20 inches per second (9 to 50 cm/second) and shallow water with sand, gravel, and cobble substrates.

Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect loach minnow critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of loach minnows, and cause loach minnows to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 31% and the reduction in the survival of young fish through the first year as high as 33%. These effects are estimated to be of a magnitude great enough to reduce numbers of loach minnow. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the loach minnow's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the loach minnow.

Gasterosteidae

UNARMORED THREESPINE STICKLEBACK
Gasterosteus aculeatus williamsoni

Unarmored threespine stickleback exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated unarmored threespine stickleback exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 48%. We estimate that Unarmored threespine stickleback s exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 56%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than species with greater adult survival. Unarmored threespine stickleback females are short-lived (about 1 year), mature early, and lay 50-300 eggs into nests guarded by males. A comparison of 88 freshwater fish species, found that longevity and age at maturity were the best predictors of elasticity patterns (Vélez-Espino et al., 2006). Elasticity analyses performed on freshwater fish with similar longevity and reproductive lifespans to the Unarmored threespine stickleback found that population growth rates for these species were highly susceptible to perturbations in juvenile survival and fecundity (Vélez-Espino et al., 2006). In combination, these two stages accounted for over 90% of the total elasticity of the population growth rate in all species with these life-history traits. These reproductive parameters, as defined in our analysis, include both egg production and larval / juvenile survival rate to the next year's spawn.

We anticipate the unarmored threespine stickleback's reproductive performance would be reduced substantially. The unarmored threespine stickleback's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the unarmored threespine stickleback's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Unarmored threespine stickleback s may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected unarmored threespine stickleback population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately unarmored threespine sticklebacks are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the unarmored threespine stickleback .

Gobiidae

TIDEWATER GOBY

Eucyclogobius newberryi

Tidewater goby exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimated tidewater goby exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 36%. We estimate that tidewater gobies exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through

the first year and that reduction could be as much as, but is not likely to be greater than, 40%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than species with greater adult survival. Tidewater goby females are short-lived (about 1 year), mature early, and can reproduce all year. A comparison of 88 freshwater fish species, found that longevity and age at maturity were the best predictors of elasticity patterns (Vélez-Espino et al., 2006). Elasticity analyses performed on freshwater fish with similar longevity and reproductive lifespans to the tidewater goby found that population growth rates for these species were highly susceptible to perturbations in juvenile survival and fecundity (Vélez-Espino et al., 2006). In combination, these two stages accounted for over 90% of the total elasticity of the population growth rate in all species with these life-history traits. These reproductive parameters, as defined in our analysis, include both egg production and larval / juvenile survival rate to the next year's spawn.

We anticipate the tidewater goby's reproductive performance would be reduced substantially. The tidewater goby's reproductive performance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the tidewater goby's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Tidewater gobies may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An affected tidewater goby population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately tidewater gobies are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration.

Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the tidewater goby.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the tidewater goby include the following: Persistent, shallow (in the range of about 0.1 to 2 m), still-to-slow-moving aquatic habitat most commonly ranging in salinity from less than 0.5 ppt to about 10 to 12 ppt; Substrates (e.g., sand, silt, mud) suitable for the construction of burrows for reproduction; Submerged and emergent aquatic vegetation, such as *Potamogeton pectinatus* and *Ruppia maritima*, that provides protection from predators; and Presence of a sandbar(s) across the mouth of a lagoon or estuary during the late spring, summer, and fall that closes or partially closes the lagoon or estuary, thereby providing relatively stable water levels and salinity.

Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect tidewater goby critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of tidewater gobies, and cause tidewater goby to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 36% and the reduction in the survival of young fish through the first year as high as 40%. These effects are estimated to be of a magnitude great enough to reduce numbers of tidewater goby. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the tidewater goby's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the tidewater goby.

Goodeidae

WHITE RIVER SPRINGFISH (ASH SPRING)

Crenichthys baileyi baileyi

White River springfish exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate White River springfish exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much

as, but is not likely to be greater than, 48%. We estimate that White River springfish exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 56%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

Springfish (genus *Crenichthys*) are uniquely adapted to survive in high temperatures (e.g. 84 to 97 F for Railroad Valley springfish, U.S. Fish and Wildlife Service 1996) and only occur in hot spring environments in Southern Nevada. Their thermal tolerance has enabled them to evolve in habitats that are otherwise uninhabitable for other fish species that are native to the region. Thus, historic populations have benefited from the lack of competition and predation by other fishes. Although life history information is limited, Springfish are believed to be relatively short lived (3 to 4 years), spawn infrequently (average two spawnings per year), and deposit few eggs per spawn (10 to 17; Kopec 1949). Such a reproductive strategy is unlike any other that was previously described in the *Population Response* section and would appear to be highly vulnerable to stressors which reduce fecundity or survival of larvae, juveniles or adults. The vulnerability of this strategy was evidenced by the decline in the Springfish populations which occurred following the introduction of nonnative fish species (Tuttle et al. 1990, U.S. Fish and Wildlife Service 1996 & 1998). These reductions were attributed in large part to predation by nonnative fishes on springfish larvae (U.S. Fish and Wildlife Service 1998). The effects of cyanide on fecundity, egg hatchability, and larval/juvenile survival could result in similar impacts on the springfish populations, as well as, worsen effects caused by nonnative fish predation.

The reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the White River springfish's potential recruitment substantially. The White River springfish's potential recruitment would be diminished because of (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates diminish survival through the first winter. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the White River springfish's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. White River springfish may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory

mechanisms, if they exist, to be overwhelmed. An effected White River springfish population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately White River springfish are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the White River springfish.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the White River springfish include the following: warm water springs and their outflows and surrounding areas that provide vegetation for cover and habitat for insects and other invertebrates on which the species feeds. By warm water springs and their outflows, we mean water of sufficient quality for the species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience individual and population growth sufficient for the critical habitat to serve its intended conservation function.

Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to the criterion concentration. This approval could adversely affect White River springfish critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of White River springfish, and cause White River springfish to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high 48% and the reduction in the survival of young fish through the first year as high as 56%. These effects are estimated to be of a magnitude great enough to reduce numbers of White River springfish. Continued approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the White River springfish's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the White River springfish.

HIKO WHITE RIVER SPRINGFISH

Crenichthys baileyi grandis

Hiko White River springfish exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we

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estimate Hiko White River springfish exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 48%. We estimate that Hiko White River springfish exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 56%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

Springfish (genus *Crenichthys*) are uniquely adapted to survive in high temperatures (e.g. 84 to 97 F for Railroad Valley springfish, U.S. Fish and Wildlife Service 1996) and only occur in hot spring environments in Southern Nevada. Their thermal tolerance has enabled them to evolve in habitats that are otherwise uninhabitable for other fish species that are native to the region. Thus, historic populations have benefited from the lack of competition and predation by other fishes. Although life history information is limited, Springfish are believed to be relatively short lived (3 to 4 years), spawn infrequently (average two spawnings per year), and deposit few eggs per spawn (10 to 17; Kopec 1949). Such a reproductive strategy is unlike any other that was previously described in the *Population Response* section and would appear to be highly vulnerable to stressors which reduce fecundity or survival of larvae, juveniles or adults. The vulnerability of this strategy was evidenced by the decline in the Springfish populations which occurred following the introduction of nonnative fish species (Tuttle et al. 1990, U.S. Fish and Wildlife Service 1996 & 1998). These reductions were attributed in large part to predation by nonnative fishes on springfish larvae (U.S. Fish and Wildlife Service 1998). The effects of cyanide on fecundity, egg hatchability, and larval/juvenile survival could result in similar impacts on the springfish populations, as well as, worsen effects caused by nonnative fish predation.

The reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the Hiko White River springfish's potential recruitment substantially. The Hiko White River springfish's potential recruitment would be diminished because of (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates diminish survival through the first winter. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Hiko White River springfish's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Hiko White River springfish

may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Hiko White River springfish population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Hiko White River springfish are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Hiko White River springfish.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the Hiko White River springfish include the following: warm water springs and their outflows and surrounding areas that provide vegetation for cover and habitat for insects and other invertebrates on which the species feeds. By warm water springs and their outflows, we mean water of sufficient quality for the species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience individual and population growth sufficient for the critical habitat to serve its intended conservation function.

Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to the criterion concentration. This approval could adversely affect Hiko White River springfish critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of Hiko White River springfish, and cause Hiko White River springfish to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high 48% and the reduction in the survival of young fish through the first year as high as 56%. These effects are estimated to be of a magnitude great enough to reduce numbers of Hiko White River springfish. Continued approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the Hiko White River springfish's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the Hiko White River springfish.

RAILROAD VALLEY SPRINGFISH

Crenichthys nevadae

Railroad Valley springfish exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Railroad Valley springfish exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 48%. We estimate that Railroad Valley springfish exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 56%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

Springfish (genus *Crenichthys*) are uniquely adapted to survive in high temperatures (e.g. 84 to 97 F for Railroad Valley springfish, U.S. Fish and Wildlife Service 1996) and only occur in hot spring environments in Southern Nevada. Their thermal tolerance has enabled them to evolve in habitats that are otherwise uninhabitable for other fish species that are native to the region. Thus, historic populations have benefited from the lack of competition and predation by other fishes. Although life history information is limited, Springfish are believed to be relatively short lived (3 to 4 years), spawn infrequently (average two spawnings per year), and deposit few eggs per spawn (10 to 17; Kopec 1949). Such a reproductive strategy is unlike any other that was previously described in the *Population Response* section and would appear to be highly vulnerable to stressors which reduce fecundity or survival of larvae, juveniles or adults. The vulnerability of this strategy was evidenced by the decline in the Springfish populations which occurred following the introduction of nonnative fish species (Tuttle et al. 1990, U.S. Fish and Wildlife Service 1996 & 1998). These reductions were attributed in large part to predation by nonnative fishes on springfish larvae (U.S. Fish and Wildlife Service 1998). The effects of cyanide on fecundity, egg hatchability, and larval/juvenile survival could result in similar impacts on the springfish populations, as well as, worsen effects caused by nonnative fish predation.

The reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the Railroad Valley springfish's potential recruitment substantially. The Railroad Valley springfish's potential recruitment would be diminished because of (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates diminish survival through the first winter. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Railroad Valley springfish's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Railroad Valley springfish may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Railroad Valley springfish population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Railroad Valley springfish are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Railroad Valley springfish.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the Railroad Valley springfish include the following: clear, unpolluted thermal spring waters ranging in temperature from 84 to 97 F in pools, flowing channels, and marshy areas with aquatic plants, insects and mollusks. By clear, unpolluted thermal spring waters, we mean water of sufficient quality for the species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience individual and population growth sufficient for the critical habitat to serve its intended conservation function.

Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to the criterion concentration. This approval could adversely affect Railroad Valley springfish critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of Railroad Valley springfish, and cause Railroad Valley springfish to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high 48% and the reduction in the survival of young fish through the first year as high as 56%. These effects are estimated to be of a magnitude great enough to reduce numbers of Railroad Valley springfish. Continued approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the Railroad Valley springfish's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the Railroad Valley springfish.

Osmeridae

DELTA SMELT

Hypomesus transpacificus

Delta smelt exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth,

swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate delta smelt exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 48%. We estimate that delta smelt exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 56%.

As noted previously, the young fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like delta smelt, than on species with greater adult survival, such as sturgeon. Delta smelt live only 1 year and die after spawning. Female smelt lay between 1,600 and 2,600 eggs in the spring. The number and hatchability of eggs together with the survival of larval and juvenile smelt are what drives annual population abundance in this annual fish species.

We anticipate the delta smelt's annual abundance would be reduced substantially. The delta smelt's annual abundance would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year to spawn, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the delta smelt's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish. Delta smelt may also experience effects on growth,

swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected delta smelt population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur and the delta smelt's current population abundance, we conclude ultimately delta smelt are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the delta smelt.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the delta smelt include water of sufficient quality for the species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience adequate individual and population growth. Approval of the CCC in State and Tribal water quality standards would authorized States and Tribes to continue to manage cyanide in waters to the criteria concentration. This approval could adversely affect delta smelt critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of delta smelt, and cause delta smelt to experience adverse effects to growth swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 48% and the reduction in the survival of young fish through the first year as high as 56%. These effects are estimated to be of a magnitude great enough to reduce numbers of delta smelt. Continued approval of the CCC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the delta smelt's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for the conservation of the delta smelt.

Percidae

SLACKWATER DARTER

Etheostoma boschungii

Slackwater darters exposed to cyanide at the CCC are likely to experience reduced survival, reproduction, growth, swimming performance, condition, and exhibit developmental abnormalities, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Few data are available to estimate the magnitude of effects that could occur. We developed a quantitative estimate of the effects on fecundity and juvenile survival from exposure to cyanide at the CCC (Table 13). Compared to control populations, we estimate slackwater darters exposed to cyanide at the CCC could experience a significant reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 65%. We estimate that slackwater darters exposed to cyanide at the CCC could experience a significant reduction in the survival of young fish

through the first year and that reduction could be as much as, but is not likely to be greater than, 74%.

As noted previously, the juveniles that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and increase the interval between spawning events.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on shorter-lived fish, like darters, than on longer-lived species, such as sturgeon. Slackwater darter females are short-lived (up to 4 years) and reproduce in no more than three years. The slackwater darter has the potential to spawn multiple times per season, but its spawning period is short, about 1 month, and it is not known how many clutches are spawned (Hartup 2005).

Hartup (2005) conducted field research on the slackwater darter and holiday darter (*E. breviostrum*) and developed species-specific estimates for fecundity, adult survival, and population sizes, then used these data to conduct population viability analyses for the two species. Average slackwater darter fecundity was estimated as 92 and 197 eggs, respectively, for one-batch and two-batch fecundity. Based on estimates of adult survival, Hartup (2005) calculated the adult fertility rate would need to average 0.896 for the slackwater darter population to be stable and 0.818 for the holiday darter population to be stable (the fertility rate was defined as the number of female offspring per female aged i per year). Assuming the slackwater darter population was stable, Hartup estimated the average proportion of spawned eggs needing to survive through the first year as 0.019 or 0.009, for one- and two-batch fecundity, respectively. For the holiday darter, the two-batch estimate was also 0.009. An elasticity analysis for the slackwater darter and holiday darter identified fertilities (defined as fecundity plus juvenile survival) as having the largest relative contribution to population growth rates for each species, as opposed to adult survival (Hartup 2005).

The reductions we estimate in hatched eggs and survival of young fish through the first year would reduce Hartup's (2005) estimated slackwater darter fertility (0.896) substantially. Incorporating our estimates, the revised fertility rates would be reduced substantially, as follows: based upon (a) reductions in numbers of hatched eggs (0.31), (b) reductions in young fish surviving through the first year (0.23), and (c) the additive effect of reduced fecundity and survival (0.07). These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the slackwater darter's reproduction by reducing the number of eggs females spawn, reducing the hatchability of spawned eggs, and reducing the survival of young fish through the first year. Slackwater darters may also experience effects on growth, swimming performance, condition, and development. Based upon our estimates of the magnitude of effects, we would not expect a reduction in density-dependence, if any, to compensate for the reductions in fecundity and juvenile survivorship. We would anticipate a consequent reduction in numbers of slackwater darters. An effected slackwater darter population's growth rate could stabilize at a reduced absolute population number, or the population could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude slackwater darters could become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. The proposed chronic criterion could reduce the reproduction, numbers, and distribution of the slackwater darter.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the slackwater darter include water of sufficient quality for species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience individual and population growth. Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to these levels. This approval could adversely affect the quality of water to the degree that it would impair individual reproduction and survival of slackwater darters, and cause darters to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high 65% and the reduction in the survival of young fish through the first year as high as 74%. These effects are estimated to be of a magnitude great enough to reduce numbers of slackwater darters. Approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the slackwater darter.

VERMILION DARTER

Etheostoma chermocki

Vermilion darters exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we

estimate vermilion darters exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 65%. We estimate that vermilion darters exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 74%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like darters, than on species with greater adult survival, such as sturgeon. Comprehensive studies of vermilion darter life history have not been completed, but it is believed that its life history attributes are similar to those of other snubnose darters, which reach sexual maturity at 1 year of age. Most darters are short-lived (up to 4 years) and reproduce in no more than three years, and spawning occurs in spring.

Hartup (2005) conducted field research on the slackwater darter and holiday darter (*E. brevirostrum*) and developed species-specific estimates for fecundity, adult survival, and population sizes, then used these data to perform population viability analyses for the two species. Average slackwater darter annual fecundity was estimated as 92 and 197 eggs spawned, respectively, for one-batch and two-batch fecundity. Two-batch fecundity for the holiday darter was averaged 175 eggs spawned. Based on estimates of adult survival, Hartup (2005) estimated the adult fertility rate (number of female offspring surviving to year 1) would need to average 0.896 for the slackwater darter population to be stable and 0.818 for the holiday darter population to be stable. Assuming the slackwater darter population was stable, Hartup estimated the average proportion of spawned eggs needing to survive through the first year as 0.019 or 0.009, for one- and two-batch fecundity, respectively. For the holiday darter, the two-batch estimate was also 0.009. An elasticity analysis for the slackwater darter and holiday darter (*E. brevirostrum*) identified fertilities (defined as fecundity plus juvenile survival) as having the largest relative contribution to population growth rates for each species, as opposed to adult survival (Hartup 2005).

Since there are no data on vermilion darters, we infer the vermilion darter's fecundity and survival are no greater than those estimated for the slackwater darter and holiday darter. Consequently, the reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the vermilion darter's fertility substantially. The vermilion darter's fertility rates would be reduced based upon (a) reductions in numbers

of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the vermilion darter's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Vermilion darters may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected vermilion darter population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately vermilion darters are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the vermilion darter.

RELICT DARTER

Etheostoma chienense

Relict darters exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate relict darters exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 65%. We estimate that relict darters exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 74%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors,

including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the CCC. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like darters, than on species with greater adult survival, such as sturgeon. We have no information on the relict darter's lifespan, survival, or fecundity. We assume the relict darter is like other darters: females are short-lived (up to 4 years) and reproduce in no more than three years, and spawning occurs in spring.

Hartup (2005) conducted field research on the slackwater darter and holiday darter (*E. brevirostrum*) and developed species-specific estimates for fecundity, adult survival, and population sizes, then used these data to perform population viability analyses for the two species. Average slackwater darter annual fecundity was estimated as 92 and 197 eggs spawned, respectively, for one-batch and two-batch fecundity. Two-batch fecundity for the holiday darter was averaged 175 eggs spawned. Based on estimates of adult survival, Hartup (2005) estimated the adult fertility rate (number of female offspring surviving to year 1) would need to average 0.896 for the slackwater darter population to be stable and 0.818 for the holiday darter population to be stable. Assuming the slackwater darter population was stable, Hartup estimated the average proportion of spawned eggs needing to survive through the first year as 0.019 or 0.009, for one- and two-batch fecundity, respectively. For the holiday darter, the two-batch estimate was also 0.009. An elasticity analysis for the slackwater darter and holiday darter (*E. brevirostrum*) identified fertilities (defined as fecundity plus juvenile survival) as having the largest relative contribution to population growth rates for each species, as opposed to adult survival (Hartup 2005).

Since there are no data on relict darters, we infer the relict darter's fecundity and survival are no greater than those estimated for the slackwater darter and holiday darter. Consequently, the reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the relict darter's fertility substantially. The relict darter's fertility rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the relict darter's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the

survivorship of young fish in their first year. Relict darters may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected relict darter population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately relict darters are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the relict darter.

ETOWAH DARTER

Etheostoma etowahae

Etowah darters exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Etowah darters exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 65%. We estimate that Etowah darters exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 74%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like darters, than on species with greater adult survival, such as sturgeon. We have no information on the Etowah darter's lifespan, survival, or fecundity. We assume the Etowah darter is like

other darters: females are short-lived (up to 4 years) and reproduce in no more than three years, and spawning occurs in spring.

Hartup (2005) conducted field research on the slackwater darter and holiday darter (*E. brevirostrum*) and developed species-specific estimates for fecundity, adult survival, and population sizes, then used these data to perform population viability analyses for the two species. Average slackwater darter annual fecundity was estimated as 92 and 197 eggs spawned, respectively, for one-batch and two-batch fecundity. Two-batch fecundity for the holiday darter was averaged 175 eggs spawned. Based on estimates of adult survival, Hartup (2005) estimated the adult fertility rate (number of female offspring surviving to year 1) would need to average 0.896 for the slackwater darter population to be stable and 0.818 for the holiday darter population to be stable. Assuming the slackwater darter population was stable, Hartup estimated the average proportion of spawned eggs needing to survive through the first year as 0.019 or 0.009, for one- and two-batch fecundity, respectively. For the holiday darter, the two-batch estimate was also 0.009. An elasticity analysis for the slackwater darter and holiday darter (*E. brevirostrum*) identified fertilities (defined as fecundity plus juvenile survival) as having the largest relative contribution to population growth rates for each species, as opposed to adult survival (Hartup 2005).

Since there are no data on Etowah darters, we infer the Etowah darter's fecundity and survival are no greater than those estimated for the slackwater darter and holiday darter. Consequently, the reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the Etowah darter's fertility substantially. The Etowah darter's fertility rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Etowah darter's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Etowah darters may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Etowah darter population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Etowah darters are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Etowah darter.

FOUNTAIN DARTER

Etheostoma fonticola

Fountain darters exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column. Exposure of fountain darters to cyanide at the criterion maximum concentration (CMC) is likely to reduce survival.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate fountain darters exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 81%. We estimate that fountain darters exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 86%. In addition, we estimate that fountain darters exposed to cyanide at the CMC are likely to experience a substantial reduction in survival and that reduction could be as much as, but is not likely to be greater than, 58.2%. Effects of cyanide at the CMC on the survival of other fountain darter life stages is expected to be of lesser magnitude.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like fountain darters, than on species with greater adult survival, such as sturgeon. Fountain darter females are short-lived (1-2 years), mature early, and spawn year-round with two peaks in August and late winter. In the San Marcos River, mature ova were collected from individuals about 3.5 months old (Linam et al. 1993). Bonner et al. (1998) reported mean egg production (± 1 SD) is 760 (± 310) per two breeding pairs during a 33-day period at 23 degrees C, under laboratory conditions. Annual fecundity is probably substantial.

Hartup (2005) conducted field research on the slackwater darter and holiday darter (*E. brevirostrum*) and developed species-specific estimates for fecundity, adult survival, and population sizes, then used these data to perform population viability analyses for the two

species. Average slackwater darter annual fecundity was estimated as 92 and 197 eggs spawned, respectively, for one-batch and two-batch fecundity. Two-batch fecundity for the holiday darter was averaged 175 eggs spawned. Based on estimates of adult survival, Hartup (2005) estimated the adult fertility rate (number of female offspring surviving to year 1) would need to average 0.896 for the slackwater darter population to be stable and 0.818 for the holiday darter population to be stable. Assuming the slackwater darter population was stable, Hartup estimated the average proportion of spawned eggs needing to survive through the first year as 0.019 or 0.009, for one- and two-batch fecundity, respectively. For the holiday darter, the two-batch estimate was also 0.009. An elasticity analysis for the slackwater darter and holiday darter (*E. brevirostrum*) identified fertilities (defined as fecundity plus juvenile survival) as having the largest relative contribution to population growth rates for each species, as opposed to adult survival (Hartup 2005).

The fountain darter is shorter-lived but more fecund than the slackwater darter and holiday darter. Nevertheless, we anticipate the fountain darter's fertility would be reduced substantially. The fountain darter's fertility rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the fountain darter's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Fountain darters may also experience effects on growth, swimming performance, condition, and development. Exposure to cyanide at the acute criterion could also substantially reduce the survivorship of juvenile fountain darters. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected fountain darter population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately fountain darters are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the acute and chronic criteria could reduce the reproduction, numbers, and distribution of the fountain darter.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the fountain darter include adequate flows, undisturbed substrate, aquatic vegetation including filamentous green algae, and water quality. Approval of the CCC and CMC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentrations. This approval could adversely affect fountain darter critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of fountain darters, and cause fountain darters to experience adverse effects to growth, swimming performance,

condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 31% and the reduction in the survival of young fish through the first year as high as 33%. Reduction in survivorship of juvenile fish exposed at the CMC could be as high as 58.2%. These effects are estimated to be of a magnitude great enough to reduce numbers of fountain darter. Continued approval of the CCC and CMC could adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the fountain darter's extirpation from critical habitat containing cyanide at the CCC and CMC. Impacts to water quality resulting from management of cyanide to the CCC and CMC would diminish the ability of critical habitat to provide for the conservation of the fountain darter.

NIANGUA DARTER

Etheostoma nianguae

Niangua darters exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Niangua darters exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 65%. We estimate that Niangua darters exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 74%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like Niangua darters, than on species with greater adult survival, such as sturgeon. The Niangua darter

reaches sexual maturity at 1 year of age and can live up to 4 years. Estimates of fecundity are based upon counts of mature ova from collected fish: the number of mature ova averaged 189.8 for four females of age-group I, 387.5 for two females of age-group II. A female of age-group IV had 748 mature eggs.

Niangua darter females are short-lived (up to 4 years) and reproduce in no more than three years. The Niangua darter spawns from March to June.

Hartup (2005) conducted field research on the slackwater darter and holiday darter (*E. brevirostrum*) and developed species-specific estimates for fecundity, adult survival, and population sizes, then used these data to perform population viability analyses for the two species. Average slackwater darter annual fecundity was estimated as 92 and 197 eggs spawned, respectively, for one-batch and two-batch fecundity. Two-batch fecundity for the holiday darter was averaged 175 eggs spawned. Based on estimates of adult survival, Hartup (2005) estimated the adult fertility rate (number of female offspring surviving to year 1) would need to average 0.896 for the slackwater darter population to be stable and 0.818 for the holiday darter population to be stable. Assuming the slackwater darter population was stable, Hartup estimated the average proportion of spawned eggs needing to survive through the first year as 0.019 or 0.009, for one- and two-batch fecundity, respectively. For the holiday darter, the two-batch estimate was also 0.009. An elasticity analysis for the slackwater darter and holiday darter (*E. brevirostrum*) identified fertilities (defined as fecundity plus juvenile survival) as having the largest relative contribution to population growth rates for each species, as opposed to adult survival (Hartup 2005).

The Niangua darter's fecundity is somewhat greater than the slackwater darter's and holiday darter's fecundity and their lifespans are similar. The reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the Niangua darter's fertility substantially. The Niangua darter's fertility rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Niangua darter's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Niangua darters may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Niangua darter population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Niangua darters are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of

the chronic criterion could reduce the reproduction, numbers, and distribution of the Niangua darter.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the Niangua darter include water of sufficient quality for species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience individual and population growth. Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to these levels. This approval could adversely affect the quality of water to the degree that it would impair individual reproduction and survival of Niangua darters, and cause darters to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high 65% and the reduction in the survival of young fish through the first year as high as 74%. These effects are estimated to be of a magnitude great enough to reduce numbers of Niangua darters. Approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the Niangua darter.

WATERCRESS DARTER

Etheostoma nuchale

Watercress darters exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate watercress darters exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 65%. We estimate that watercress darters exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 74%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like darters, than on species with greater adult survival, such as sturgeon. We have no information on the watercress darter's lifespan, survival, or fecundity. We assume the relict darter is like other darters: females are short-lived (up to 4 years) and reproduce in no more than three years, and spawning occurs in spring.

Hartup (2005) conducted field research on the slackwater darter and holiday darter (*E. brevirostrum*) and developed species-specific estimates for fecundity, adult survival, and population sizes, then used these data to perform population viability analyses for the two species. Average slackwater darter annual fecundity was estimated as 92 and 197 eggs spawned, respectively, for one-batch and two-batch fecundity. Two-batch fecundity for the holiday darter was averaged 175 eggs spawned. Based on estimates of adult survival, Hartup (2005) estimated the adult fertility rate (number of female offspring surviving to year 1) would need to average 0.896 for the slackwater darter population to be stable and 0.818 for the holiday darter population to be stable. Assuming the slackwater darter population was stable, Hartup estimated the average proportion of spawned eggs needing to survive through the first year as 0.019 or 0.009, for one- and two-batch fecundity, respectively. For the holiday darter, the two-batch estimate was also 0.009. An elasticity analysis for the slackwater darter and holiday darter (*E. brevirostrum*) identified fertilities (defined as fecundity plus juvenile survival) as having the largest relative contribution to population growth rates for each species, as opposed to adult survival (Hartup 2005).

Since there are no data on watercress darters, we infer the watercress darter's fecundity and survival are no greater than those estimated for the slackwater darter and holiday darter. Consequently, the reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the watercress darter's fertility substantially. The watercress darter's fertility rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the watercress darter's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Watercress darters may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory

mechanisms, if they exist, to be overwhelmed. An effected watercress darter population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately watercress darters are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the watercress darter.

OKALOOSA DARTER

Etheostoma okaloosae

Okaloosa darters exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Okaloosa darters exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 65%. We estimate that Okaloosa darters exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 74%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like darters, than on species with greater adult survival, such as sturgeon. Fecundity of Okaloosa darters is very low - mean mature ova were only 29 (Ogilvie 1980).

Hartup (2005) conducted field research on the slackwater darter and holiday darter (*E. brevirostrum*) and developed species-specific estimates for fecundity, adult survival, and population sizes, then used these data to perform population viability analyses for the two species. Average slackwater darter annual fecundity was estimated as 92 and 197 eggs spawned, respectively, for one-batch and two-batch fecundity. Two-batch fecundity for the holiday darter was averaged 175 eggs spawned. Based on estimates of adult survival, Hartup (2005) estimated the adult fertility rate (number of female offspring surviving to year 1) would need to average 0.896 for the slackwater darter population to be stable and 0.818 for the holiday darter population to be stable. Assuming the slackwater darter population was stable, Hartup estimated the average proportion of spawned eggs needing to survive through the first year as 0.019 or 0.009, for one- and two-batch fecundity, respectively. For the holiday darter, the two-batch estimate was also 0.009. An elasticity 1 analysis for the slackwater darter and holiday darter (*E. brevirostrum*) identified fertilities (defined as fecundity plus juvenile survival) as having the largest relative contribution to population growth rates for each species, as opposed to adult survival (Hartup 2005).

The Okaloosa darter's fecundity is much less than the slackwater darter's and holiday darter's fecundity. The reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the Okaloosa darter's fertility substantially. The Okaloosa darter's fertility rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Okaloosa darter's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Okaloosa darters may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Okaloosa darter population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Okaloosa darters are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Okaloosa darter.

DUSKYTAIL DARTER
Etheostoma percnurum

Duskytail darters exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate duskytail darters exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 65%. We estimate that duskytail darters exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 74%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like duskytail darters, than on species with greater adult survival, such as sturgeon. Duskytail darter adult survival is very low. A very small percentage of the population survives to age 3. Duskytail darters can spawn as 1-year-olds. Spawning frequency ranged from 5-7 clutches per year, and fecundity from 135-189 (Layman 1991)

Hartup (2005) conducted field research on the slackwater darter and holiday darter (*E. breviostrum*) and developed species-specific estimates for fecundity, adult survival, and population sizes, then used these data to perform population viability analyses for the two species. Average slackwater darter annual fecundity was estimated as 92 and 197 eggs spawned, respectively, for one-batch and two-batch fecundity. Two-batch fecundity for the holiday darter was averaged 175 eggs spawned. Based on estimates of adult survival, Hartup (2005) estimated the adult fertility rate (number of female offspring surviving to year 1) would need to average 0.896 for the slackwater darter population to be stable and 0.818 for the holiday darter population to be stable. Assuming the slackwater darter population was stable, Hartup estimated the average proportion of spawned eggs needing to survive through the first year as 0.019 or 0.009, for one- and two-batch fecundity,

respectively. For the holiday darter, the two-batch estimate was also 0.009. An elasticity analysis for the slackwater darter and holiday darter (*E. brevirostrum*) identified fertilities (defined as fecundity plus juvenile survival) as having the largest relative contribution to population growth rates for each species, as opposed to adult survival (Hartup 2005).

The duskytail darter's fecundity and survival approximates or is somewhat less than the slackwater darter's and holiday darter's fecundity and survival. Consequently, the reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the duskytail darter's fertility substantially. The duskytail darter's fertility rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the duskytail darter's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Duskytail darters may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected duskytail darter population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately duskytail darters are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the duskytail darter.

BAYOU DARTER

Etheostoma rubrum

Bayou darters exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate bayou darters exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 65%. We estimate that bayou darters exposed to cyanide at

the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 74%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like bayou darters, than on species with greater adult survival, such as sturgeon. Most Bayou darters start spawning after their first year and do not live beyond the age of three. Depending on the size of the female, clutches can range from 20-75 eggs. A single female likely spawns at least twice per reproductive season based on the size classes of ova.

Bayou darter females are short-lived (up to 3 years) and reproduce in no more than three years. The bayou darter spawns from March to June. Hartup (2005) conducted field research on the slackwater darter and holiday darter (*E. brevirostrum*) and developed species-specific estimates for fecundity, adult survival, and population sizes, then used these data to perform population viability analyses for the two species. Average slackwater darter annual fecundity was estimated as 92 and 197 eggs spawned, respectively, for one-batch and two-batch fecundity. Two-batch fecundity for the holiday darter was averaged 175 eggs spawned. Based on estimates of adult survival, Hartup (2005) estimated the adult fertility rate (number of female offspring surviving to year 1) would need to average 0.896 for the slackwater darter population to be stable and 0.818 for the holiday darter population to be stable. Assuming the slackwater darter population was stable, Hartup estimated the average proportion of spawned eggs needing to survive through the first year as 0.019 or 0.009, for one- and two-batch fecundity, respectively. For the holiday darter, the two-batch estimate was also 0.009. An elasticity analysis for the slackwater darter and holiday darter (*E. brevirostrum*) identified fertilities (defined as fecundity plus juvenile survival) as having the largest relative contribution to population growth rates for each species, as opposed to adult survival (Hartup 2005).

The bayou darter's fecundity approximates or is slightly less than the slackwater and holiday darter's. The reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the bayou darter's fertility substantially. The bayou darter's fertility would be diminished because of (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less

sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the bayou darter's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Bayou darters may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected bayou darter population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately bayou darters are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the bayou darter.

CHEROKEE DARTER

Etheostoma scotti

Cherokee darters exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Cherokee darters exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 65%. We estimate that Cherokee darters exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 74%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration.

Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like Cherokee darters, than on species with greater adult survival, such as sturgeon. We have no information on the Cherokee darter's lifespan, survival, or fecundity. We assume the Cherokee darter is like other darters: females are short-lived (up to 4 years) and reproduce in no more than three years, and spawning occurs in spring.

Hartup (2005) conducted field research on the slackwater darter and holiday darter (*E. brevirostrum*) and developed species-specific estimates for fecundity, adult survival, and population sizes, then used these data to perform population viability analyses for the two species. Average slackwater darter annual fecundity was estimated as 92 and 197 eggs spawned, respectively, for one-batch and two-batch fecundity. Two-batch fecundity for the holiday darter was averaged 175 eggs spawned. Based on estimates of adult survival, Hartup (2005) estimated the adult fertility rate (number of female offspring surviving to year 1) would need to average 0.896 for the slackwater darter population to be stable and 0.818 for the holiday darter population to be stable. Assuming the slackwater darter population was stable, Hartup estimated the average proportion of spawned eggs needing to survive through the first year as 0.019 or 0.009, for one- and two-batch fecundity, respectively. For the holiday darter, the two-batch estimate was also 0.009. An elasticity analysis for the slackwater darter and holiday darter (*E. brevirostrum*) identified fertilities (defined as fecundity plus juvenile survival) as having the largest relative contribution to population growth rates for each species, as opposed to adult survival (Hartup 2005).

Since there are no data on Cherokee darters, we infer the Cherokee darter's fecundity and survival are no greater than those estimated for the slackwater darter and holiday darter. Consequently, the reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the Cherokee darter's fertility substantially. The Cherokee darter's fertility rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Cherokee darter's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Cherokee darters may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Cherokee darter population's decline could stabilize at a reduced absolute population number or could continue to

decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Cherokee darters are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Cherokee darter.

MARYLAND DARTER

Etheostoma sellare

Maryland darters exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Maryland darters exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 65%. We estimate that Maryland darters exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 74%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like darters, than on species with greater adult survival, such as sturgeon. We have no information on the Maryland darter's lifespan, survival, or fecundity. We assume the Maryland darter is like other darters: females are short-lived (up to 4 years) and reproduce in no more than three years, and spawning occurs in spring.

Hartup (2005) conducted field research on the slackwater darter and holiday darter (*E. brevirostrum*) and developed species-specific estimates for fecundity, adult survival, and

population sizes, then used these data to perform population viability analyses for the two species. Average slackwater darter annual fecundity was estimated as 92 and 197 eggs spawned, respectively, for one-batch and two-batch fecundity. Two-batch fecundity for the holiday darter was averaged 175 eggs spawned. Based on estimates of adult survival, Hartup (2005) estimated the adult fertility rate (number of female offspring surviving to year 1) would need to average 0.896 for the slackwater darter population to be stable and 0.818 for the holiday darter population to be stable. Assuming the slackwater darter population was stable, Hartup estimated the average proportion of spawned eggs needing to survive through the first year as 0.019 or 0.009, for one- and two-batch fecundity, respectively. For the holiday darter, the two-batch estimate was also 0.009. An elasticity analysis for the slackwater darter and holiday darter (*E. brevirostrum*) identified fertilities (defined as fecundity plus juvenile survival) as having the largest relative contribution to population growth rates for each species, as opposed to adult survival (Hartup 2005).

Since there are no data on Maryland darters, we infer the Maryland darter's fecundity and survival are no greater than those estimated for the slackwater darter and holiday darter. Consequently, the reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the Maryland darter's fertility substantially. The Maryland darter's fertility rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Maryland darter's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Maryland darters may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Maryland darter population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Maryland darters are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Maryland darter.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the Maryland darter include water of sufficient quality for species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience individual and population growth. Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to these levels. This approval could adversely affect the quality of water to the degree that it would impair

individual reproduction and survival of Maryland darters, and cause darters to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high 65% and the reduction in the survival of young fish through the first year as high as 74%. These effects are estimated to be of a magnitude great enough to reduce numbers of Maryland darters. Approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the Maryland darter.

BLUEMASK DARTER

Etheostoma sp.

Bluemask darters exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate bluemask darters exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 65%. We estimate that bluemask darters exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 74%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like darters, than on species with greater adult survival, such as sturgeon. We have no information on the bluemask darter's lifespan, survival, or fecundity. We assume the bluemask darter is

like other darters: females are short-lived (up to 4 years) and reproduce in no more than three years, and spawning occurs in spring.

Hartup (2005) conducted field research on the slackwater darter and holiday darter (*E. brevirostrum*) and developed species-specific estimates for fecundity, adult survival, and population sizes, then used these data to perform population viability analyses for the two species. Average slackwater darter annual fecundity was estimated as 92 and 197 eggs spawned, respectively, for one-batch and two-batch fecundity. Two-batch fecundity for the holiday darter was averaged 175 eggs spawned. Based on estimates of adult survival, Hartup (2005) estimated the adult fertility rate (number of female offspring surviving to year 1) would need to average 0.896 for the slackwater darter population to be stable and 0.818 for the holiday darter population to be stable. Assuming the slackwater darter population was stable, Hartup estimated the average proportion of spawned eggs needing to survive through the first year as 0.019 or 0.009, for one- and two-batch fecundity, respectively. For the holiday darter, the two-batch estimate was also 0.009. An elasticity analysis for the slackwater darter and holiday darter (*E. brevirostrum*) identified fertilities (defined as fecundity plus juvenile survival) as having the largest relative contribution to population growth rates for each species, as opposed to adult survival (Hartup 2005).

Since there are no data on bluemask darters, we infer the bluemask darter's fecundity and survival are no greater than those estimated for the slackwater darter and holiday darter. Consequently, the reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the bluemask darter's fertility substantially. The bluemask darter's fertility rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the bluemask darter's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Bluemask darters may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected bluemask darter population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately bluemask darters are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the bluemask darter.

BOULDER DARTER

Etheostoma wapiti

Boulder darters exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate boulder darters exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 59%. We estimate that boulder darters exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 74%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like boulder darters, than on species with greater adult survival, such as sturgeon. No life history studies have been conducted on this species. We assume the boulder darter is like other darters: females are short-lived (up to 4 years) and reproduce in no more than three years, and spawning occurs in spring.

Hartup (2005) conducted field research on the slackwater darter and holiday darter (*E. brevirostrum*) and developed species-specific estimates for fecundity, adult survival, and population sizes, then used these data to perform population viability analyses for the two species. Average slackwater darter annual fecundity was estimated as 92 and 197 eggs spawned, respectively, for one-batch and two-batch fecundity. Two-batch fecundity for the holiday darter was averaged 175 eggs spawned. Based on estimates of adult survival, Hartup (2005) estimated the adult fertility rate (number of female offspring surviving to year 1) would need to average 0.896 for the slackwater darter population to be stable and 0.818 for the holiday darter population to be stable. Assuming the slackwater darter

population was stable, Hartup estimated the average proportion of spawned eggs needing to survive through the first year as 0.019 or 0.009, for one- and two-batch fecundity, respectively. For the holiday darter, the two-batch estimate was also 0.009. An elasticity analysis for the slackwater darter and holiday darter (*E. brevirostrum*) identified fertilities (defined as fecundity plus juvenile survival) as having the largest relative contribution to population growth rates for each species, as opposed to adult survival (Hartup 2005).

Since there are no data on boulder darters, we infer the boulder darter's fecundity and survival are no greater than those estimated for the slackwater darter and holiday darter. Consequently, the reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the boulder darter's fertility substantially. The boulder darter's fertility rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the boulder darter's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Boulder darters may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected boulder darter population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately boulder darters are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the boulder darter.

AMBER DARTER

Percina antesella

Amber darters exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success,

and survival of young first-year fish (Table 13). Compared to control populations, we estimate amber darters exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 63%. We estimate that amber darters exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 72%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species, like amber darters, that have fewer opportunities to spawn than on species with greater adult survival, such as sturgeon. Amber darter females are short-lived (approximately 3 years) and probably reproduce in only two years. The amber darter spawns from late fall to early spring.

Hartup (2005) conducted field research on the slackwater darter and holiday darter (*E. brevirostrum*) and developed species-specific estimates for fecundity, adult survival, and population sizes, then used these data to perform population viability analyses for the two species. Average slackwater darter annual fecundity was estimated as 92 and 197 eggs spawned, respectively, for one-batch and two-batch fecundity. Two-batch fecundity for the holiday darter was averaged 175 eggs spawned. Based on estimates of adult survival, Hartup (2005) estimated the adult fertility rate (number of female offspring surviving to year 1) would need to average 0.896 for the slackwater darter population to be stable and 0.818 for the holiday darter population to be stable. Assuming the slackwater darter population was stable, Hartup estimated the average proportion of spawned eggs needing to survive through the first year as 0.019 or 0.009, for one- and two-batch fecundity, respectively. For the holiday darter, the two-batch estimate was also 0.009. An elasticity analysis for the slackwater darter and holiday darter (*E. brevirostrum*) identified fertilities (defined as fecundity plus juvenile survival) as having the largest relative contribution to population growth rates for each species, as opposed to adult survival (Hartup 2005).

There are no data on the fecundity and survival of amber darters. We infer the amber darter's fecundity and survival are no greater than those estimated for the slackwater darter and holiday darter. Consequently, the reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the amber darter's fertility substantially. The amber darter's fertility rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the

first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the amber darter's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Amber darters may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected amber darter population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately amber darters are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the amber darter.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the amber darter include water of sufficient quality for species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience individual and population growth. Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to these levels. This approval could adversely affect the quality of water to the degree that it would impair individual reproduction and survival of amber darters, and cause darters to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high 63% and the reduction in the survival of young fish through the first year as high as 72%. These effects are estimated to be of a magnitude great enough to reduce numbers of amber darters. Approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the amber darter.

GOLDLINE DARTER

Percina aurolineata

Goldline darters exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate goldline darters exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 63%. We estimate that goldline darters exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 72%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like goldline darters, than on species with greater adult survival, such as sturgeon. The life history of the goldline darter is unknown. We assume the goldline darter is like other darters: females are short-lived (up to 4 years) and reproduce in no more than three years.

Hartup (2005) conducted field research on the slackwater darter and holiday darter (*E. brevirostrum*) and developed species-specific estimates for fecundity, adult survival, and population sizes, then used these data to perform population viability analyses for the two species. Average slackwater darter annual fecundity was estimated as 92 and 197 eggs spawned, respectively, for one-batch and two-batch fecundity. Two-batch fecundity for the holiday darter was averaged 175 eggs spawned. Based on estimates of adult survival, Hartup (2005) estimated the adult fertility rate (number of female offspring surviving to year 1) would need to average 0.896 for the slackwater darter population to be stable and 0.818 for the holiday darter population to be stable. Assuming the slackwater darter population was stable, Hartup estimated the average proportion of spawned eggs needing to survive through the first year as 0.019 or 0.009, for one- and two-batch fecundity, respectively. For the holiday darter, the two-batch estimate was also 0.009. An elasticity analysis for the slackwater darter and holiday darter (*E. brevirostrum*) identified fertilities (defined as fecundity plus juvenile survival) as having the largest relative contribution to population growth rates for each species, as opposed to adult survival (Hartup 2005).

There are no data on the fecundity and survival of goldline darters. We assume the goldline darter's fecundity and survival are no greater than those estimated for the

slackwater darter and holiday darter. Consequently, the reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the amber darter's fertility substantially. The goldline darter's fertility rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the goldline darter's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Goldline darters may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected goldline darter population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately goldline darters are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the goldline darter.

CONASAUGA LOGPERCH

Percina jenkinsi

Conasauga logperch exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Conasauga logperch exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 63%. We estimate that Conasauga logperch exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 72%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like *Conasauga logperch*, than on species with greater adult survival, such as sturgeon. *Conasauga logperch* females are short-lived (probably up to 4 years) and probably reproduce in no more than three years. The *Conasauga logperch*'s spawning is probably limited to spring.

Hartup (2005) conducted field research on the slackwater darter and holiday darter (*E. brevirostrum*) and developed species-specific estimates for fecundity, adult survival, and population sizes, then used these data to perform population viability analyses for the two species. Average slackwater darter annual fecundity was estimated as 92 and 197 eggs spawned, respectively, for one-batch and two-batch fecundity. Two-batch fecundity for the holiday darter was averaged 175 eggs spawned. Based on estimates of adult survival, Hartup (2005) estimated the adult fertility rate (number of female offspring surviving to year 1) would need to average 0.896 for the slackwater darter population to be stable and 0.818 for the holiday darter population to be stable. Assuming the slackwater darter population was stable, Hartup estimated the average proportion of spawned eggs needing to survive through the first year as 0.019 or 0.009, for one- and two-batch fecundity, respectively. For the holiday darter, the two-batch estimate was also 0.009. An elasticity analysis for the slackwater darter and holiday darter (*E. brevirostrum*) identified fertilities (defined as fecundity plus juvenile survival) as having the largest relative contribution to population growth rates for each species, as opposed to adult survival (Hartup 2005). There are no data on the fecundity and survival of *Conasauga logperch*. We infer the *Conasauga logperch*'s fecundity and survival are no greater than those estimated for the slackwater darter and holiday darter. Consequently, the reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the *Conasauga logperch*'s fertility substantially. The *Conasauga logperch*'s fertility rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the *Conasauga logperch*'s reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the

survivorship of young fish in their first year. Conasauga logperch may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Conasauga logperch population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Conasauga logperch are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Conasauga logperch.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the Conasauga logperch include water of sufficient quality for species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience individual and population growth. Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to these levels. This approval could adversely affect the quality of water to the degree that it would impair individual reproduction and survival of Conasauga logperch, and cause logperch to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high 63% and the reduction in the survival of young fish through the first year as high as 72%. These effects are estimated to be of a magnitude great enough to reduce numbers of Conasauga logperch. Approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the Conasauga logperch.

LEOPARD DARTER

Percina pantherina

Leopard darter exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate leopard darter exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 63%. We estimate that leopard darter exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through

the first year and that reduction could be as much as, but is not likely to be greater than, 72%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like leopard darter, than on species with greater adult survival, such as sturgeon. Leopard darter females are short-lived (up to 4 years) and reproduce in no more than three years. The leopard darter spawns from March to April.

Hartup (2005) conducted field research on the slackwater darter and holiday darter (*E. brevirostrum*) and developed species-specific estimates for fecundity, adult survival, and population sizes, then used these data to perform population viability analyses for the two species. Average slackwater darter annual fecundity was estimated as 92 and 197 eggs spawned, respectively, for one-batch and two-batch fecundity. Two-batch fecundity for the holiday darter was averaged 175 eggs spawned. Based on estimates of adult survival, Hartup (2005) estimated the adult fertility rate (number of female offspring surviving to year 1) would need to average 0.896 for the slackwater darter population to be stable and 0.818 for the holiday darter population to be stable. Assuming the slackwater darter population was stable, Hartup estimated the average proportion of spawned eggs needing to survive through the first year as 0.019 or 0.009, for one- and two-batch fecundity, respectively. For the holiday darter, the two-batch estimate was also 0.009. An elasticity analysis for the slackwater darter and holiday darter (*E. brevirostrum*) identified fertilities (defined as fecundity plus juvenile survival) as having the largest relative contribution to population growth rates for each species, as opposed to adult survival (Hartup 2005).

There is limited data on the fecundity of leopard darters and no information on survival. Robison's (1978) and Hartup's (2005) estimates of fecundity are not directly comparable because their methods differed. Nevertheless, the leopard darter's life history is similar to that of other darters, including the slackwater and holiday darter. The reductions we estimate in hatched eggs and survival of young fish through the first year would reduce the leopard darter's fertility substantially. The leopard darter's fertility rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in

reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the leopard darter's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Leopard darters may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected leopard darter population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately leopard darter are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the leopard darter.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the leopard darter include water of sufficient quality for species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience individual and population growth. Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to these levels. This approval could adversely affect the quality of water to the degree that it would impair individual reproduction and survival of leopard darter, and cause darters to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high 63% and the reduction in the survival of young fish through the first year as high as 72%. These effects are estimated to be of a magnitude great enough to reduce numbers of leopard darter. Approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the leopard darter.

ROANOKE LOGPERCH

Percina rex

Roanoke logperch exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were

available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Roanoke logperch exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 63%. We estimate that Roanoke logperch exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 72%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like most darters, than on species with greater adult survival, such as sturgeon. Roanoke logperch females live 5-6 years.

Hartup (2005) conducted field research on the slackwater darter and holiday darter (*E. brevirostrum*) and developed species-specific estimates for fecundity, adult survival, and population sizes, then used these data to perform population viability analyses for the two species. Average slackwater darter annual fecundity was estimated as 92 and 197 eggs spawned, respectively, for one-batch and two-batch fecundity. Two-batch fecundity for the holiday darter was averaged 175 eggs spawned. Based on estimates of adult survival, Hartup (2005) estimated the adult fertility rate (number of female offspring surviving to year 1) would need to average 0.896 for the slackwater darter population to be stable and 0.818 for the holiday darter population to be stable. Assuming the slackwater darter population was stable, Hartup estimated the average proportion of spawned eggs needing to survive through the first year as 0.019 or 0.009, for one- and two-batch fecundity, respectively. For the holiday darter, the two-batch estimate was also 0.009. An elasticity analysis for the slackwater darter and holiday darter (*E. brevirostrum*) identified fertilities (defined as fecundity plus juvenile survival) as having the largest relative contribution to population growth rates for each species, as opposed to adult survival (Hartup 2005).

The Roanoke logperch is longer-lived and more fecund than the slackwater darter and holiday darter. Nevertheless, we anticipate the Roanoke logperch's fertility would be reduced substantially. The Roanoke logperch's fertility rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of

cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Roanoke logperch's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Roanoke logperch may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Roanoke logperch population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Roanoke logperch are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Roanoke logperch.

SNAIL DARTER

Percina tanasi

Snail darter exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate snail darter exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 63%. We estimate that snail darter exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 72%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration.

Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like snail darter, than on species with greater adult survival, such as sturgeon. Snail darter females are short-lived (up to 4 years) and reproduce in no more than three years. The snail darter spawns in early February through April and average fecundity is about 600 eggs.

Hartup (2005) conducted field research on the slackwater darter and holiday darter (*E. brevirostrum*) and developed species-specific estimates for fecundity, adult survival, and population sizes, then used these data to perform population viability analyses for the two species. Average slackwater darter annual fecundity was estimated as 92 and 197 eggs spawned, respectively, for one-batch and two-batch fecundity. Two-batch fecundity for the holiday darter was averaged 175 eggs spawned. Based on estimates of adult survival, Hartup (2005) estimated the adult fertility rate (number of female offspring surviving to year 1) would need to average 0.896 for the slackwater darter population to be stable and 0.818 for the holiday darter population to be stable. Assuming the slackwater darter population was stable, Hartup estimated the average proportion of spawned eggs needing to survive through the first year as 0.019 or 0.009, for one- and two-batch fecundity, respectively. For the holiday darter, the two-batch estimate was also 0.009. An elasticity analysis for the slackwater darter and holiday darter (*E. brevirostrum*) identified fertilities (defined as fecundity plus juvenile survival) as having the largest relative contribution to population growth rates for each species, as opposed to adult survival (Hartup 2005).

The snail darter has a similar lifespan but is more fecund than the slackwater darter and holiday darter. We anticipate the snail darter's fertility would be reduced substantially, but less so than the slackwater or holiday darter's fertility. The snail darter's fertility rates would be reduced based upon (a) reductions in numbers of hatched eggs, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the snail darter's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. snail darter may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected snail darter population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately snail darter are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of

the chronic criterion could reduce the reproduction, numbers, and distribution of the snail darter.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the snail darter include water of sufficient quality for species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience individual and population growth. Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to these levels. This approval could adversely affect the quality of water to the degree that it would impair individual reproduction and survival of snail darter, and cause darters to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high 63% and the reduction in the survival of young fish through the first year as high as 72%. These effects are estimated to be of a magnitude great enough to reduce numbers of snail darter. Approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the snail darter.

Poeciliidae

BIG BEND GAMBUSIA

Gambusia gaigei

Big Bend gambusia exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity and survival of young first-year fish (Table 13). Compared to control populations, we estimate Big Bend gambusia exposed to cyanide at the CCC could experience a substantial reduction in the number of young produced and that reduction could be as much as, but is not likely to be greater than, 48%. We estimate that Big Bend gambusia exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 56%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

Big Bend gambusia is a live-bearing fish, that is short lived (life expectancy 2 years; U.S. Fish and Wildlife Service 2009), has a prolonged reproductive period (5 to 8 months), and females may produce 50 or more young in the peak season. Such a life history strategy would appear to be highly sensitive to reductions in fecundity and survival of young fish. An elasticity analysis for other short-lived fish species indicates that population growth is, in fact, most influenced by changes in fecundity and juvenile survival (Vélez-Espino et al. 2006). Although gambusia are viviparous, we believe that our estimates of effects on Big Bend gambusia fecundity, which were based on studies with oviparous fish, are applicable. Koyo et al. (2000) described the dynamics of oocyte (egg) and embryonic development in *Gambusia affinis*, the mosquitofish. The process of oocyte development, fertilization, and “hatching” in viviparous and oviparous fishes are comparable, except that fertilization and “hatching” occur internally in viviparous species. Thus, the effects of cyanide on oocyte development described in the *Chronic Toxicity to Fish* section are likely to occur in Gambusia, with similar outcomes. In studies with oviparous fish, only the largest and most mature oocytes were spawned (ovulation) and capable of being fertilized. Similarly, in the viviparous gambusia only the largest and most mature oocytes are capable of being fertilized in the follicle. Thus reductions in the number of mature oocytes, by cyanide, would reduce the number of eggs capable of being fertilized, the number of embryos that are “hatched” (internally), and the number of live young that are “born”. We would also expect predicted effects on juvenile survival to be the same for live-bearing species.

The reductions we estimate in the number of young produced and survival of young fish through the first year would reduce the Big Bend gambusia’s potential recruitment substantially. The Big Bend gambusia’s potential recruitment would be diminished because of (a) reductions in numbers of young produced, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the young that are produced in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Big Bend gambusia’s reproduction by reducing the number of young produced by females, and reducing the survivorship of young fish in their first year. Big Bend gambusia may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Big Bend gambusia population’s decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Big Bend gambusia are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Big Bend gambusia.

SAN MARCOS GAMBUSIA
Gambusia georgei

San Marcos gambusia exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity and survival of young first-year fish (Table 13). Compared to control populations, we estimate San Marcos gambusia exposed to cyanide at the CCC could experience a substantial reduction in the number of young produced and that reduction could be as much as, but is not likely to be greater than, 48%. We estimate that San Marcos gambusia exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 56%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

Little is known about the reproductive capabilities of San Marcos gambusia except that it is a live-bearing fish and two individuals kept in laboratory aquaria produced 12, 30, and 60 young although the largest clutch appeared to have been aborted and did not survive (Edwards et al. 1980). However, more is known about the closely related Big Bend gambusia (*Gambusia gaigei*) which we will use to provide insight into the reproductive traits of San Marcos gambusia. Big Bend gambusia is short lived (life expectancy 2 years; U.S. Fish and Wildlife Service 2009), has a prolonged reproductive period (5 to 8 months), and females may produce 50 or more young in the peak season. Such a life history strategy would appear to be highly sensitive to reductions in fecundity and survival of young fish. An elasticity analysis for other short-lived fish species indicates that population growth is, in fact, most influenced by changes in fecundity and juvenile survival (Vélez-Espino et al. 2006). Although gambusia are viviparous, we believe that our estimates of effects on San Marcos gambusia fecundity, which were based on studies with oviparous fish, are applicable. Koyo et al. (2000) described the dynamics of oocyte (egg) and embryonic development in *Gambusia affinis*, the mosquitofish. The process of oocyte development, fertilization, and “hatching” in viviparous and oviparous fishes are comparable, except that fertilization and “hatching” occur internally in viviparous species. Thus, the effects of cyanide on oocyte development described in the *Chronic Toxicity to Fish* section are likely to occur in Gambusia, with similar outcomes. In

studies with oviparous fish, only the largest and most mature oocytes were spawned (ovulation) and capable of being fertilized. Similarly, in the viviparous gambusia only the largest and most mature oocytes are capable of being fertilized in the follicle. Thus reductions in the number of mature oocytes, by cyanide, would reduce the number of eggs capable of being fertilized, the number of embryos that are “hatched” (internally), and the number of live young that are “born”. We would also expect predicted effects on juvenile survival to be the same for live-bearing species.

The reductions we estimate in the number of young produced and survival of young fish through the first year would reduce the San Marcos gambusia’s potential recruitment substantially. The San Marcos gambusia’s potential recruitment would be diminished because of (a) reductions in numbers of young produced, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the young that are produced in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the San Marcos gambusia’s reproduction by reducing the number of young produced by females, and reducing the survivorship of young fish in their first year. San Marcos gambusia may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected San Marcos gambusia population’s decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately San Marcos gambusia are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the San Marcos gambusia.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the San Marcos gambusia were not identified in the final rule designating critical habitat. We describe them here to include water of sufficient quality for the species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience individual and population growth sufficient for the critical habitat to serve its intended conservation function.

Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to the criterion concentration. This approval could adversely affect San Marcos gambusia critical habitat by diminishing the quality of water to the degree that it would impair individual reproduction and survival of San Marcos gambusia, and cause San Marcos gambusia to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of young produced could be as high 48% and the reduction in the survival of young fish through the first

year as high as 56%. These effects are estimated to be of a magnitude great enough to reduce numbers of San Marcos gambusia. Continued approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in the San Marcos gambusia's extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the San Marcos gambusia.

CLEAR CREEK GAMBUSIA

Gambusia heterochir

Clear Creek gambusia exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity and survival of young first-year fish (Table 13). Compared to control populations, we estimate Clear Creek gambusia exposed to cyanide at the CCC could experience a substantial reduction in the number of young produced and that reduction could be as much as, but is not likely to be greater than, 48%. We estimate that Clear Creek gambusia exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 56%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The Clear Creek gambusia is viviparous and females can produce several broods per year. In Clear Creek, females are reproductive for 7 months (March-September) and all stream reaches inhabited by Clear Creek gambusia have pregnant females during the midsummer reproductive period. In the lab (at 25 C), females produced up to 50 young every 42 days. However, in the cooler waters of Clear Creek (20 C) the estimated interbrood interval is 60 days. It's not clear how long Clear Creek gambusia live, but the closely related Big Bend gambusia (*Gambusia gaigei*) has a life expectancy of 2 years (U.S. Fish and Wildlife Service 2009). If the Clear Creek gambusia has similar longevity, their life history strategy (short-lived with extended reproductive periods and relatively low numbers of young per brood) would appear to be highly sensitive to reductions in fecundity and survival of young fish. An elasticity analysis for other short-lived fish species indicates that population growth is, in fact, most influenced by changes

in fecundity and juvenile survival (Vélez-Espino et al. 2006). Although gambusia are viviparous, we believe that our estimates of effects on Clear Creek gambusia fecundity, which were based on studies with oviparous fish, are applicable. Koyo et al. (2000) described the dynamics of oocyte (egg) and embryonic development in *Gambusia affinis*, the mosquitofish. The process of oocyte development, fertilization, and “hatching” in viviparous and oviparous fishes are comparable, except that fertilization and “hatching” occur internally in viviparous species. Thus, the effects of cyanide on oocyte development described in the *Chronic Toxicity to Fish* section are likely to occur in Gambusia, with similar outcomes. In studies with oviparous fish, only the largest and most mature oocytes were spawned (ovulation) and capable of being fertilized. Similarly, in the viviparous gambusia only the largest and most mature oocytes are capable of being fertilized in the follicle. Thus reductions in the number of mature oocytes, by cyanide, would reduce the number of eggs capable of being fertilized, the number of embryos that are “hatched” (internally), and the number of live young that are “born”. We would also expect predicted effects on juvenile survival to be the same for live-bearing species.

The reductions we estimate in the number of young produced and survival of young fish through the first year would reduce the Clear Creek gambusia’s potential recruitment substantially. The Clear Creek gambusia’s potential recruitment would be diminished because of (a) reductions in numbers of young produced, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the young that are produced in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Clear Creek gambusia’s reproduction by reducing the number of young produced by females, and reducing the survivorship of young fish in their first year. Clear Creek gambusia may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Clear Creek gambusia population’s decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Clear Creek gambusia are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Clear Creek gambusia.

PECOS GAMBUSIA
Gambusia nobilis

Pecos gambusia exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth,

swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity and survival of young first-year fish (Table 13). Compared to control populations, we estimate Pecos gambusia exposed to cyanide at the CCC could experience a substantial reduction in the number of young produced and that reduction could be as much as, but is not likely to be greater than, 48%. We estimate that Pecos gambusia exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 56%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

Pecos gambusia bear live young. Females produce up to 40 young per brood during spring and summer. The interbrood interval for Pecos gambusia is unknown, but other species in the Poeciliidae family typically bear young every 1-2 months while reproductive. It's not clear how long Pecos gambusia live, but the closely related Big Bend gambusia (*Gambusia gaigei*) has a life expectancy of 2 years (U.S. Fish and Wildlife Service 2009). If the Pecos gambusia has similar longevity, their life history strategy (short-lived with extended reproductive periods and relatively low numbers of young per brood) would appear to be highly sensitive to reductions in fecundity and survival of young fish. An elasticity analysis for other short-lived fish species indicates that population growth is, in fact, most influenced by changes in fecundity and juvenile survival (Vélez-Espino et al. 2006). Although gambusia are viviparous, we believe that our estimates of effects on Pecos gambusia fecundity, which were based on studies with oviparous fish, are applicable. Koyo et al. (2000) described the dynamics of oocyte (egg) and embryonic development in *Gambusia affinis*, the mosquitofish. The process of oocyte development, fertilization, and "hatching" in viviparous and oviparous fishes are comparable, except that fertilization and "hatching" occur internally in viviparous species. Thus, the effects of cyanide on oocyte development described in the *Chronic Toxicity to Fish* section are likely to occur in Gambusia, with similar outcomes. In studies with oviparous fish, only the largest and most mature oocytes were spawned (ovulation) and capable of being fertilized. Similarly, in the viviparous gambusia only the largest and most mature oocytes are capable of being fertilized in the follicle. Thus reductions in the number of mature oocytes, by cyanide, would reduce the number of eggs capable of being fertilized, the number of embryos that are "hatched" (internally), and the number of live young that are "born". We would also expect predicted effects on juvenile survival to be the same for live-bearing species.

The reductions we estimate in the number of young produced and survival of young fish through the first year would reduce the Pecos gambusia's potential recruitment substantially. The Pecos gambusia's potential recruitment would be diminished because of (a) reductions in numbers of young produced, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the young that are produced in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates diminish survival through the first winter. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Pecos gambusia's reproduction by reducing the number of young produced by females, and reducing the survivorship of young fish in their first year. Pecos gambusia may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Pecos gambusia population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Pecos gambusia are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Pecos gambusia.

GILA TOPMINNOW (including YAQUI TOPMINNOW)

Poeciliopsis occidentalis

Gila topminnow exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity and survival of young first-year fish (Table 13). Compared to control populations, we estimate Gila topminnow exposed to cyanide at the CCC could experience a substantial reduction in the number of young produced and that reduction could be as much as, but is not likely to be greater than, 48%. We estimate that Gila topminnow exposed to cyanide at the CCC could experience a substantial reduction in the survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 56%.

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As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

Topminnow bear live young and two broods are carried simultaneously (one further along in development than the other). Brood size is from 1 to 20, and gestation time is 24-28 days. Breeding season is from January to August, with some populations capable of breeding all year if temperatures and food availability are suitable. Life span is approximately 1 year; however, this varies with season of birth and fluctuations in environmental conditions in the habitat. Such a life history strategy (short-lived with extended reproductive periods and relatively low numbers of young per brood) would appear to be highly sensitive to reductions in fecundity and survival of young fish. An elasticity analysis for other short-lived fish species indicates that population growth is, in fact, most influenced by changes in fecundity and juvenile survival (Vélez-Espino et al. 2006). Although topminnows are viviparous, we believe that our estimates of effects on Gila topminnow fecundity, which were based on studies with oviparous fish, are applicable. Koyo et al. (2000) described the dynamics of oocyte (egg) and embryonic development in another live-baring species in the Poeciliidae family, *Gambusia affinis* (the mosquitofish). The process of oocyte development, fertilization, and “hatching” in viviparous and oviparous fishes are comparable, except that fertilization and “hatching” occur internally in viviparous species. Thus, the effects of cyanide on oocyte development described in the *Chronic Toxicity to Fish* section are likely to occur in *Gambusia*, with similar outcomes. In studies with oviparous fish, only the largest and most mature oocytes were spawned (ovulation) and capable of being fertilized. Similarly, in the viviparous *Gambusia* only the largest and most mature oocytes are capable of being fertilized in the follicle. Thus reductions in the number of mature oocytes, by cyanide, would reduce the number of eggs capable of being fertilized, the number of embryos that are “hatched” (internally), and the number of live young that are “born”. We would also expect predicted effects on juvenile survival to be the same for live-bearing species.

The reductions we estimate in the number of young produced and survival of young fish through the first year would reduce the Gila topminnow’s potential recruitment substantially. The Gila topminnow’s potential recruitment would be diminished because of (a) reductions in numbers of young produced, (b) reductions in young fish surviving through the first year, and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the young that are produced in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates diminish survival through the first winter. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time needed between batched spawning events.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Gila topminnow’s reproduction by reducing the number of young produced by females, and reducing the survivorship of young fish in their first year. Gila

topminnow may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Gila topminnow population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Gila topminnow are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Gila topminnow.

Salmonidae

BULL TROUT

Salvelinus confluentus

Bull trout exposed to cyanide at the criterion continuous concentration (CCC) are likely to experience reduced survival and reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the *Overview* section. The effects of chronic toxicity could also be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column. Exposure of bull trout to cyanide at the criterion maximum concentration (CMC) is likely to reduce survival.

Relatively few studies are available for estimating the magnitude of effects to the bull trout that could occur following exposure to cyanide at criterion concentrations. However, data are available to develop quantitative estimates of the effects of such exposure on bull trout fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate that bull trout exposed to cyanide at the CCC are likely to experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 87%. We estimate that bull trout exposed to cyanide at the CCC are likely to experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 90%. In addition, we estimate that juvenile bull trout exposed to cyanide at the CMC are likely to experience a substantial reduction in survival and that reduction could be as much as, but is not likely to be greater than, 99.9%. Effects of cyanide at the CMC on the survival of other bull trout life stages is expected to be of lesser magnitude.

Although these effects are based on modeled estimates, direct toxicity tests with other chemicals indicate that bull trout can be very sensitive to the adverse effects of water pollutants. Several studies have compared the sensitivity of bull trout to rainbow trout. Among the species considered in the cyanide criteria document (and in the BE), the rainbow trout was the most sensitive and, thus, was the species on which the acute and chronic cyanide criteria were based. Bull trout have been found to be less sensitive than rainbow trout to some metals (cadmium and zinc; Hansen et al. 2002a), but as sensitive to other metals (copper; Hansen et al. 2002b) and as sensitive or more sensitive to some

herbicides (Fairchild et al. 2006). Dioxin, like cyanide, is a potent reproductive toxin. Cook et al. (2000) reported that, among fish that have been tested, bull trout was the most sensitive to dioxin. The bull trout was three times more sensitive than the lake trout (the next most sensitive species) and more sensitive than the brook trout or the rainbow trout.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation).

Rieman and McIntyre (1993) characterized the demographic and habitat requirements needed for bull trout conservation. As part of their evaluation they considered the consequences of habitat disturbances in terms of their potential impacts on reproduction, survival and extinction risk. Using a population model, they simulated four types of bull trout populations that differ in growth and maturation rates in order to capture the variation exhibited in natural populations, e.g. resident versus migratory life forms. For each simulated population they varied the survival from egg to age one to determine the level of mortality each population could sustain without collapsing. They next calculated the level of survival from egg to fry stage needed to attain those minimum, egg-to-age-one, survival rates. The required egg-to-fry survival rates were highest for slow growth/late maturity populations (0.25 – 0.49) and lowest for the fast growth populations (0.03 - 0.05). Rieman and McIntyre (1993) reported that egg-to-fry survival rates of 0.25 to 0.50 may approach the highest values possible in many streams. For slow growth populations, the egg-to-fry survival rates would have to be at or near the highest possible rates in order to sustain the population. Thus, reductions in egg-to-fry survival caused by exposure to cyanide would result in an egg-to-age-one survival rate that would not sustain the population. For fast growth populations, the egg-to-fry survival rates are lower than the highest possible rates, so these populations may be able to absorb additional mortality at this life stage. However, the large reduction in egg-to-fry survival caused by cyanide (90%), coupled with reductions in the number of eggs spawned, would likely reduce the egg-to-age-one survival rate below the minimum required to sustain the population.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the bull trout's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year by 87 to 90 percent. Bull trout may also experience effects on growth, swimming performance, condition, and development. Exposure to cyanide at the acute criterion could also substantially reduce the survivorship of juvenile bull trout. Because of the high magnitude of these effects, we would expect

density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An affected bull trout population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect that is likely to occur at the chronic criterion level, we conclude that ultimately bull trout are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the acute and chronic criteria at the rangewide scale is likely to reduce the reproduction, numbers, and distribution of the bull trout at the rangewide scale.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the bull trout include water of sufficient quality for species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience individual and population growth. Approval of the CCC and CMC in state water quality standards would authorize states to manage cyanide in waters to these levels. This approval could adversely affect the quality of water to the degree that it would impair individual reproduction and survival of the bull trout, and cause adverse effects to bull trout growth, swimming performance, condition, and development. We estimate that implementation of the proposed action could reduce water quality conditions in critical habitat to the extent that a reduction in the number of hatched eggs could be as high as 87% and a reduction in the survival of young fish through the first year could be as high as 90%. Reduction in survivorship of juvenile fish exposed at the CMC could be as high as 99.9%. These effects are estimated to be of a magnitude great enough to reduce numbers of bull trout using areas of critical habitat that support breeding and rearing areas for the bull trout. Approval of the CCC and CMC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation of affected populations from critical habitat containing cyanide at the CCC and CMC. Impacts to water quality resulting from management of cyanide to the CCC and CMC would diminish the ability of critical habitat to provide for conservation of the bull trout.

LITTLE KERN GOLDEN TROUT

Oncorhynchus aquabonita whitei

Little Kern golden trout exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Little Kern golden trout exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much

as, but is not likely to be greater than, 60%. We estimate that Little Kern golden trout exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 69%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like darters, than on species with greater adult survival, such as sturgeon.

Little Kern golden trout appear to be intermediate. Females reach reproductive maturity in 3-4 years and live up to 9 years. While their fecundity appears relatively low, females contain between 41 and 65 eggs per year, they appear to reproduce each year after reaching maturity. Nevertheless, we expect that reductions in fecundity and survival of young fish through the first year would have a substantial population-level effect on Little Kern golden trout.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Little Kern golden trout's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Little Kern golden trout may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Little Kern golden trout population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Little Kern golden trout are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion likely reduces the reproduction, numbers, and distribution of the Little Kern golden trout.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the Little Kern golden trout include water of sufficient quality for species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience individual and population growth. Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to these levels. This approval could adversely affect the quality of water to the degree that it would impair

individual reproduction and survival of the Little Kern golden trout, and cause trout to experience adverse effects to growth, swimming performance, condition, and development. We estimate the reduction in the number of hatched eggs could be as high as 60% and the reduction in the survival of young fish through the first year as high as 69%. These effects are estimated to be of a magnitude great enough to reduce numbers of Little Kern golden trout. Approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the Little Kern golden trout.

APACHE TROUT
Oncorhynchus apache

Apache trout exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column. Exposure of Apache trout to cyanide at the criterion maximum concentration (CMC) is likely to reduce survival.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Apache trout exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 87%. We estimate that Apache trout exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 90%. In addition, we estimate that juvenile Apache trout exposed to cyanide at the CMC are likely to experience a substantial reduction in survival and that reduction could be as much as, but is not likely to be greater than, >99.9%. Effects of cyanide at the CMC on the survival of other Apache trout life stages is expected to be of lesser magnitude.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth

rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like Apache trout, than on species with greater adult survival, such as sturgeon.

We expect that reductions in fecundity and survival of young fish through the first year would have a greater population-level effect on Apache trout than on longer-lived species, because the majority of Apache trout females spawn only twice in their lifetime. It is estimated that female Apache trout produce between 72 and 1,083 eggs per female, depending on size class. If we reduce the number of hatched eggs by 87% and reduce the survival of fish in their first year by 90% then few fish will survive to the adult stage. If we consider the effects additive, then only 0.2 fish will survive to the adult stage. The combined effect could be less than additive (i.e. between 0.2 and 0.6) if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. The reductions we estimate in hatched eggs and juvenile survivorship could reduce, by an order of magnitude, the number of individuals surviving to adulthood. These reductions in reproduction could be exacerbated further if reductions in juvenile growth rates result in smaller females reproducing in a given year. As we noted in the Status/Baseline section, the number of eggs female Gila trout produce is proportional to their mass.

Apache trout are considered a subspecies of Gila trout and share a very similar life history and the same threats. Brown et al. (2001) performed a population viability analysis for the Gila trout to explore potential management strategies. A base model was constructed to be used as a benchmark for comparison of the effects of different management strategies. Fecundity was estimated from the overall mean count of ova from field-stripped fish. Among their findings, the model was sensitive to large changes in fecundity. Halving fecundity significantly increased the probability of extinction as compared to the base model. We would anticipate a similar response pattern for Apache trout. Exposure to cyanide at the chronic criterion affects not only fecundity, but also hatchability, and juvenile survival of Apache trout. The combined effects could be much greater than what was analyzed by Brown et al. (2001).

Brown et al. (2001) identified catastrophic events as having a much larger influence on the viability of Gila trout than population size, fecundity, or population structure. This conclusion applied to the relative importance of each variable separately and not in combination. Any combination of variables would pose a greater risk to continued viability than any one risk alone. The reductions in reproduction and survival we estimate could occur would diminish the ability of the Apache trout's populations to recover from a catastrophic event, such as a forest fire or drought, and would increase the risk of extirpation of an effected population. Cyanide toxicity could result in the extant population persisting at a reduced population size prior to a catastrophic event. If the local population initially survives the catastrophic event, reductions in reproduction and survival could lengthen the time to recover and leave the population more vulnerable to extirpation.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Apache trout's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Apache trout may also experience effects on growth, swimming performance, condition, and development. Exposure to cyanide at the acute criterion could also substantially reduce the survivorship of juvenile Apache trout. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Apache trout population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Apache trout are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the acute and chronic criteria could reduce the reproduction, numbers, and distribution of the Apache trout.

LAHONTAN CUTTHROAT TROUT

Oncorhynchus clarkii henshawi

Lahontan cutthroat trout exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the *Overview* section. The effects of chronic toxicity could also be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column. Exposure of Lahontan cutthroat trout to cyanide at the criterion maximum concentration (CMC) is likely to reduce survival.

Relatively few studies are available for estimating the magnitude of effects that could occur following exposure of this species to cyanide at criterion concentrations. However, data are available to develop quantitative estimates of the effects of such exposure on Lahontan cutthroat trout fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate that Lahontan cutthroat trout exposed to cyanide at the CCC are likely to experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 80%. We estimate that Lahontan cutthroat trout exposed to cyanide at the CCC are likely to experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 85%. In addition, we estimate that juvenile Lahontan cutthroat trout exposed to cyanide at the CMC are likely to experience a substantial reduction in survival and that reduction could be as much as, but is not likely to be greater than, 43%. Effects of cyanide at the CMC on the survival of other life stages of Lahontan cutthroat trout is expected to be of lesser magnitude.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also reduce productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section above on *Population Responses to Reductions in Fecundity and Juvenile Survival*, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like Lahontan cutthroat trout where the majority of females die after their first spawning, than on species with greater adult survival, such as sturgeon.

In summary, exposure to cyanide concentrations at the chronic criterion are likely to substantially reduce the Lahontan cutthroat trout's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Lahontan cutthroat trout may also experience adverse effects on growth, swimming performance, condition, and development. Exposure to cyanide at the acute criterion could also substantially reduce the survivorship of juvenile Lahontan cutthroat trout. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An affected Lahontan cutthroat trout population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect that is likely to occur at the criterion concentration, we conclude that ultimately Lahontan cutthroat trout are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the acute and chronic criteria at the rangewide scale is likely to reduce the reproduction, numbers, and distribution of the Lahontan cutthroat trout at the rangewide scale.

PAIUTE CUTTHROAT TROUT

Oncorhynchus clarkii seleniris

Paiute cutthroat trout exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Paiute cutthroat trout exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 60%. We estimate that Paiute cutthroat trout

exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 69%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like Paiute cutthroat trout, than on species with greater adult survival, such as sturgeon.

We expect that reductions in fecundity and survival of young fish through the first year would have a greater population-level effect on Paiute cutthroat trout than on longer-lived species, because female Paiute cutthroat trout spawn few times in their lifetime.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Paiute cutthroat trout's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Paiute cutthroat trout may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Paiute cutthroat trout population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Paiute cutthroat trout are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion likely reduces the reproduction, numbers, and distribution of the Paiute cutthroat trout.

GREENBACK CUTTHROAT MOUNTAIN TROUT

Oncorhynchus clarki stomias

Greenback cutthroat trout exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Greenback cutthroat trout exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 60%. We estimate that Greenback cutthroat trout exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 69%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn than on species with greater adult survival, such as sturgeon.

The reductions we estimate in hatched eggs and survival of young fish through the first year could reduce substantially the number of individuals surviving to adulthood. These reductions in reproduction could be exacerbated further if reductions in juvenile growth rates result in smaller females reproducing in a given year. The number of eggs female greenback trout produce is proportional to their mass.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Greenback cutthroat trout's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Greenback cutthroat trout may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Greenback cutthroat trout population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Greenback cutthroat trout are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion likely reduces the reproduction, numbers, and distribution of the Greenback cutthroat trout.

GILA TROUT

Oncorhynchus gilae

Gila trout exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Gila trout exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 60%. We estimate that Gila trout exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 69%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like Gila trout, than on species with greater adult survival, such as sturgeon.

We expect that reductions in fecundity and survival of young fish through the first year would have a greater population-level effect on Gila trout than on longer-lived species, because the majority of Gila trout females spawn only twice in their lifetime. Brown et al. (2001) estimated that female fish produced between 62 and 197 eggs per spawn, depending on size class. In the Gila Trout Recovery Plan (Service 2003) the Service estimated that for every 100 eggs that hatch about half will survive to the juvenile life stage. Of those approximately 50 fish, only about 6 will survive to the subadult stage and of those 6 subadults, only 2 will survive to the adult life stage. This estimate provides a good reference for characterizing the effects of chronic cyanide toxicity. If we simply reduce the number of hatched eggs by 60% and assume survivorship remains the same for other transitions, then only 0.7 fish will survive to the adult stage. If we reduce the survival of fish in their first year by 69% and assume survivorship remains the same for other transitions, then only 0.6 fish will survive to the adult stage. If we consider the

effects additive, then only 0.2 fish will survive to the adult stage. The combined effect could be less than additive (i.e. between 0.2 and 0.6) if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. The reductions we estimate in hatched eggs and juvenile survivorship could reduce, by an order of magnitude, the number of individuals surviving to adulthood. These reductions in reproduction could be exacerbated further if reductions in juvenile growth rates result in smaller females reproducing in a given year. As we noted in the Status/Baseline section, the number of eggs female Gila trout produce is proportional to their mass.

Brown et al. (2001) performed a population viability analysis for the Gila trout to explore potential management strategies. A base model was constructed to be used as a benchmark for comparison of the effects of different management strategies. Fecundity was estimated from the overall mean count of ova from field-stripped fish. Among their findings, the model was sensitive to large changes in fecundity. Halving fecundity significantly increased the probability of extinction as compared to the base model. Exposure to cyanide at the chronic criterion affects not only fecundity, but also hatchability, and juvenile survival. The combined effects could be much greater than what was analyzed by Brown et al. (2001).

Brown et al. (2001) identified catastrophic events as having a much larger influence on the viability of Gila trout than population size, fecundity, or population structure. This conclusion applied to the relative importance of each variable separately and not in combination. Any combination of variables would pose a greater risk to continued viability than any one risk alone. The reductions in reproduction and survival we estimate could occur would diminish the ability of the Gila trout's populations to recover from a catastrophic event, such as a forest fire or drought, and would increase the risk of extirpation of an effected population. Cyanide toxicity could result in the extant population persisting at a reduced population size prior to a catastrophic event. If the local population initially survives the catastrophic event, reductions in reproduction and survival could lengthen the time to recover and leave the population more vulnerable to extirpation. Brown et al. (2001) found the Gila trout's extinction risk was sensitive to the number of populations. For example, increasing the number of Gila trout populations in the model from 10 to 16 significantly reduced risk of extinction by 15%.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Gila trout's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Gila trout may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Gila trout population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Gila trout are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the

chronic criterion likely reduces the reproduction, numbers, and distribution of the Gila trout.

ATLANTIC SALMON

Salmo salar

Gulf of Maine Distinct Population Segment

Atlantic salmon exposed to cyanide at the chronic criterion concentration are likely to experience reduced survival, reproduction, and may also experience effects on growth, swimming performance, condition, and development, as described above in the Effects Overview. The effects of chronic toxicity could be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. However, data were available to develop quantitative estimates of the effects on fecundity, hatching success, and survival of young first-year fish (Table 13). Compared to control populations, we estimate Atlantic salmon exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than, 36%. We estimate that Atlantic salmon exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than, 41%.

As noted previously, the young first-year fish that do survive could experience reduced growth rates which would increase their vulnerability to a host of potential stressors, including temperature, flow, and inter- and intraspecific competition for food and cover. It could also delay reproductive maturity and productivity.

The reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the chronic criterion concentration. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section on Population Level Effects, we noted that reductions in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to spawn, like Atlantic salmon, than on species with greater adult survival, such as sturgeon.

Atlantic salmon typically live about 5 years, spending 1-3 years as freshwater smolts prior to migrating to the ocean as adults where they will develop in about 2-3 years into mature salmon and then return to their natal freshwater rivers to spawn. We expect that reductions in fecundity and survival of young fish through the first year would have a greater population-level effect on Atlantic salmon than on longer-lived species, because the majority of Atlantic salmon females spawn only once in their lifetime. Females deposit 7,000-8,000 eggs per spawn. Studies in Maine indicate less than 10% of the eggs spawned will survive to emerge as feeding fry. The Gulf of Maine DPS has declined to

critically low levels. Adult returns, juvenile abundance estimates and survival have continued to decline since the listing. In 2004, total adult returns to the eight rivers still supporting wild Atlantic salmon populations within the DPS were estimated to range from 60 to 113 individuals. No adults were documented in three of the eight rivers. Declining smolt production has also been documented in recent years, despite fry stocking. For example, from 1996 through 1999, annual smolt production in the Narraguagus River was estimated to average about 3,000 fish. Smolt production declined significantly in 2000 and for the past three years has averaged only about 1,500 fish per year. Overwinter survival in the Narraguagus River since 1997 has only averaged about 12%, approximately half of the survival rate of previous years and significantly less than the 30% previously accepted for the region. These estimates provide a good reference for characterizing the effects of chronic cyanide toxicity.

Based on a minimum of 7000 eggs per spawn, if we simply reduce the number of hatched eggs by 36% assuming overwinter survival is at the previously accepted level of 30%, then only 134 smolts (as opposed to 210) will survive through the first year. If we reduce the smolt survivorship by 41% then only 124 fish will survive through the first year. If we consider the effects additive then only 79 smolts will survive through the first year. The combined effect could be less than additive (i.e. between 79 and 134) if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. If we consider that in some rivers survival is as low as 12%, the effects of cyanide at the CCC could result in as little as 32 smolts per spawn surviving through the first year.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Atlantic salmon's reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and reducing the survivorship of young fish in their first year. Atlantic salmon may also experience effects on growth, swimming performance, condition, and development. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Atlantic salmon population's decline could stabilize at a reduced absolute population number or could continue to decline until it is extirpated. Based upon the magnitude of effect we anticipate could occur, we conclude ultimately Atlantic salmon are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion likely reduces the reproduction, numbers, and distribution of the Atlantic salmon.

7.2 Effects to Invertebrates

Direct effects to invertebrates are anticipated for two species within the genus *Gammarus*, the Illinois cave amphipod (*Gammarus acherondytes*), and Noel's amphipod (*Gammarus desperatus*). No direct effects are anticipated for other invertebrate species (see Appendix B for screening methodology).

ILLINOIS CAVE AMPHIPOD

Gammarus acherondytes

NOEL'S AMPHIPOD
Gammarus desperatus

EC_A values for the Illinois cave amphipod and Noel's amphipod were derived using EPA's Interspecies Correlation Estimate (ICE) model for the genus *Gammarus* with *Daphnia magna* as the surrogate species (EPA 2003b). The lower 95% confidence interval of the LC₅₀ value was divided by the cyanide-specific value of 1.21 to derive the acute EC_A (24.5 ug/L) and by the invertebrate ACR to derive the chronic EC_A (3.92 ug/L). The proposed chronic criterion (5.2 ug/L) exceeds the chronic EC_A for these species. On that basis, exposure of these amphipods to cyanide concentrations at these criteria is likely to cause adverse effects.

Two, acute 96-hour LC₅₀ values were found in the literature for species within the genus *Gammarus*: *G. fasciatus* (LC₅₀ value = 903 ug/L) and *G. pseudolimnaeus* (142.9 ug/L) (Ewell et al 1986, Smith et al.1978). Smith et al. (1978) also derived a lethal threshold concentration at which the first signs of lethality appear of 74 ug/L for *G. pseudolimnaeus*. Although the measured LC₅₀ values are an order of magnitude greater than that derived using the lower 95% confidence interval of the ICE estimate (29.6 ug/L), it is not unusual for species that are closely related taxonomically to have widely variable sensitivities to particular toxicants. For cyanide, numerous LC₅₀ values for species within the family Cyprinidae were measured and the results varied by a factor of 18 between the most and least sensitive species. The measured LC₅₀ values for *Gammarus* represent the only two such estimates known for this group, a large genus comprised of over 200 species.

In the only study in which chronic toxicity was tested in freshwater amphipods (Smith et al.1979, Oseid and Smith 1979), *G. pseudolimnaeus* was found to be 6 to 25 times more sensitive to cyanide than the isopod species *Asellus communis* for acute and chronic endpoints. When *G. pseudolimnaeus* was housed alone, overall mass (free individuals plus eggs and young) in tanks containing cyanide concentrations of 16 ug/L and 32 ug/L and above was significantly lower than controls. Smith et al. (1979) and Oseid and Smith (1979) did not believe that reductions in the 16 ug/L tank were due to cyanide exposure, but provided no alternate explanation for the decline. Effects of cyanide exposure were heightened for *G. pseudolimnaeus* when exposed concurrently with *A. communis* in the same test tank. While the aggressive and competitive *G. pseudolimnaeus* greatly out-competed *A. communis* in tanks containing the control or 4 ug/L treatment, a significant shift took place in tanks with concentrations of 9 ug/L or greater cyanide. When exposed to cyanide in tanks containing *A. communis*, total mass of *G. pseudolimnaeus* was reduced 63% compared to controls at 9 ug/L and 97% at 21 ug/L, the next highest concentration tested. This impact occurred at concentrations lower than when *G. pseudolimnaeus* was exposed alone. Smith et al. (1979) and Oseid and Smith (1979) speculated that decreased survival of *G. pseudolimnaeus* may have been caused by predation by *A. communis*. Thus, cyanide not only exerts direct effects on *G. pseudolimnaeus*, but can shift the competitive advantage to more tolerant species in mixed communities. Smith et al. (1979) and Oseid and Smith (1979) concluded that

where a mixed community exists, *Gammarus* is likely to be excluded in the presence of cyanide pollution. No effects were seen at cyanide concentrations of 4 ug/L and below. The differential sensitivity reported by Smith et al. (1979) and Oseid and Smith (1979) was supported by Ewell et al. (1986), who did not look at chronic effects, but found that acute sensitivity of *G. fasciatus* was nearly twice that of *A. intermedius*.

These results may be applicable to the listed amphipods, as interspecific interactions with other amphipod and isopod species are believed to be important factors in the biology of Gammerids. The Illinois cave amphipod was regularly recorded in caves containing several species of amphipod and isopod species, as well as other invertebrates and fish (Webb et al. 1998). Noel's amphipod is part of a complex of *Gammarus* species that co-occur together in the Pecos River Basin of New Mexico and Texas. Resource partitioning according to substrate and water depth has been examined for these species, but is not fully understood. While fungi and detritus have been suggested as important food sources for amphipods and isopods, it is also recognized that these species can be predators on other species and even exhibit cannibalistic behaviors in high density situations.

Both the chronic EC_A and the chronic data available indicate that cyanide at criteria concentrations may result in the loss of individuals of the Illinois cave amphipod and Noel's amphipod. Effects of cyanide appear to be exasperated when amphipods are exposed in situations of resource competition or possibly predation, when a population may experience a loss of mass that is greater than 0% and less than 63%. The Illinois cave amphipod and Noel's amphipod are known to occur in systems that contain other invertebrate species that may occupy a similar niche and exhibit more tolerance to cyanide, making these two species more susceptible to the effects of cyanide under these conditions. Because both of these species exist in populations that are geographically isolated from one another, the ability of these amphipods to recolonize habitat is limited. For these reasons, exposure of these two species to cyanide concentrations at or near the proposed chronic criterion value is likely to contribute to the elimination of a population unit. The loss of a population unit for either amphipod species would substantially reduce its reproduction, numbers, or distribution at the rangewide scale.

7.3 Effects to Mussels

Listed mussels are not anticipated to experience direct effects of cyanide at criteria concentrations (see Appendix B for screening methodology and discussion of direct effects to mussels), but were screened in based on indirect effects to host fish species. For most mussel species, transformation on a host fish is a required element of their life cycle that cannot be bypassed. Host fish availability and density have been found to be significant factors influencing mussel persistence in particular habitats (Haag and Warren 1998). Contact between glochidia and suitable host fishes is a low-probability event even in healthy populations (Neves et al. 1997), and despite the large number of glochidia produced by an individual mussel, infestation rates tend to be very low (Haag and Warren 1999, Layzer et al. 2003). Host fish generalists, whose glochidia can transform on multiple species, release glochidia in large mucous webs that entangle fish

indiscriminately. Host fish specialists, relying on one or few species to transform, tend to employ specialized lures that mimic prey items of host fish (Strayer et al. 2004). This strategy likely reduces the probability of infestation in unsuitable fish hosts, which can result in immune system incompatibility, and discharge of glochidia. Glochidia that fail to infest a suitable host or that have been sloughed off by an unsuitable host will not transform to the adult stage and will survive only as long as their energy reserves last, from a few days to up to two weeks. The odds of an individual glochidium infesting a host and completing transformation have been estimated at 4 in 100,000. Given those odds, any reduction of host fish species populations caused by exposure to cyanide at either the acute or chronic criterion levels is likely to cause an adverse effect to the listed mussel species.

In addition, mussel glochidia infested on fish are completely parasitic and are dependent on the host for oxygen, nutrition, and overall survival. Once contact is made with a suitable host, successful glochidia encyst on the gills, fins, or skin for a period of several weeks. Any mortality to host fish during this period will necessarily result in mortality to infested glochidia. Thus, the host fish become surrogates for the listed mussels during the parasitic stage.

Estimating Acute and Chronic EC_A Values for Host Fish Species

In estimating risks to glochidia host species, the Service derived acute and chronic assessment effects concentrations (EC_A) estimates according to the prioritization described in Figures 2 and 4 of the 2004 Draft Methodology for Conducting Biological Evaluations of Aquatic Life Criteria (Table 15). Although host fish species identified for listed mussels were themselves not listed species, the host fish species is an obligate part of the mussel lifecycle to which it has a parasitic dependence while attached, and is treated with the same conservatism as a listed species.

1. When acute toxicity data were available for a host species, the species mean acute value from Table 1 of the BE was used to derive the acute EC_A, and, when appropriate, the chronic EC_A. However, when only one LC₅₀ was available for a species, the lower 95% confidence interval of the LC₅₀, when available, was used to derive the EC_A.
2. For yellow perch (*Perca flavescens*), the LC₅₀ reported in Table 1 of the BE could not be reproduced from the original source, so an acute EC_A value was calculated from the mean of all 96-hour LC₅₀ values (converted to free CN) reported in Smith et al. (1978).
3. For fish with no species-specific data, ICE models were derived at the lowest taxonomic level with adequate data (EPA 2003b). The lower 95% confidence interval of the predicted LC₅₀ was used to determine EC_A values.
4. For species for which no ICE models were available, the 5th percentile LC₅₀ from the appropriate SSD was used in lieu of the mean LC₅₀.

All acute EC_A values were calculated by division of the LC₅₀ by the cyanide-specific 1.21 factor derived by the Service.

Table 15. Surrogate taxa used to estimate LC₅₀ values for evaluation of to host fish effects

Surrogate taxa used to estimate host fish LC ₅₀	LC ₅₀ (ug CN/L)	Acute EC _A (ug/L)	Chronic EC _A (ug/L)
Actinopterygii (class)	66.5 ¹	54.93	2.86
Cypriniformes (order)	84.5 ¹	69.88	3.64
Cyprinidae (family)	101.7 ²	84.07	4.38
<i>Pimephales promelas</i> (species)	138.4 ³	114.38	5.96
Cyprinodontiformes			
<i>Poecilia reticulata</i> (species)	187.8 ³	155.21	8.09
Perciformes (order)	90.8 ¹	75.04	3.91
Centrarchidae (family)	73.2 ²	60.46	3.15
<i>Lepomis</i> (genus)	89.5 ²	73.99	3.85
<i>Lepomis cyanellus</i> (species)	126.2 ²	104.03	5.43
<i>Lepomis macrochirus</i> (species)	126.1 ³	104.21	5.43
<i>Micropterus</i> (genus)			
<i>Micropterus salmoides</i> (species)	95.7 ⁴	79.09	4.12
<i>Pomoxis</i> (genus)			
<i>Pomoxis nigromaculatus</i> (species)	84.5 ⁴	70.66	3.64
Percidae (family)	42.3 ²	34.97	1.82
<i>Etheostoma</i> (genus)	40.0 ²	33.07	1.72
<i>Perca</i> (genus)			
<i>Perca flavescens</i> (species)	93.3 ⁵	77.11	4.02
Salmoniformes			
<i>Salmo salar</i> (species)	90 ³	74.38	3.88
<i>Salmo trutta</i> (species)	54.9 ²	45.38	2.36
Siluriformes (order)			
Ictaluridae (family)	182.8 ²	151.06	7.87
<i>Ictalurus punctatus</i> (species)	190.3 ²	83.83	8.20

¹ LC₅₀ based on 5th percentile estimate from species sensitivity distribution (SSD), Table 2 – Cyanide BE.

² LC₅₀ estimate based on lower bound of the 95% CI from ICE model (Appendix D)

³ LC₅₀ based on measured value from the Cyanide BE (Table 1);

⁴ LC₅₀ based on lower bound of the 95% CI of the measured value from the Cyanide BE (Table 1);

⁵ LC₅₀ based on mean measured value from Smith et al 1978

Effects to Listed Mussel Species

Table 16 identifies host fish species for all currently listed mussel species, with estimates of their acute and chronic EC_A values, and predicted maximum effects to fecundity and juvenile survival. We used listed species accounts, NatureServe, recent FWS Recovery Plans and 5-Year Reviews, the open literature, and consultation with species experts to update and expand the list of known hosts for these species.

Two of the listed mussels, the fat pocketbook (*Potamilus capax*) and the scaleshell mussel (*Leptodea leptodon*), have an identified obligate relationship with a single host, the freshwater drum (*Aplodinotus grunniens*). The remaining listed mussels either lack a known obligate relationship with a host fish or host fish relationships are unknown. Of the 300 identified North American mussel species, there have been no host fish species identified for a significant percentage, and it is likely that the complement of host fish remains incomplete for many other species. Individual mussel species have been found to have up to 25 fish species known to serve as suitable hosts, though host fish specificity to one species or a group of species related by taxonomy or food guild can be common in mussels. To assess effects to these species, we looked at the range of all fish species that have been identified as hosts for listed mussels (Table 17). We then totaled the number of species identified as hosts in each family, as well as the frequency that each family was represented as a host fish for a listed mussel species, as identified in Table 15. Fish within families that may be sensitive to cyanide at concentrations below acute or chronic criteria, accounted for 96% of the species diversity (Table 18), and 98% of all fish identified as hosts for listed mussels (Table 19). Extrapolating these results to fish with no known hosts or a potentially incomplete list of hosts, we can assume that any mussel is likely to have at least one host fish for which adverse effects cannot be ruled out at exposure to cyanide at criteria concentrations. Therefore, for mussels lacking a known obligate relationship with a host fish, in the absence of information to the contrary, it was assumed that the species was likely to have at least one host fish that is sensitive to cyanide at the criteria concentrations, whether one is currently identified or not. On that basis, these mussel species are likely to be adversely affected at cyanide criteria concentrations due to potential reductions in host fish abundance.

Table 16. Cyanide sensitivity of fish species that serve as hosts for glochidia of listed mussels.

Listed Mussel	Host Fish	Acute EC _A (ug/L)	Chronic EC _A (ug/L)	Source/surrogate taxa for LC ₅₀ values (Table 15)	Estimated reduction in fecundity and larvae/juvenile survival of host fish due based on surrogate species data sets (Appndix F)		
					Fathead Minnow (Reduction in the mean number of eggs spawned)	Brook Trout (Reduction in the mean number of eggs spawned)	Bluegill (reduction in larvae/juvenile survival)
MUSSELS WITH OBLIGATE HOST FISH							
Fat Pocketbook <i>Potamilus capax</i>	Freshwater drum	75.04	3.91	Perciformes (order)	36% (24%, 47%)	24% (0%, 53%)	40% (0%, 79%)
Scaleshell Mussel <i>Leptodea leptodon</i>	Freshwater drum	75.04	3.91	Perciformes (order)	36% (24%, 47%)	24% (0%, 53%)	40% (0%, 79%)
MUSSELS WITH NO KNOWN OBLIGATE HOST FISH							
Cumberland Elktoe <i>Alasmidonta atropurpurea</i>	Rainbow darter Redline darter Fantail darter	33.07	1.72	<i>Etheostoma</i> (genus)	65% (60%, 70%)	40% (20%, 58%)	74% (46%, 88%)
	Banded sculpin	54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
	Rock bass	60.46	3.15	<i>Centrarchidae</i> (family)	44% (35%, 53%)	28% (0%, 54%)	52% (0%, 81%)
	Northern hogsucker	69.88	3.64	Cypriniformes (order)	39% (28%, 49%)	26% (0%, 54%)	44% (0%, 80%)
	Longear sunfish	73.99	3.85	<i>Lepomis</i> (genus)	36% (24%, 47%)	24% (0%, 54%)	41% (0%, 79%)
	Whitetail shiner	84.07	4.38	Cyprinidae (family)	31% (18%, 44%)	22% (0%, 53%)	33% (0%, 78%)
Dwarf Wedgemussel <i>Alasmidonta heterodon</i>	Tesselated darter Johnny darter	33.07	1.72	<i>Etheostoma</i> (genus)	65% (60%, 70%)	40% (20%, 58%)	74% (46%, 88%)
	Roanoke darter	34.97	1.82	Percidae (family)	63% (58%, 68%)	39% (18%, 57%)	72% (43%, 87%)
	Mottled sculpin Slimy sculpin	54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
	Atlantic salmon	74.38	3.88	<i>Salmo salar</i> (species)	36% (24%, 47%)	24% (0%, 54%)	41% (0%, 79%)
Appalachian Elktoe <i>Alasmidonta raveneliana</i>	Banded sculpin Mottled sculpin	54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
	Blackbanded darter	33.07	1.72	<i>Etheostoma</i> (genus)	65% (60%, 70%)	40% (20%, 58%)	74% (46%, 88%)
Fat Threeridge <i>Amblema neislerii</i>	Redear sunfish	73.99	3.85	<i>Lepomis</i> (genus)	36% (24%, 47%)	24% (0%, 54%)	41% (0%, 79%)

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	Largemouth bass	79.09	4.12	<i>Micropterus salmoides</i> (species)	34% (21%, 45%)	23% (0%, 53%)	38% (0%, 79%)	
	Weed shiner	84.04	4.38	Cyprinidae (family)	31% (18%, 44%)	22% (0%, 53%)	33% (0%, 78%)	
	Bluegill	104.21	5.43	<i>Lepomis macrochirus</i> (species)	---	---	---	
Ouachita Rock Pocketbook <i>Arkansia wheeleri</i>	White crappie	54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)	
	Warmouth Smallmouth bass	60.46	3.15	Centrarchidae (family)	44% (35%, 53%)	28% (0%, 54%)	52% (0%, 81%)	
	Black crappie	70.66	3.64	<i>Pomoxis nigromaculatus</i> (species)	39% (24%, 49%)	26% (0%, 54%)	44% (0%, 80%)	
	River carpsucker	69.88	3.64	Cypriniformes (order)	39% (28%, 49%)	26% (0%, 54%)	44% (0%, 80%)	
	Longear sunfish Orangespotted sunfish	73.99	3.85	<i>Lepomis</i> (genus)	36% (24%, 47%)	24% (0%, 54%)	41% (0%, 79%)	
	Freshwater drum	75.04	3.91	Perciformes (order)	36% (24%, 47%)	24% (0%, 53%)	40% (0%, 79%)	
	Largemouth bass	79.67	4.12	<i>Micropterus salmoides</i> (species)	34% (21%, 45%)	23% (0%, 53%)	38% (0%, 79%)	
	Dusky shiner Bleeding shiner Golden shiner Emerald shiner	84.07	4.38	Cyprinidae (family)	31% (18%, 44%)	22% (0%, 53%)	33% (0%, 78%)	
	Green sunfish	104.03	5.43	<i>Lepomis cyanellus</i> (species)	---	---	---	
	Bluegill	104.21	5.43	<i>Lepomis macrochirus</i> (species)	---	---	---	
	Birdwing Pearlymussel <i>Conradilla caelata</i>	Greenside darter Tennessee snubnose darter Banded darter	33.07	1.72	<i>Etheostoma</i> (genus)	65% (60%, 70%)	40% (20%, 58%)	74% (46%, 88%)
	Fanshell <i>Cyprogenia stegaria</i>	Banded darter Greenside darter Tennessee snubnose darter	33.07	1.72	<i>Etheostoma</i> (genus)	65% (60%, 70%)	40% (20%, 58%)	74% (46%, 88%)
Blotchside logperch Logperch Tangerine darter		34.97	1.82	Percidae (family)	63% (58%, 68%)	39% (18%, 57%)	72% (43%, 87%)	
Mottled sculpin Banded sculpin		54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)	
Fantail darter Banded darter Tangerine darter Greenside darter Tennessee snubnose darter		33.07	1.72	<i>Etheostoma</i> (genus)	65% (60%, 70%)	40% (20%, 58%)	74% (46%, 88%)	

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	Gilt darter Channel darter Logperch Blotchside logperch	34.97	1.82	Percidae (family)	63% (58%, 68%)	39% (18%, 57%)	72% (43%, 87%)
	Black sculpin	54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
Chipola Slabshell <i>Elliptio chipolaensis</i>	Bluegill	104.21	5.43	<i>Lepomis macrochirus</i> (species)	---	---	---
Tar River Spiny mussel <i>Elliptio steinstansana</i>	Bluehead chub Satinfin shiner White shiner Pinewoods shiner	84.07	4.38	Cyprinidae (family)	31% (18%, 44%)	22% (0%, 53%)	33% (0%, 78%)
Purple Bankclimber <i>Elliptioideus sloatianus</i>	Blackbanded darter	33.07	1.72	<i>Etheostoma</i> (genus)	65% (60%, 70%)	40% (20%, 58%)	74% (46%, 88%)
	Eastern mosquitofish	54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
	Greater Jumprock	69.88	3.64	Cypriniformes (order)	39% (28%, 49%)	26% (0%, 54%)	44% (0%, 80%)
	Guppy	155.21	8.09	<i>Poecilia reticulata</i> (species)	---	---	---
Cumberlandian Combshell <i>Epioblasma brevidens</i>	Wounded darter Redline darter Bluebreast darter Snubnose darter Greenside darter	33.07	1.72	<i>Etheostoma</i> (genus)	65% (60%, 70%)	40% (20%, 58%)	74% (46%, 88%)
	Logperch	34.97	1.82	Percidae (family)	63% (58%, 68%)	39% (18%, 57%)	72% (43%, 87%)
	Banded sculpin Mottled sculpin Black sculpin	54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
Oyster Mussel <i>Epioblasma capsaeformis</i>	Wounded darter Redline darter Bluebreast darter Greenside darter Fantail darter	33.07	1.72	<i>Etheostoma</i> (genus)	65% (60%, 70%)	40% (20%, 58%)	74% (46%, 88%)
	Dusky darter	34.97	1.82	Percidae (family)	63% (58%, 68%)	39% (18%, 57%)	72% (43%, 87%)
	Banded sculpin Black sculpin Mottled sculpin	54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
Curtis Pearly Mussel	Rainbow darter	33.07	1.72	<i>Etheostoma</i> (genus)	65% (60%, 70%)	40% (20%, 58%)	74% (46%, 88%)

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<i>Epioblasma florentina curtisii</i>	Banded sculpin Mottled sculpin	54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
Yellow Blossom (Pearlymussel) <i>Epioblasma florentina florentina</i>	Not known						
Tan Riffleshell <i>Epioblasma florentina walkeri</i>	Fantail darter Greenside darter Redline darter Snubnose darter	33.07	1.72	<i>Etheostoma</i> (genus)	65% (60%, 70%)	40% (20%, 58%)	74% (46%, 88%)
	Banded sculpin Mottled sculpin	54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
Upland Combshell <i>Epioblasma metastrata</i>	Not known						
Catspaw (Purple Cat's Paw Pearlymussel) <i>Epioblasma obliquata obliquata</i>	Blackside darter Logperch	34.97	1.82	Percidae (family)	63% (58%, 68%)	39% (18%, 57%)	72% (43%, 87%)
	Mottled sculpin	54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
	Rock bass	60.46	3.15	Centrarchidae (family)	44% (35%, 53%)	28% (0%, 54%)	52% (0%, 81%)
	Stonecat	151.06	7.87	Ictaluridae (family)	---	---	---
White Catspaw Pearlymussel <i>Epioblasma obliquata perobliqua</i>	Not known						
Southern Acornshell <i>Epioblasma othcaloogensis</i>	Not known						
Southern Combshell <i>Epioblasma penita</i>	Not known						
Tubercled Blossom <i>Epioblasma torulosa torulosa</i>	Not known						
Turgid Blossom <i>Epioblasma turgidula</i>	Not known						
Green Blossom <i>Epioblasma torulosa gubernaculum</i>	Not known						
Northern Riffleshell <i>Epioblasma torulosa rangiana</i>	Banded darter Bluebreast darter Johnny darter Iowa darter	33.07	1.72	<i>Etheostoma</i> (genus)	65% (60%, 70%)	40% (20%, 58%)	74% (46%, 88%)
	Brown trout	45.38	2.36	<i>Salmo trutta</i> (species)	55% (24%, 61%)	34% (9%, 56%)	63% (23%, 84%)

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	Banded sculpin Mottled sculpin	54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
Shiny Pigtoe <i>Fusconaia cor</i>	Whitetail shiner Common shiner Warpaint shiner Telescope shiner	84.07	4.38	Cyprinidae (family)	31% (18%, 44%)	22% (0%, 53%)	33% (0%, 78%)
Fine-rayed Pigtoe <i>Fusconaia cuneolus</i>	Mottled sculpin	54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
	River chub Central stoneroller Telescope shiner Tennessee shiner Whitetail shiner	84.07	4.38	Cyprinidae (family)	31% (18%, 44%)	22% (0%, 53%)	33% (0%, 78%)
	Fathead minnow	114.38	5.96	<i>Pimephales promelas</i> (species)	---	---	---
	Cracking Pearlymussel <i>Hemistena lata</i>	Not known					
Pink Mucket <i>Lampsilis abrupta</i>	Walleye Sauger	34.97	1.82	Percidae (family)	63% (58%, 68%)	39% (18%, 57%)	72% (43%, 87%)
	Spotted bass Smallmouth bass	60.46	3.15	Centrarchidae (family)	44% (35%, 53%)	28% (0%, 54%)	52% (0%, 81%)
	Freshwater drum	75.04	3.91	Perciformes (order)	36% (24%, 47%)	24% (0%, 53%)	40% (0%, 79%)
	Largemouth bass	79.67	4.12	<i>Micropterus salmoides</i> (species)	34% (21%, 45%)	23% (0%, 53%)	38% (0%, 79%)
Fine-lined Pocketbook <i>Lampsilis altilis</i>	Redeye bass Spotted bass	60.46	3.15	Centrarchidae (family)	44% (35%, 53%)	28% (0%, 54%)	52% (0%, 81%)
	Largemouth bass	79.09	4.12	<i>Micropterus salmoides</i> (species)	34% (21%, 45%)	23% (0%, 53%)	38% (0%, 79%)
	Green sunfish	104.03	5.43	<i>Lepomis cyanellus</i> (species)	---	---	---
Higgins Eye <i>Lampsilis higginsii</i>	Sauger Walleye	34.97	1.82	Percidae (family)	63% (58%, 68%)	39% (18%, 57%)	72% (43%, 87%)
	Smallmouth bass	60.46	3.15	Centrarchidae (family)	44% (35%, 53%)	28% (0%, 54%)	52% (0%, 81%)
	Black crappie	70.66	3.64	<i>Pomoxis nigromaculatus</i> (species)	39% (24%, 49%)	26% (0%, 54%)	44% (0%, 80%)
	Freshwater drum	75.04	3.91	Perciformes (order)	36% (24%, 47%)	24% (0%, 53%)	40% (0%, 79%)
	Largemouth bass	79.09	4.12	<i>Micropterus salmoides</i>	34% (21%, 45%)	23% (0%, 53%)	38% (0%, 79%)
	Yellow perch		4.02	<i>Perca flavescens</i> (species)	35% (24%, 46%)	23% (0%, 53%)	38% (0%, 79%)

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Orangenacre Mucket <i>Lampsilis perovalis</i>	Chain pickerel	54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
	Redeye bass	60.46	3.15	Centrarchidae (family)	44% (35%, 53%)	28% (0%, 54%)	52% (0%, 81%)
	Spotted bass						
	Largemouth bass	79.67	4.12	<i>Micropterus salmoides</i> (species)	34% (21%, 45%)	23% (0%, 53%)	38% (0%, 79%)
Arkansas Fatmucket <i>Lampsilis powelli</i>	Spotted bass	60.46	3.15	Centrarchidae (family)	44% (35%, 53%)	28% (0%, 54%)	52% (0%, 81%)
	Largemouth bass	79.09	4.12	<i>Micropterus salmoides</i> (species)	34% (21%, 45%)	23% (0%, 53%)	38% (0%, 79%)
Speckled Pocketbook <i>Lampsilis streckeri</i>	Warmouth	60.46	3.15	Centrarchidae (family)	44% (35%, 53%)	28% (0%, 54%)	52% (0%, 81%)
	Spotted bass						
	Smallmouth bass	73.99	3.85	<i>Lepomis</i> (genus)	36% (24%, 47%)	24% (0%, 54%)	41% (0%, 79%)
	Shadow bass						
Longear sunfish	104.03	5.43	<i>Lepomis cyanellus</i> (species)	---	---	---	
Green sunfish	104.21	5.43	<i>Lepomis macrochirus</i> (species)	---	---	---	
Shinyrayed pocketbook <i>Lampsilis subangulata</i>	Eastern mosquitofish	54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
	Spotted bass	60.46	3.15	Centrarchidae (family)	44% (35%, 53%)	28% (0%, 54%)	52% (0%, 81%)
	Largemouth bass	79.09	4.12	<i>Micropterus salmoides</i> (species)	34% (21%, 45%)	23% (0%, 53%)	38% (0%, 79%)
	Bluegill	104.21	5.43	<i>Lepomis macrochirus</i> (species)	---	---	---
	Guppy	155.21	8.09	<i>Poecilia reticulata</i> (species)	---	---	---
Alabama Lampmussel <i>Lampsilis virescens</i>	Not known						
Carolina Heelsplitter <i>Lasmigon decorata</i>	Not known						
Louisiana Pearlshell <i>Margaritifera hembeli</i>	Brown madtom	151.06	7.87	Ictaluridae (family)	---	---	---
	Blackspotted topminnow	54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
	Striped shiner	84.07	4.38	Cyprinidae (family)	31% (18%, 44%)	22% (0%, 53%)	33% (0%, 78%)
	Redfin shiner						
Golden shiner							
Alabama Moccasinshell <i>Medionidus acutissimus</i>	Tuskaloosa darter Redfin darter Blackbanded darter Southern sand darter Johnny darter Speckled darter	33.07	1.72	<i>Etheostoma</i> (genus)	65% (60%, 70%)	40% (20%, 58%)	74% (46%, 88%)

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	Saddleback darter Naked sand darter Logperch	34.97	1.82	Percidae (family)	63% (58%, 68%)	39% (18%, 57%)	72% (43%, 87%)
	Blackspotted topminnow	54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
Coosa Moccasinshell <i>Medionidus parvulus</i>	Blackbanded darter	34.97	1.82	Percidae (family)	63% (58%, 68%)	39% (18%, 57%)	72% (43%, 87%)
Gulf Moccasinshell <i>Medionidus penicillatus</i>	Blackbanded darter Brown darter	33.07	1.72	<i>Etheostoma</i> (genus)	65% (60%, 70%)	40% (20%, 58%)	74% (46%, 88%)
	Eastern mosquitofish	54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
	Guppy	155.21	8.09	<i>Poecilia reticulata</i> (species)	---	---	---
Ochlockonee Moccasinshell <i>Medionidus simpsonianus</i>	Not known						
Ring Pink <i>Obovaria retusa</i>	Not known						
Littlewing Pearlymussel <i>Pegis fibula</i>	Redline darter Greenside darter Emerald darter	33.07	1.72	<i>Etheostoma</i> (genus)	65% (60%, 70%)	40% (20%, 58%)	74% (46%, 88%)
	Banded sculpin	54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
White Wartyback <i>Plethobasus cicatricosus</i>	Not known						
Orangefoot Pimpleback <i>Plethobasus cooperianus</i>	Not known						
Clubshell <i>Pleurobema clava</i>	Blackside darter Logperch	34.97	1.82	Percidae (family)	63% (58%, 68%)	39% (18%, 57%)	72% (43%, 87%)
	Striped shiner Central stoneroller	84.07	4.38	<i>Cyprinidae</i> (family)	31% (18%, 44%)	22% (0%, 53%)	33% (0%, 78%)
James Spinymussel <i>Pleurobema collina</i>	Fantail darter	33.07	1.72	<i>Etheostoma</i> (genus)	65% (60%, 70%)	40% (20%, 58%)	74% (46%, 88%)
	Pumpkinseed	73.99	3.85	<i>Lepomis</i> (genus)	36% (24%, 47%)	24% (0%, 54%)	41% (0%, 79%)

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	Bluehead chub Rosyside dace Satinfin shiner Rosefin shiner Blacknose dace Central stoneroller Mountain redbelly dace Swallowtail shiner Common shiner	84.07	4.38	Cyprinidae (family)	31% (18%, 44%)	22% (0%, 53%)	33% (0%, 78%)
Black Clubshell <i>Pleurobema curtum</i>	Not known						
Southern Clubshell <i>Pleurobema decisum</i>	Blacktail shiner Alabama shiner Tricolor shiner	84.07	4.38	Cyprinidae (family)	31% (18%, 44%)	22% (0%, 53%)	33% (0%, 78%)
Dark Pigtoe <i>Pleurobema furvum</i>	Blackspotted topminnow	54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
	Largescale stoneroller Alabama shiner Blacktail shiner Creek chub	84.07	4.38	Cyprinidae (family)	31% (18%, 44%)	22% (0%, 53%)	33% (0%, 78%)
Southern Pigtoe <i>Pleurobema georgianum</i>	Alabama shiner Blacktail shiner Tricolor shiner	84.07	4.38	Cyprinidae (family)	31% (18%, 44%)	22% (0%, 53%)	33% (0%, 78%)
Cumberland Pigtoe <i>Pleurobema gibberum</i>	Telescope shiner Striped shiner	84.07	4.38	Cyprinidae (family)	31% (18%, 44%)	22% (0%, 53%)	33% (0%, 78%)
Flat Pigtoe <i>Pleurobema marshalli</i>	Not known						
Ovate Clubshell <i>Pleurobema perovatum</i>	Not known						
Rough Pigtoe <i>Pleurobema plenum</i>	Not known						
Oval pigtoe <i>Pleurobema pyriforme</i>	Eastern mosquitofish	54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
	Sailfin shiner	84.07	4.38	Cyprinidae (family)	31% (18%, 44%)	22% (0%, 53%)	33% (0%, 78%)
	Guppy	155.21	8.09	<i>Poecilia reticulata</i> (species)	---	---	---
Heavy Pigtoe <i>Pleurobema taitianum</i>	Not known						
Alabama Heelsplitter <i>Potamilus inflatus</i>	Freshwater drum	75.04	3.91	Perciformes (order)	36% (24%, 47%)	24% (0%, 53%)	40% (0%, 79%)

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Triangular Kidneyshell <i>Ptychobranchus greeni</i>	Warrior darter Tuskaloosa darter Blackbanded darter	33.07	1.72	<i>Etheostoma</i> (genus)	65% (60%, 70%)	40% (20%, 58%)	74% (46%, 88%)
	Logperch	34.97	1.82	Percidae (family)	63% (58%, 68%)	39% (18%, 57%)	72% (43%, 87%)
Rough rabbitsfoot <i>Quadrula cylindrical strigillata</i>	Whitetail shiner Spotfin shiner Bigeye chub	84.07	4.38	Cyprinidae (family)	31% (18%, 44%)	22% (0%, 53%)	33% (0%, 78%)
Winged Mapleleaf <i>Quadrula fragosa</i>	Blue catfish	151.06	7.87	Ictaluridae (family)	---	---	---
	Channel catfish	83.83	8.20	<i>Ictalurus punctatus</i> (species)	---	---	---
Cumberland Monkeyface <i>Quadrula intermedia</i>	Streamline chub Blotched chub	84.07	4.38	Cyprinidae (family)	31% (18%, 44%)	22% (0%, 53%)	33% (0%, 78%)
Appalachian Monkeyface <i>Quadrula sparsa</i>	Not known						
Stirrupshell <i>Quadrula stapes</i>	Not known						
Pale Lilliput <i>Toxolasma cylindrellus</i>	Not known						
Purple Bean <i>Villosa perpurpurea</i>	Fantail darter Greenside darter	33.07	1.72	<i>Etheostoma</i> (genus)	65% (60%, 70%)	40% (20%, 58%)	74% (46%, 88%)
	Black sculpin Mottled sculpin Banded sculpin	54.93	2.86	Actinopterygii (class)	48% (39%, 56%)	30% (1%, 55%)	56% (3%, 82%)
	Fantail darter Striped darter	33.07	1.72	<i>Etheostoma</i> (genus)	65% (60%, 70%)	40% (20%, 58%)	74% (46%, 88%)

Table 17. Fish species identified as hosts of listed mussels.

Family	Common Name	Genus species
Catostomidae		
	Northern hogsucker	<i>Hypentelium nigricans</i>
	Greater jumprock	<i>Moxostoma lachneri</i>
Centrarchidae		
	Shadow bass	<i>Ambloplites ariommus</i>
	Rock bass	<i>Ambloplites rupestris</i>
	Warmouth	<i>Chaenobryttus gulosus</i>
	Green sunfish	<i>Lepomis cyanellus</i>
	Bluegill	<i>Lepomis macrochirus</i>
	Longear sunfish	<i>Lepomis megalotis</i>
	Redear sunfish	<i>Lepomis microlophus</i>
	Redeye bass	<i>Micropterus coosae</i>
	Smallmouth bass	<i>Micropterus dolomieu</i>
	Spotted bass	<i>Micropterus punctulatus</i>
	Largemouth bass	<i>Micropterus salmoides</i>
	Black crappie	<i>Pomoxis nigromaculatus</i>
Cottidae		
	Black sculpin	<i>Cottus baileyi</i>
	Mottled sculpin	<i>Cottus bairdii</i>
	Banded sculpin	<i>Cottus carolinae</i>
	Slimy sculpin	<i>Cottus cognatus</i>
Cyprinidae		
	Central stoneroller	<i>Campostoma anomalum</i>
	Largescale stoneroller	<i>Campostoma oligolepis</i>
	Rosyside dace	<i>Clinostomus funduloides</i>
	Satinfin shiner	<i>Cyprinella analostana</i>
	Alabama shiner	<i>Cyprinella callistia</i>
	Whitetail shiner	<i>Cyprinella galactura</i>
	Spotfin shiner	<i>Cyprinella spiloptera</i>
	Tricolor shiner	<i>Cyprinella trichroistia</i>
	Blacktail shiner	<i>Cyprinella venusta</i>
	Streamline chub	<i>Erimystax dissimilis</i>
	Blotched chub	<i>Erimystax insignis</i>
	Bigeye chub	<i>Hybopsis amblops</i>
	Striped shiner	<i>Luxilus chrysocephalus</i>
	Common shiner	<i>Luxilus cornutus</i>
	Rosefin shiner	<i>Lythrurus ardens</i>
	Pinewoods shiner	<i>Lythrurus matutinus</i>
	Redfin shiner	<i>Lythrurus umbratilis</i>
	Bluehead chub	<i>Nocomis leptcephalus</i>
	River chub	<i>Nocomis micropogon</i>

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	Golden shiner	<i>Notemigonus crysoleucas</i>
	White shiner	<i>Notropis albeolus</i>
	Warpaint shiner	<i>Notropis coccogenis</i>
	Tennessee shiner	<i>Notropis leuciodus</i>
	Swallowtail shiner	<i>Notropis procne</i>
	Telescope shiner	<i>Notropis telescopus</i>
	Weed shiner	<i>Notropis texanus</i>
	Mountain redbelly	<i>Phoxinus oreas</i>
	Sailfin shiner	<i>Pteronotropis hypselopterus</i>
	Blacknose dace	<i>Rhinichthys atratulus</i>
	Creek chub	<i>Semotilus atromaculatus</i>
Esocidae		
	Chain pickerel	<i>Esox niger</i>
Fundulidae		
	Blackspotted topminnow	<i>Fundulus olivaceus</i>
Ictaluridae		
	Blue catfish	<i>Ictalurus furcatus</i>
	Channel catfish	<i>Ictalurus punctatus</i>
	Stonecat	<i>Noturus flavus</i>
	Brown madtom	<i>Noturus phaeus</i>
Percidae		
	Naked sand darter	<i>Ammocrypta beanii</i>
	Emerald darter	<i>Etheostoma baileyi</i>
	Warrior darter	<i>Etheostoma bellator</i>
	Greenside darter	<i>Etheostoma blennioides</i>
	Rainbow darter	<i>Etheostoma caeruleum</i>
	Bluebreast darter	<i>Etheostoma camurum</i>
	Tuskaloosa darter	<i>Etheostoma douglasi</i>
	Brown darter	<i>Etheostoma edwini</i>
	Fantail darter	<i>Etheostoma flabellare</i>
	Southern sand darter	<i>Etheostoma meridianum</i>
	Johnny darter	<i>Etheostoma nigrum</i>
	Tesselated darter	<i>Etheostoma olmstedii</i>
	Redline darter	<i>Etheostoma rufilineatum</i>
	Tennessee snubnose darter	<i>Etheostoma simoterum</i>
	Snubnose darter	<i>Etheostoma simoterum</i>
	Speckled darter	<i>Etheostoma stigmaeum</i>
	Striped darter	<i>Etheostoma virgatum</i>
	Wounded darter	<i>Etheostoma vulneratum</i>
	Redfin darter	<i>Etheostoma whipplei</i>
	Banded darter	<i>Etheostoma zonale</i>
	Yellow perch	<i>Perca flavescens</i>
	Tangerine darter	<i>Percina aurantiaca</i>
	Blotchside logperch	<i>Percina burtoni</i>
	Logperch	<i>Percina caprodes</i>

	Channel darter	<i>Percina copelandi</i>
	Gilt dater	<i>Percina evides</i>
	Blackside darter	<i>Percina maculata</i>
	Blackbanded darter	<i>Percina nigrofasciata</i>
	Roanoke darter	<i>Percina roanoka</i>
	Dusky darter	<i>Percina sciera</i>
	Saddleback darter	<i>Percina vigil</i>
	Fathead minnow	<i>Pimephales promelas</i>
	Sauger	<i>Sander canadensis</i>
	Walleye	<i>Sander vitreus</i>
Poecilidae		
	Eastern mosquitofish	<i>Gambusia holbrooki</i>
	Guppy	<i>Poecilia reticulata</i>
Salmonidae		
	Atlantic salmon	<i>Salmo salar</i>
	Brown trout	<i>Salmo trutta</i>
Sciaenidae		
	Freshwater drum	<i>Aplodinotus grunniens</i>

Table 18. Species diversity of host fish for glochidia of listed mussels.

Host fish by species		
Family	# species	% all species
Percidae* (darters, perch)	35	34
Cyprinidae* (shiners, chub, dace, stonerollers)	34	33
Centrarchidae* (bass, bluegill, sunfish)	15	15
Cottidae* (sculpin)	4	4
Ictaluridae (catfish)	4	4
Catostomidae* (suckers)	3	3
Poecilidae* (mosquitofish, guppies)	2	2
Salmonidae* (salmon, trout)	2	2
Sciaenidae* (drum)	1	1
Esocidae* (pickerel)	1	1
Fundulidae* (topminnows)	1	1
TOTAL	102	100

*Sensitive to cyanide concentrations below EPA's proposed Aquatic Life Criteria.

Table 19. Frequency of occurrence of host fish for glochidia of listed mussels.

Host fish by frequency of occurrence		
Family	# occurrences	% all occurrences
Percidae* (darters, perch)	82	36%
Cyprinidae* (shiners, chub, dace, stonerollers)	51	22%
Centrarchidae* (bass, bluegill, sunfish)	42	18%
Cottidae* (sculpin)	26	11%
Poeciliidae* (mosquitofish, guppies)	8	4%
Sciaenidae* (drum)	6	3%
Ictaluridae (catfish)	4	2%
Catostomidae* (suckers)	3	1%
Fundulidae* (topminnows)	3	1%
Salmonidae* (salmon, trout)	2	1%
Esocidae* (pickerel)	1	<1%
TOTAL	228	100%

*Sensitive to cyanide concentrations below EPA's proposed Aquatic Life Criteria

Effects to Mussel Populations

There are no host fish for listed mussel species currently identified for which acute effects are anticipated at the criteria values. For species where hosts have not yet been identified, the probability of acute effects to host fish appears to be small. The greatest effect to mussel populations due to host fish effects is anticipated to result from declines in host fish abundance due to adverse effects to their fecundity and juvenile survival caused by exposure to cyanide at chronic criterion levels.

For effects to host fish caused by exposure to cyanide at chronic criterion levels, estimates were derived using methodologies for listed fish (Appendix F). Based on those calculations, host fish species for which adverse effects are anticipated at the chronic criterion may experience a reduction in eggs hatched that could be as much as, but not likely greater than, 31% to 63% compared to unexposed control populations (Table 16). Additionally, reductions in juvenile survival are expected at magnitudes that could be as high as 33% to 72% of control populations. Given the importance of fecundity and juvenile survival on overall effects to population abundance, this magnitude of adverse effect to the reproduction of host fish species is likely to translate into a decreased abundance of affected host fish species.

For listed mussels, any perturbation that limits fertilization rates and survivability of the glochidia, decreases host fish abundance, or decreases host fish community composition is detrimental to mussel population viability and, ultimately, the species as a whole (Downing et al. 1993, Neves 1993, Neves et al. 1997). Densities of host-specialist mussels, particularly those lacking elaborate host-attracting mechanisms, have been correlated to densities of host fish present in two drainage basins of Alabama (Haag and Warren 1998). No correlation was found for host-generalists or host-specialists with attractant lures. These data suggest that mussel species that showed a positive correlation may exhibit a density-dependence with host fish, limited by their abundance. It has been

hypothesized that a gradual underlying decline of host fish abundance may play a major role in the steady decline of endangered mussel populations.

Although adult survival is typically the most influential life stage in population growth models of long-lived species like mussels, modeled effects to changes in reproduction can also have a significant influence on population growth. In simulation modeling performed for the three-ridge mussel (*Amblema plicata*), deterministic methods using life history tables estimated that a 20% drop in the average number of young produced by females would negatively impact the population growth rate, resulting in a yearly population decline of 4.3% (Hart et al. 2004). Simulations incorporating environmental stochasticity predicted a 98% percent decline in the number of individuals after 100 years.

To determine the effects of exposure to chronic criteria concentrations of cyanide on listed mussel populations, species were grouped into one of the following four categories according to the number and diversity of fish species that have been identified as suitable hosts, and the estimated effects on those host species at criteria concentrations:

Category 1. Host-fish obligates, where the host fish is sensitive to cyanide exposure at criteria concentrations;

Category 2. Host-fish generalists or specialists, where most or all of known hosts are sensitive to cyanide exposure at criteria concentrations;

Category 3. No host fish identified; and

Category 4. Few fish hosts identified. While those which are currently known may be either sensitive or insensitive to cyanide at criteria concentrations, there are no data to indicate whether these fish species represent obligate hosts, a significant portion of the species assemblage of host fish, or a minor portion.

Category 1:

The mussel species listed below are host fish obligates. Using the methods described herein, the obligate host fish for these species, the freshwater drum, may experience a reduction in eggs hatched that could be as much as, but not likely greater than, 36% compared to unexposed control populations (Table 16). Additionally, reductions in juvenile survival are expected at magnitudes that could be as high as 40% of control populations. Given the importance of fecundity and juvenile survival on overall effects to population abundance, this magnitude of adverse effect to the reproduction of host fish species is likely to translate into a decreased abundance of the freshwater drum. If the abundance of an obligate host fish species decreases as a result of cyanide exposure, increased glochidia mortality is likely to occur as a result of their inability to attach to a suitable host. Since attachment of glochidia to a suitable host is a rare and necessary event in the mussel reproductive cycle, reductions in host fish abundance are likely to cause adverse impacts to mussel reproduction and population numbers. Since the obligate host fish identified for the following mussel species are likely to exhibit

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population declines at cyanide criteria concentrations, these mussel species are likely to be subject to reduced reproduction, numbers, and distribution:

Scaleshell Mussel *Leptodea leptodon*
Fat Pocketbook *Potamilus capax*

Category 2

The mussel species listed below are either host fish specialists or generalists. Using the methods described herein, most of the identified host fish species for these mussels are likely to be subject to reduced levels of reproduction and juvenile survival as a result of exposure to cyanide at criteria concentrations (Table 16). Some host fish species identified for these mussels may not experience adverse effects to reproduction and juvenile survival at these concentrations (Table 16). There may also be host fish for these mussels which have yet to be identified that will be either be sensitive to or tolerant of cyanide exposure at criteria concentrations. If the abundance of any host fish species decreases as a result of cyanide exposure, increased glochidia mortality is likely to occur as a result of their inability to attach to a suitable host or the host dies during infestation. Since attachment of glochidia to a suitable host is a rare and necessary event in the mussel reproductive cycle, reductions in host fish abundance or the death of individual host fish that are infested by glochidia are likely to cause adverse impacts to mussel reproduction and population numbers. Since the majority of fish hosts identified for the following mussel species are likely to exhibit population declines at cyanide criteria concentrations, these mussel species are likely to be subject to reduced reproduction, numbers, and distribution:

Cumberland Elktoe	<i>Alasmidonta atropurpurea</i>
Dwarf Wedgemussel	<i>Alasmidonta heterodon</i>
Fat Threeridge	<i>Amblema neislerii</i>
Ouachita Rock Pocketbook	<i>Arkansia wheeleri</i>
Birdwing Pearlymussel	<i>Conradilla caelata</i>
Fanshell	<i>Cyprogenia stegaria</i>
Dromedary Pearlymussel	<i>Dromus dromas</i>
Tar Spiny mussel	<i>Elliptio steinstansana</i>
Purple Bankclimber	<i>Elliptoideus sloatianus</i>
Cumberland Combshell	<i>Epioblasma brevidens</i>
Oyster Mussel	<i>Epioblasma capsaeformis</i>
Curtis Pearlymussel	<i>Epioblasma florentina curtisii</i>
Tan Riffleshell	<i>Epioblasma florentina walkeri</i>
Catspaw (Purple cat's paw pearlymussel)	<i>Epioblasma obliquata obliquata</i>
Northern Riffleshell	<i>Epioblasma turulosa rangiana</i>
Shiny Pigtoe	<i>Fusconaia cor</i>
Fine-rayed Pigtoe	<i>Fusconaia cuneolus</i>
Pink Mucket	<i>Lampsilis abrupta</i>
Fine-lined Pocketbook	<i>Lampsilis altilis</i>
Higgins Eye	<i>Lampsilis higginsii</i>
Orangenacre Mucket	<i>Lampsilis perovalis</i>

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Speckled Pocketbook	<i>Lampsilis streckeri</i>
Shinyrayed pocketbook	<i>Lampsilis subangulata</i>
Louisiana Pearlshell	<i>Margaritifera hembeli</i>
Alabama Moccasinshell	<i>Medionidus acutissimus</i>
Gulf Moccasinshell	<i>Medionidus penicillatus</i>
Littlewing Pearlymussel	<i>Pegis fibula</i>
Clubshell	<i>Pleurobema clava</i>
James Spiny mussel	<i>Pleurobema collina</i>
Southern Clubshell	<i>Pleurobema decisum</i>
Dark Pigtoe	<i>Pleurobema furvum</i>
Southern Pigtoe	<i>Pleurobema georgianum</i>
Oval Pigtoe	<i>Pleurobema pyriforme</i>
Triangular Kidneyshell	<i>Ptychobranhus greeni</i>
Rough Rabbitsfoot	<i>Quadrula cylindrical strigillata</i>
Purple Bean	<i>Villosa perpurpurea</i>

Category 3

No fish hosts have been identified for 23 of the listed mussel species considered in this biological opinion. While host fish are a necessary factor in the reproduction of freshwater mussels, there has not been adequate study to identify which fish can serve for hosts for a significant portion of North American mussels, including those which are threatened and endangered. In cases of uncertainty, it is Service policy to error on the side of listed species. Therefore, for purposes of this analysis, we are assuming that the mussel species in this category are either host fish specialists or generalists but that all of the host fish species are sensitive to cyanide exposure at criteria concentrations to an extent that their populations are likely to be reduced. This effect is likely to cause increased glochidia mortality due to their inability to attach to a suitable host or death of the host. Since attachment of glochidia to a suitable host is a rare and necessary event in the mussel reproductive cycle, reductions in host fish abundance or death of individual host fish that are infested by glochidia are likely to cause declines in the reproduction, numbers, and distribution of the following mussel species:

Yellow Blossom	<i>Epioblasma florentina florentina</i>
Upland Combshell	<i>Epioblasma metastriata</i>
White Catspaw Pearlymussel	<i>Epioblasma obliquata perobliqua</i>
Southern Acornshell	<i>Epioblasma othcaloogensis</i>
Southern Combshell	<i>Epioblasma penita</i>
Tubercled Blossom	<i>Epioblasma torulosa torulosa</i>
Turgid Blossom	<i>Epioblasma turgidula</i>
Green Blossom	<i>Epioblasma torulosa gubernaculum</i>
Cracking Pearlymussel	<i>Hemistena lata</i>
Alabama Lampmussel	<i>Lampsilis virescens</i>
Carolina Heelsplitter	<i>Lasmigon decorate</i>
Ochlockonee Moccasinshell	<i>Medionidus simpsonianus</i>
Ring Pink	<i>Obovaria retusa</i>
White Wartyback	<i>Plethobasus cicatricosus</i>

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Orangefoot Pimpleback	<i>Plethobasus cooperianus</i>
Black Clubshell	<i>Pleurobema curtum</i>
Flat Pigtoe	<i>Pleurobema marshalli</i>
Ovate Clubshell	<i>Pleurobema perovatum</i>
Rough Pigtoe	<i>Pleurobema plenum</i>
Heavy Pigtoe	<i>Pleurobema taitianum</i>
Appalachian Monkeyface	<i>Quadrula sparsa</i>
Stirrupshell	<i>Quadrula stapes</i>
Pale Lilliput	<i>Toxolasma cylindrellus</i>

Category 4:

For several species, few host fish have been identified. While those which are currently known may be either sensitive or insensitive to cyanide at criteria concentrations, there are no data to indicate whether these fish represent obligate hosts, a significant portion of the species assemblage of host fish, or a minor portion. As noted above, in cases of uncertainty, it is Service policy to error on the side of listed species. Therefore, for purposes of this analysis, we are assuming that the mussel species in this category are either host fish specialists or generalists but that many or all of the host fish species are sensitive to cyanide exposure at criteria concentrations to an extent that their populations are likely to be reduced. This effect is likely to cause increased glochidia mortality due to their inability to attach to a suitable host or death of the host. Since attachment of glochidia to a suitable host is a rare and necessary event in the mussel reproductive cycle, reductions in host fish abundance or death of individual host fish that are infested by glochidia are likely to cause declines in the reproduction, numbers, and distribution of the following mussel species:

Appalachian Elktoe	<i>Alasmidonta raveneliana</i>
Chipola Slabshell	<i>Elliptio chipolaensis</i>
Arkansas Fatmucket	<i>Lampsilis powelli</i>
Coosa Moccasinshell	<i>Medionidus parvulus</i>
Cumberland Pigtoe	<i>Pleurobema gibberum</i>
Alabama Heelsplitter	<i>Potamilus inflatus</i>
Winged Mapleleaf	<i>Quadrula fragosa</i>
Cumberland Monkeyface	<i>Quadrula intermedia</i>
Cumberland Bean	<i>Villosa trabalis</i>

Effects to Critical Habitat for Listed Mussels

Category 1 Species:

No critical habitat has been designated for species in this category.

Category 2 Species:

The physical and biological features of critical habitat essential to the conservation of mussels include water of sufficient quality for normal behavior, growth, and survival of all life stages of the mussel and its host fish. Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to these levels. This

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approval is likely to adversely affect the quality of water within critical habitat for the following mussel species to the degree that it would impair individual reproduction and survival of host fish, and cause host fish to experience adverse effects to growth, swimming performance, condition, and development (Table 16). For those reasons, we conclude that management of cyanide to the CCC level in water within critical habitat for the following mussel species is likely to degrade or preclude the proper function of the primary constituent elements of critical habitat that support water quality for normal behavior, growth, and survival of all life stages of the mussel and its host fish:

Cumberland Elktoe	<i>Alasmidonta atropurpurea</i>
Fat Threeridge	<i>Amblema neislerii</i>
Purple Bankclimber	<i>Elliptoideus sloatianus</i>
Cumberlandian Combshell	<i>Epioblasma brevidens</i>
Oyster Mussel	<i>Epioblasma capsaeformis</i>
Fine-lined Pocketbook	<i>Lampsilis altilis</i>
Orangenacre Mucket	<i>Lampsilis perovalis</i>
Shinyrayed pocketbook	<i>Lampsilis subangulata</i>
Alabama Moccasinshell	<i>Medionidus acutissimus</i>
Gulf Moccasinshell	<i>Medionidus penicillatus</i>
Southern Clubshell	<i>Pleurobema decisum</i>
Dark Pigtoe	<i>Pleurobema furvum</i>
Southern Pigtoe	<i>Pleurobema georgianum</i>
Oval Pigtoe	<i>Pleurobema pyriforme</i>
Triangular Kidneyshell	<i>Ptychobranthus greeni</i>
Rough Rabbitsfoot	<i>Quadrula cylindrical strigillata</i>
Purple Bean	<i>Villosa perpurpurea</i>

Category 3 Species:

The physical and biological features of critical habitat essential to the conservation of mussels include water of sufficient quality for normal behavior, growth, and survival of all life stages of the mussel and its host fish. Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to these levels. This approval is likely to adversely affect the quality of water within critical habitat for the following mussel species to the degree that it would impair individual reproduction and survival of potential host fish, and cause host fish to experience adverse effects to growth, swimming performance, condition, and development (Table 16). For those reasons, we conclude that management of cyanide to the CCC level in water within critical habitat for the following mussel species is likely to degrade or preclude the proper function of the primary constituent elements of critical habitat that support water quality for normal behavior, growth, and survival of all life stages of the mussel and its host fish:

Upland Combshell	<i>Epioblasma metastrata</i>
Southern Acornshell	<i>Epioblasma othcaloogensis</i>
Carolina Heelsplitter	<i>Lasmigon decorate</i>
Ochlockonee Moccasinshell	<i>Medionidus simpsonianus</i>
Ovate Clubshell	<i>Pleurobema perovatum</i>

Category 4 Species:

The physical and biological features of critical habitat essential to the conservation of mussels include water of sufficient quality for normal behavior, growth, and survival of all life stages of the mussel and its host fish. Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to these levels. This approval is likely to adversely affect the quality of water within critical habitat for the following mussel species to the degree that it would impair individual reproduction and survival of known and/or potential host fish, and cause host fish to experience adverse effects to growth, swimming performance, condition, and development (Table 16). For those reasons, we conclude that management of cyanide to the CCC level in water within critical habitat for the following mussel species is likely to degrade or preclude the proper function of the primary constituent elements of critical habitat that support water quality for normal behavior, growth, and survival of all life stages of the mussel and its host fish:

Appalachian Elktoe
Chipola Slabshell
Coosa Moccasinshell

Alasmidonta raveneliana
Elliptio chipolaensis
Medionidus parvulus

7.4 Effects to Amphibians

Our assessment of the sensitivity of listed amphibian species to cyanide was based on multiple lines of evidence. First, we evaluated the available information on cyanide-induced effects on amphibians. We then reviewed the approach EPA used in their Biological Evaluation to assess the sensitivity of listed amphibians to cyanide and the protectiveness of the cyanide criteria. Next, we examined additional toxicity information for amphibians, not used by EPA, and constructed regression models for predicting the acute sensitivities of amphibian genera to cyanide. Finally, we compared the predicted sensitivity of amphibians with that of rainbow trout; the most sensitive freshwater species (based on measured cyanide LC₅₀s) and the species that was used to set the acute and chronic cyanide criteria. Taken together, these data provided the basis for our effects determination.

The scientific literature for cyanide toxicity to amphibians is limited and somewhat dated. Early investigators studied the effects of cyanide on amphibian development. These experiments were generally focused on early embryogenesis including oviposited and fertilized egg morphogenesis and post gastrulation development. Repressive effects of cyanide on embryonic respiration and development were documented by several authors (Spiegelman and Moog, 1943, Lovtrup and Pigon, 1958, Nakatsuji, 1974). Others used sub-lethal exposure concentrations of cyanide as a mechanism to arrest or retard development in order to test various hypotheses regarding metabolism or physiology (Spiegelman and Steinbach, 1945; Ornstein and Gregg, 1952). Although these historical studies are important for understanding the physiological actions of cyanide on amphibians, they do not provide the traditional quantitative measures of acute and chronic toxicity (i.e. LC₅₀s, NOECs, EC_xs) that have been used in water quality criteria development.

Because cyanide-specific toxicity data (LC₅₀s) for amphibians were not available, EPA based their effects analysis on the relative sensitivity of amphibians to other pollutants (EPA 2007). They examined the rank order of amphibian LC₅₀s for seven water pollutants using data sets from ambient water quality criteria documents (Table 20). The 7 data sets included LC₅₀s for 9 amphibian species (in total), although 4 of the data sets contained LC₅₀s for only 1 amphibian species and the other 3 data sets contained data for 2 species. So among these seven criteria documents, the amphibian class was represented by no more than one or two species at a time. With so few species used to characterize the sensitivity of an entire class there is considerable uncertainty as to whether the most sensitive amphibian species are adequately represented, especially considering the large interspecies variability in cyanide toxicity observed for other taxa (see acute effects section of BO). It seems highly unlikely that the amphibians species included in these data sets were among the most sensitive amphibians. Nevertheless, for two of the seven pollutants the single amphibian species in the data set ranked among the most sensitive species/genera in the multi-taxa data sets used to develop criteria. For the remaining five pollutants the GMAVs for amphibians ranged from the 26th percentile to the 100th percentile. Considering the low number of species used to represent amphibians in the analysis and the fact that amphibians were among the most sensitive species/genera for 28% of the pollutants examined we believe that there is a more than a discountable chance that some amphibian species may be highly sensitive to cyanide. Therefore, we do not believe these results alone support EPA's determination that the listed amphibian species are not likely to be adversely affected by cyanide at criteria concentrations.

To better understand how to interpret the results from EPA's analysis we extended our evaluation to include rainbow trout; a species frequently included in criteria development and often among the more sensitive species tested (Table 20). Using data for the same seven pollutants we found that the over all pattern of rankings for rainbow trout were much like those for amphibians, i.e. most near or above the median and two or three falling among the most sensitive species. However we know that in terms of cyanide, rainbow trout is the most sensitive freshwater species that has been tested, more sensitive than the 5th percentile estimated species (EPA 1985). (That is, rainbow trout fell in the "sensitive tail" of the species sensitivity distribution.) So, there is at least one example where the "ranking profile" (for these 7 pollutants) shared by amphibians and rainbow trout was associated with a species that was highly sensitive to cyanide. In addition, we found that for these seven pollutants amphibian species were more sensitive than rainbow trout 43% of the time (3 of 7). To further investigate the relative sensitivity of amphibians to other taxa we reviewed other references on amphibian toxicology.

Table 20. Rank and corresponding percentile of GMAVs (genus mean acute values) for amphibians and rainbow trout versus all aquatic taxa and chordates (fishes) only. Data for amphibians are from Appendix D of EPAs Cyanide Biological Evaluation (EPA 2007). Data for rainbow trout are from criteria documents (see footnotes).

Chemical	Amphibian Species	Amphibian GMAV Rank Vs. Other Taxa	Rainbow (GMAV) Rank vs. Other Taxa	Percentile (Amphibians)	Percentile (Rainbow trout)	Amphibians more (+) or less (-) sensitive than Rainbow trout
Atrazine	Bufo americanus	11 of 19	4 of 19 ¹	0.58	0.21	-
Atrazine	Rana sp.	14 of 19		0.74		
Cadmium	Ambystoma gracile	29 of 57	4 of 57 ²	0.51	0.07	-
Cadmium	Xenopus laevis	33 of 57		0.58		
Diazinon	Rana clamitans	8 of 21	12 of 21 ³	0.26	0.57	+
Lindane	Pseudacris triseriata	22 of 23	10 of 23 ⁴	0.96	0.43	-
Lindane	Bufo woodhousei	23 of 23		1.00		
Nonylphenol	Bufo boreas	2 of 15	8 of 15 ⁵	0.13	0.53	+
Parathion	Pseudacris triseriata	23 of 31	25 of 31 ⁵	0.74	0.81	+
Pentachlorophenol	Rana satesbeiana	4 ⁶ of 32	3 of 32 ⁵	0.13	0.09	-

¹ Draft aquatic life ambient water quality criteria for atrazine (EPA 2003)

² 2001 update of the aquatic life ambient water quality criteria for cadmium (EPA 2001)

³ Aquatic life ambient water quality criteria for diazinon (EPA 2005)

⁴ 1995 updates: water quality criteria documents for the protection of aquatic life in ambient water (EPA 1996)

⁵ Aquatic life ambient water quality criteria for nonylphenol (EPA 2005)

⁶ Rank was changed from 5 to 4 based on GMAV ranks for pentachlorophenol (EPA 1996)

Birge et al. (2003) performed a comparative toxicity analysis for 29 amphibian species in contrast to various species of fish. Amphibian testing included seven salamander species (family Ambystomidae) and 22 frog species (families Microhylidae, Hylidae, Ranidae, and Bufonidae). Though no toxicity testing was performed for cyanide, sufficient data was produced to generate comparisons between amphibians and fish for 34 inorganic compounds and 27 organic compounds. Comparisons include all amphibian test species for 50 of these 61 compounds. Although exposure times varied among species due to differences in hatching times, comparable stages of development (eggs, embryos, and early larvae) were included in all tests. Fish species included in this study for which sensitivity to cyanide is known are the rainbow trout ($LC_{50} = 59.22\mu\text{g/g}$), largemouth bass ($101.7\mu\text{g/g}$) and fathead minnow ($138.4\mu\text{g/g}$).

When compared to rainbow trout, LC_{50} values for amphibians were more sensitive 52% of the time for metals (N=203), 36% for organics (N=44), and 49% for all compounds combined (N=247). For largemouth bass, amphibians were more sensitive 83% of the

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time (N=182), 60% for organics (N=15), and 81% for all compounds (N=197). For fathead minnow, amphibians were more sensitive 89% of the time for metals (N=18), 63% for organics (N=24), and 74% for all compounds (N=42). The generally more sensitive species of Microhylidae and Hylidae were not available for toxicity testing for several organic compounds. For the 15 most sensitive amphibian species, LC₅₀ values were below fish values (including species used above, plus channel catfish and goldfish) 74% of the time.

Bridges et al (2002), performed toxicity testing for five compounds on southern leopard frog (*Rana sphenocephala*) tadpoles and compared results with published values for the boreal toad (*Bufo boreas*), rainbow trout, fathead minnow, and bluegill. The two amphibian species showed the highest correlation of LC₅₀ values for the rainbow trout. Correlations for the fathead minnow and bluegill were much weaker. The authors suggest that rainbow trout thus may be the most appropriate species for assessing toxicity to anuran tadpoles. However, the authors also argue that since amphibians are very tolerant to some chemicals, and very sensitive to others, individual toxicity testing is suggested rather than relying on surrogate species.

The comparative toxicity data sets from Birge et al. (2003) provided an opportunity to construct ICE-like regression models that could be used to estimate cyanide LC₅₀s for amphibians (Table 21; Figures 5 and 6). Following EPA guidelines (EPA 2003b), regression models were developed to estimate the sensitivity of two amphibian genera (*Rana* and *Ambystoma*) using rainbow trout as the surrogate species.

Table 21. Estimated cyanide LC₅₀s for two amphibian genera (*Rana* and *Ambystoma*) using rainbow trout as a surrogate species

Predicted Taxon	Surrogate Species	LCI LC ₅₀ (ug/L)	MLE LC ₅₀ (ug/L)	UCI LC ₅₀ (ug/L)	Corr. Coeff. (r)	MSE	log-log a	log-log b	p	n	Chem.
Rana (genus)	Rainbow Trout	30.82	54.25	95.51	0.789	0.648	0.259	0.832	<0.001	84	32
Ambystoma (genus)	Rainbow Trout	60.56	120.61	639.63	0.638	0.415	0.966	0.629	<0.002	23	4

Figure 6. Rainbow Trout Interspecies Correlation Plot for the Genus Ambystoma

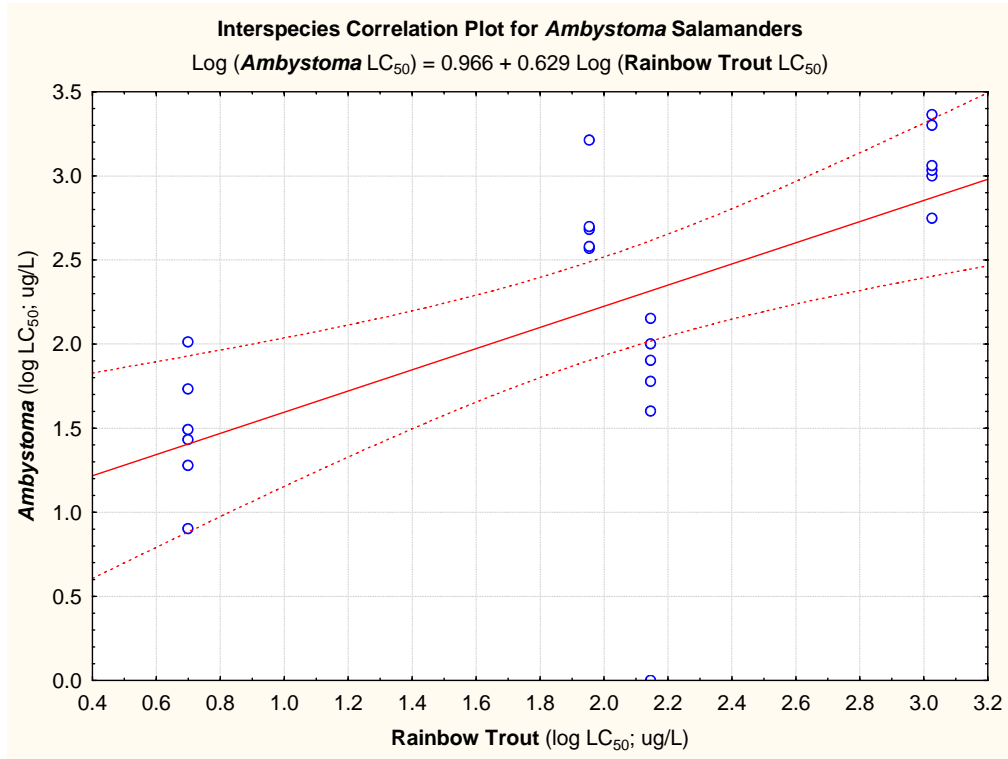
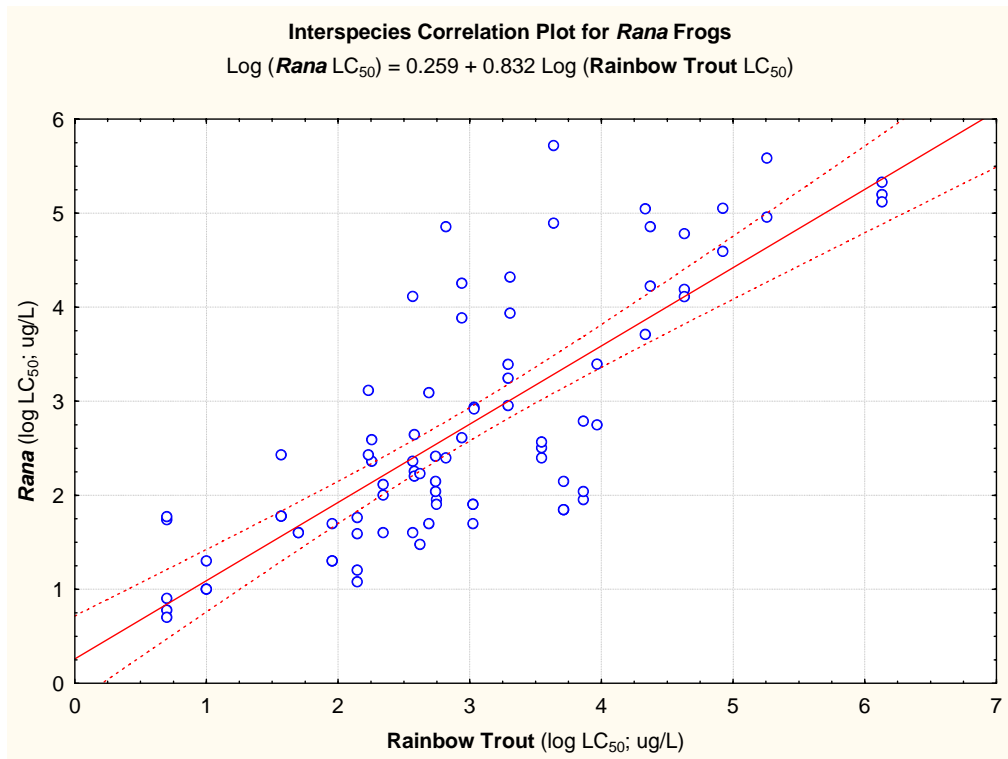


Figure 7. Rainbow Trout Interspecies Correlation Plot for the Genus Rana



Estimated LC₅₀s for the two amphibian genera, *Ambystoma* (LC₅₀ 60.56 ug CN/L) and *Rana* (LC₅₀ 30.82 ug CN/L), are approximately equal to or less than the LC₅₀ for rainbow trout (59 ug CN/L).

As previously mentioned, rainbow trout had the lowest measured cyanide LC₅₀ of all fish species considered in the cyanide criteria document as well as the cyanide BE. Based on the method described in the *Fish* section of Appendix B, the chronic EC_A for rainbow trout would be 2.54 ug CN/L (i.e. 59 ug CN/L / 23.22) and the acute EC_A would be 51.75 ug CN/L (i.e. 59 ug CN/L / 1.14). Because the chronic EC_A is below 5.2 ug CN/L rainbow trout would likely be adversely affected by exposure to cyanide at the CCC. Thus, amphibian species are estimated to be as or more sensitive to cyanide than rainbow trout and thus likely to be adversely affected by exposure to cyanide at the chronic criterion.

Individual Species and Critical Habitat Accounts

Ambystomatidae

RETICULATED FLATWOODS SALAMANDER

Ambystoma bishop

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. Based on multiple lines of evidence, including the relative sensitivity of amphibians to other pollutants, their relative sensitivity to rainbow trout and the sensitivity of rainbow trout to cyanide we conclude that reticulated flatwoods salamanders exposed to cyanide at the CCC could experience substantial reductions in recruitment due to reductions in fecundity, hatching success, and larval development and survival. Compared to control populations, we estimate rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than 52% (Appendix E). We estimate that rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than 61% (Appendix E). Based on the relative sensitivity of amphibians as compared to rainbow trout we estimate the effects of cyanide at the CCC for amphibians could be of similar or greater magnitude than effects estimated for rainbow trout. As such, reticulated flatwoods salamanders exposed to cyanide at the CCC could experience a substantial reduced egg hatching and decreased survival to metamorphosis of larval salamanders.

Although no comparable data are available for amphibians, cyanide may also adversely effect growth, locomotion, condition, and development as described above for fish in the Effects Overview. The effects of chronic toxicity could also be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

The life history of the reticulated flatwoods salamander can be characterized as a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily terrestrial. Reticulated flatwoods salamanders breed in relatively small, isolated ephemeral ponds where the larvae develop until metamorphosis. Post-metamorphic salamanders migrate out of the ponds and into the uplands where they live until they move back to ponds to breed as adults. The flatwoods salamander reproduces at 1 year of age for males and two years of age for females. Males and females court before the breeding sites flood. Females then lay their eggs, either singly or in clumps, beneath leaf litter, under logs, sphagnum moss mats, small trees, bushes or clumps of grass at dry locations in seasonal wetlands. If rainfall is insufficient to result in adequate pond flooding, breeding may not occur or, if larvae do develop, they may die before metamorphosis. Egg development from deposition to hatching occurs in approximately 2 weeks, but eggs do not hatch until they are inundated. Depending on when they are inundated, the larvae metamorphose 11 to 18 weeks after hatching. Exposure to cyanide at the CCC could result in a substantial reduction in egg hatching and survival to metamorphosis. Larval salamanders that do survive could experience reduced growth rates and reduced swimming abilities which would increase their vulnerability to a host of potential stressors including, temperature, flow, predation, and inter- and intraspecific competition for food and cover. Reduced growth rates could also delay reproductive maturity and productivity. Amphibian larvae must grow to a critical minimum body size before they can metamorphose to the terrestrial stage, and in the case of the reticulated flatwoods salamander such reduced growth rates could preclude emergence prior to pond drying.

We anticipate that the estimated reductions in hatched eggs and larval survival would reduce the reticulated flatwoods salamander's fertility rates substantially. The salamander's fertility rates would be reduced based upon (a) reductions in numbers of eggs hatched, (b) reductions in larvae surviving through the first year and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time between mating.

Reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the CCC. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). Taylor et al. (2006) constructed a model, based on extensive population data available for the marbled salamander (*Ambystoma opacum*), to look at how many years of reproductive failure would be required to result in local extinction of pond-breeding salamanders (with varying lifespans) and found that even without total reproductive failure, populations required moderate to high upland post-metamorphic survival to persist. Catastrophic reproductive failure in this study created fluctuations in the population, raised the threshold of survival required to achieve persistence, and imposed the possibility of extinction even under otherwise favorable environmental conditions. Even in populations with multiple breeding ponds, amphibian populations may be unable to recolonize areas after local extirpations due to their physiological

constraints, relatively low mobility, and site fidelity. In the case of the reticulated flatwoods salamander, only 20 populations are known and 14 (70 percent) of these populations are supported by only one breeding site. For those populations with only one breeding pond, habitat destruction associated with cyanide at CCC levels may adversely effect flatwoods salamander reproduction and survival resulting in extirpation of the population supported by that breeding pond. For populations with more than one breeding pond, habitat destruction associated with cyanide at CCC levels may result in a reduction of available breeding sites leading to a reduction in population size and range and an increased vulnerability to catastrophic events that may adversely affect breeding and survival.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the reticulated flatwoods salamander's reproduction by reducing the number of eggs laid by females, the hatchability of laid eggs, and reduction of survivorship of larval salamanders. Salamanders may also experience effects on growth, locomotion, condition, and development. The reticulated flatwoods salamander has a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily terrestrial. Although exposure to cyanide concentrations at the chronic criterion may be isolated to the aquatic phases of the salamander's life cycle (i.e. breeding activities and egg and larval development), the long-term effects of exposure could result in reduced adult growth, survival and fitness. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected flatwoods salamander population's decline could stabilize at a reduced absolute population number or could continue to decline until it's extirpated. The majority of extant populations are supported by a single breeding pond, reproductive failure at that site could result in the extirpation of the population and even those populations with multiple breeding sites may have difficulty recolonizing after local extirpations due to physiological, morphological, and behavioral constraints. Based upon the magnitude of effect we anticipate could occur, we conclude that ultimately, reticulated flatwoods salamanders are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the reticulated salamander.

Critical Habitat: Critical habitat for the reticulated flatwoods salamander has been designated in: Calhoun, Holmes, Jackson, Santa Rosa, Walton, and Washington Counties Florida; Baker and Miller Counties Georgia. The physical and biological features of critical habitat essential to the conservation of the reticulated flatwoods salamander includes breeding habitats consisting of small, acidic depressional standing bodies of water that are seasonally flooded by rainfall in late fall or early winter and dry in late spring or early summer; and are geographically isolated from other water bodies. In order for these aquatic areas to provide the necessary conditions for successful breeding and survival they must include water of sufficient quality to allow the species to carry out normal feeding, movement, sheltering, and reproductive behaviors and to experience individual and population growth. Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentrations. This approval could adversely affect the quality of water to

the degree that it would impair individual reproduction and survival of reticulated flatwoods salamanders and cause salamanders to experience adverse effects to growth, locomotion, condition, and development. Based on data for the rainbow trout, our surrogate species for assessing impacts to amphibians, we estimate the reduction in number of hatched eggs could be as much as, but is not likely to be greater than 52% and the reduction in larval survival to metamorphosis could be as much as, but is not likely to be greater than 61%. These effects are estimated to be of a magnitude great enough to reduce numbers of reticulated flatwoods salamanders and result in the extirpation of local populations. Approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the species.

FROSTED FLATWOODS SALAMANDER

Ambystoma cingulatum

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. Based on multiple lines of evidence, including the relative sensitivity of amphibians to other pollutants, their relative sensitivity to rainbow trout and the sensitivity of rainbow trout to cyanide we conclude that frosted flatwoods salamanders exposed to cyanide at the CCC could experience substantial reductions in recruitment due to reductions in fecundity, hatching success, and larval development and survival. Compared to control populations, we estimate rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than 52% (Appendix E). We estimate that rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than 61% (Appendix E). Based on the relative sensitivity of amphibians as compared to rainbow trout we estimate the effects of cyanide at the CCC for amphibians could be of similar or greater magnitude than effects estimated for rainbow trout. As such, frosted flatwoods salamanders exposed to cyanide at the CCC could experience a substantial reduced egg hatching and decreased survival to metamorphosis of larval salamanders.

Although no comparable data are available for amphibians, cyanide may also adversely effect growth, locomotion, condition, and development as described above for fish in the Effects Overview. The effects of chronic toxicity could also be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

The life history of the frosted flatwoods salamander can be characterized as a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily terrestrial. Frosted flatwoods salamanders breed in relatively small, isolated ephemeral ponds where the larvae develop until metamorphosis. Post-metamorphic salamanders migrate out of the ponds and into the uplands where they live until they

move back to ponds to breed as adults. The flatwoods salamander reproduces at 1 year of age for males and two years of age for females. Males and females court before the breeding sites flood. Females then lay their eggs, either singly or in clumps, beneath leaf litter, under logs, sphagnum moss mats, small trees, bushes or clumps of grass at dry locations in seasonal wetlands. If rainfall is insufficient to result in adequate pond flooding, breeding may not occur or, if larvae do develop, they may die before metamorphosis. Egg development from deposition to hatching occurs in approximately 2 weeks, but eggs do not hatch until they are inundated. Depending on when they are inundated, the larvae metamorphose 11 to 18 weeks after hatching. Exposure to cyanide at the CCC could result in a substantial reduction in egg hatching and survival to metamorphosis. Larval salamanders that do survive could experience reduced growth rates and reduced swimming abilities which would increase their vulnerability to a host of potential stressors including, temperature, flow, predation, and inter- and intraspecific competition for food and cover. Reduced growth rates could also delay reproductive maturity and productivity. Amphibian larvae must grow to a critical minimum body size before they can metamorphose to the terrestrial stage, and in the case of the frosted flatwoods salamander such reduced growth rates could preclude emergence prior to pond drying.

We anticipate that the estimated reductions in hatched eggs and larval survival would reduce the frosted flatwoods salamander's fertility rates substantially. The salamander's fertility rates would be reduced based upon (a) reductions in numbers of eggs hatched, (b) reductions in larvae surviving through the first year and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time between mating.

Reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the CCC. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). Taylor et al. (2006) constructed a model, based on extensive population data available for the marbled salamander (*Ambystoma opacum*), to look at how many years of reproductive failure would be required to result in local extinction of pond-breeding salamanders (with varying lifespans) and found that even without total reproductive failure, populations required moderate to high upland post-metamorphic survival to persist. Catastrophic reproductive failure in this study created fluctuations in the population, raised the threshold of survival required to achieve persistence, and imposed the possibility of extinction even under otherwise favorable environmental conditions. Even in populations with multiple breeding ponds, amphibian populations may be unable to recolonize areas after local extirpations due to their physiological constraints, relatively low mobility, and site fidelity. Surveys indicate there are 25 populations of the frosted flatwoods salamander, some of which have been inferred from the capture of a single individual. Twenty-two (88 percent) of the known frosted flatwoods salamander populations occur primarily on public land. Sixteen of the populations (64 percent of total populations of the species) on public land represent

metapopulations supported by more than one breeding site. For populations with only one breeding pond, if the habitat at that site is destroyed, recolonization would be impossible and the population supported by that breeding pond would be extirpated. For those populations with only one breeding pond, habitat destruction associated with cyanide at CCC levels may adversely effect flatwoods salamander reproduction and survival resulting in extirpation of the population supported by that breeding pond. For populations with more than one breeding pond, habitat destruction associated with cyanide at CCC levels may result in a reduction of available breeding sites leading to a reduction in population size and range and an increased vulnerability to catastrophic events that may adversely affect breeding and survival.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the frosted flatwoods salamander's reproduction by reducing the number of eggs laid by females, the hatchability of laid eggs, and reduction of survivorship of larval salamanders. Salamanders may also experience effects on growth, locomotion, condition, and development. The frosted flatwoods salamander has a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily terrestrial. Although exposure to cyanide concentrations at the chronic criterion may be isolated to the aquatic phases of the salamander's life cycle (i.e. breeding activities and egg and larval development), the long-term effects of exposure could result in reduced adult growth, survival and fitness. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected flatwoods salamander population's decline could stabilize at a reduced absolute population number or could continue to decline until it's extirpated. The majority of extant populations are supported by a single breeding pond, reproductive failure at that site could result in the extirpation of the population and even those populations with multiple breeding sites may have difficulty recolonizing after local extirpations due to physiological, morphological, and behavioral constraints. Based upon the magnitude of effect we anticipate could occur, we conclude that ultimately, frosted flatwoods salamanders are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the frosted flatwoods salamander.

Critical Habitat: Critical habitat has been designated for the frosted flatwoods salamander in: Baker, Franklin, Jefferson, Liberty, and Wakulla Counties, Florida; and in Berkeley, Charleston, and Jasper Counties, South Carolina. The physical and biological features of critical habitat essential to the conservation of the frosted flatwoods salamander includes breeding habitats consisting of small, acidic depressional standing bodies of water that are seasonally flooded by rainfall in late fall or early winter and dry in late spring or early summer; and are geographically isolated from other water bodies. In order for these aquatic areas to provide the necessary conditions for successful breeding and survival they must include water of sufficient quality to allow the species to carry out normal feeding, movement, sheltering, and reproductive behaviors and to experience individual and population growth. Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentrations. This approval could adversely affect the quality

of water to the degree that it would impair individual reproduction and survival of frosted flatwoods salamanders and cause salamanders to experience adverse effects to growth, locomotion, condition, and development. Based on data for the rainbow trout, our surrogate species for assessing impacts to amphibians, we estimate the reduction in number of hatched eggs could be as much as, but is not likely to be greater than 52% and the reduction in larval survival to metamorphosis could be as much as, but is not likely to be greater than 61%. These effects are estimated to be of a magnitude great enough to reduce numbers of frosted flatwoods salamanders and result in the extirpation of local populations. Approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the species.

CALIFORNIA TIGER SALAMANDER

Ambystoma californiense

Central California population

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. Based on multiple lines of evidence, including the relative sensitivity of amphibians to other pollutants, their relative sensitivity to rainbow trout and the sensitivity of rainbow trout to cyanide we conclude that California tiger salamanders exposed to cyanide at the CCC could experience substantial reductions in recruitment due to reductions in fecundity, hatching success, and larval development and survival. Compared to control populations, we estimate rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than 52% (Appendix E). We estimate that rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than 61% (Appendix E). Based on the relative sensitivity of amphibians as compared to rainbow trout we estimate the effects of cyanide at the CCC for amphibians could be of similar or greater magnitude than effects estimated for rainbow trout. As such, California tiger salamanders exposed to cyanide at the CCC could experience a substantial reduced egg hatching and decreased survival to metamorphosis of larval salamanders.

Although no comparable data are available for amphibians, cyanide may also adversely effect growth, locomotion, condition, and development as described above for fish in the Effects Overview. The effects of chronic toxicity could also be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

The life history of the California tiger salamander can be characterized as a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily terrestrial. Adult California tiger salamanders mate in vernal pools and similar water

bodies, and the females lay their eggs in the water. Females attach their eggs singly or, in rare circumstances, in groups of two to four, to twigs, grass stems, vegetation, or debris. In ponds with little or no vegetation, females may attach eggs to objects, such as rocks and boards on the bottom. After breeding, adults leave the pool and return to small mammal burrows in surrounding uplands, although they may continue to come out nightly for approximately the next two weeks to feed. In drought years, the seasonal pools may not form and the adults may not breed. The eggs hatch in 10 to 14 days with newly hatched salamanders (larvae) ranging in size from 11.5 to 14.2 mm (0.5 to 0.6 in) in total length. The larval stage of the California tiger salamander usually lasts three to six months, because most seasonal ponds and pools dry up during the summer, although some larvae in Contra Costa and Alameda Counties may remain in their breeding sites over the summer. Amphibian larvae must grow to a critical minimum body size before they can metamorphose (change into a different physical form) to the terrestrial stage. One study found larvae metamorphosed and left the breeding pools 60 to 94 days after the eggs had been laid, with larvae developing faster in smaller, more rapidly drying pools. The longer the inundation period, the larger the larvae and metamorphosed juveniles are able to grow, and the more likely they are to survive and reproduce. The larvae perish if a site dries before they complete metamorphosis. There was a strong positive correlation between inundation period and total number of metamorphosing juvenile amphibians, including tiger salamanders. Size at metamorphosis is positively correlated with stored body fat and survival of juvenile amphibians, and negatively correlated with age at first reproduction

Exposure to cyanide at the CCC could result in a substantial reduction in egg hatching and survival to metamorphosis. Larval salamanders that do survive could experience reduced growth rates and reduced swimming abilities which would increase their vulnerability to a host of potential stressors including, temperature, flow, predation, and inter- and intraspecific competition for food and cover. Reduced growth rates could also delay reproductive maturity and productivity. Because amphibian larvae must grow to a critical minimum body size before they can metamorphose to the terrestrial stage, reduced growth rates of the California tiger salamander could preclude emergence prior to pond drying, resulting in larval mortalities.

We anticipate that the estimated reductions in hatched eggs and larval survival would reduce the California tiger salamander's fertility rates substantially. The salamander's fertility rates would be reduced based upon (a) reductions in numbers of eggs hatched, (b) reductions in larvae surviving through the first year and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time between mating.

Reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the CCC. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section of Population Level Effects, we noted that

reduction in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to reproduce, like California tiger salamanders. Lifetime reproductive success for California and other tiger salamanders is low. One study found the average female bred 1.4 times and produced 8.5 young that survived to metamorphosis per reproductive effort. This resulted in roughly 11 metamorphic offspring over the lifetime of a female. Most California tiger salamanders in this study did not reach sexual maturity until four or five years old. While individuals may survive for more than 10 years, many breed only once, and one study estimated that less than five percent of metamorphic juveniles survive to become breeding adults. The mechanisms for recruitment are clearly dependent on a number of factors such as migration, terrestrial survival, and population turnover, whose interaction is not well understood.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the California tiger salamander's reproduction by reducing the number of eggs laid by females, the hatchability of laid eggs, and a reduction of survivorship of larval salamanders. Salamanders may also experience effects on growth, locomotion, condition, and development. The California tiger salamander has a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily terrestrial. Although exposure to cyanide concentrations at the chronic criterion may be isolated to the aquatic phases of the salamander's life cycle (i.e. breeding activities and egg and larval development), the long-term effects of exposure could result in reduced adult growth, survival and fitness. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected California tiger salamander population's decline could stabilize at a reduced absolute population number or could continue to decline until it's extirpated. For extant populations supported by a single breeding pond, reproductive failure at that site could result in the extirpation of the population and even those populations with multiple breeding sites may have difficulty recolonizing after local extirpations due to physiological, morphological, and behavioral constraints. Based upon the magnitude of effect we anticipate could occur, we conclude that ultimately, California tiger salamanders are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the California Tiger salamander.

Critical Habitat: The exact locations of the critical habitat are depicted on maps in the federal register. The physical and biological features of critical habitat essential to the conservation of the California tiger salamander includes standing bodies of fresh water (including natural and manmade (e.g., stock)) ponds, vernal pools, and other ephemeral or permanent water bodies which typically support inundation during winter rains and hold water for a minimum of 12 weeks in a year of average rainfall. In order for these aquatic areas to provide the necessary conditions for successful breeding and survival they must include water of sufficient quality to allow the species to carry out normal feeding, movement, sheltering, and reproductive behaviors and to experience individual and population growth. Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentrations. This approval could adversely affect the quality of water to the degree

that it would impair individual reproduction and survival of California tiger salamanders and cause salamanders to experience adverse effects to growth, locomotion, condition, and development. Based on data for the genus *Oncorhynchus*, our surrogate species for assessing impacts to amphibians, we estimate the reduction in number of hatched eggs could be as much as, but is not likely to be greater than 52% and the reduction in larval survival to metamorphosis could be as much as, but is not likely to be greater than 61%. These effects are estimated to be of a magnitude great enough to reduce numbers of California tiger salamanders and result in the extirpation of local populations. Approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the species.

CALIFORNIA TIGER SALAMANDER

Ambystoma californiense

Santa Barbara County population

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. Based on multiple lines of evidence, including the relative sensitivity of amphibians to other pollutants, their relative sensitivity to rainbow trout and the sensitivity of rainbow trout to cyanide we conclude that California tiger salamanders exposed to cyanide at the CCC could experience substantial reductions in recruitment due to reductions in fecundity, hatching success, and larval development and survival. Compared to control populations, we estimate rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than 52% (Appendix E). We estimate that rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than 61% (Appendix E). Based on the relative sensitivity of amphibians as compared to rainbow trout we estimate the effects of cyanide at the CCC for amphibians could be of similar or greater magnitude than effects estimated for rainbow trout. As such, California tiger salamanders exposed to cyanide at the CCC could experience a substantial reduced egg hatching and decreased survival to metamorphosis of larval salamanders.

Although no comparable data are available for amphibians, cyanide may also adversely effect growth, locomotion, condition, and development as described above for fish in the Effects *Overview*. The effects of chronic toxicity could also be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

This population is restricted to Santa Barbara County, California. The life history of the California tiger salamander can be characterized as a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily terrestrial. Adult California tiger salamanders mate in vernal pools and similar water bodies, and the

females lay their eggs in the water. Females attach their eggs singly or, in rare circumstances, in groups of two to four, to twigs, grass stems, vegetation, or debris. In ponds with little or no vegetation, females may attach eggs to objects, such as rocks and boards on the bottom. After breeding, adults leave the pool and return to small mammal burrows in surrounding uplands, although they may continue to come out nightly for approximately the next two weeks to feed. In drought years, the seasonal pools may not form and the adults may not breed. The eggs hatch in 10 to 14 days with newly hatched salamanders (larvae) ranging in size from 11.5 to 14.2 mm (0.5 to 0.6 in) in total length. The larval stage of the California tiger salamander usually lasts three to six months, because most seasonal ponds and pools dry up during the summer, although some larvae in Contra Costa and Alameda Counties may remain in their breeding sites over the summer. Amphibian larvae must grow to a critical minimum body size before they can metamorphose (change into a different physical form) to the terrestrial stage. One study found larvae metamorphosed and left the breeding pools 60 to 94 days after the eggs had been laid, with larvae developing faster in smaller, more rapidly drying pools. The longer the inundation period, the larger the larvae and metamorphosed juveniles are able to grow, and the more likely they are to survive and reproduce. The larvae perish if a site dries before they complete metamorphosis. There was a strong positive correlation between inundation period and total number of metamorphosing juvenile amphibians, including tiger salamanders. Size at metamorphosis is positively correlated with stored body fat and survival of juvenile amphibians, and negatively correlated with age at first reproduction

Exposure to cyanide at the CCC could result in a substantial reduction in egg hatching and survival to metamorphosis. Larval salamanders that do survive could experience reduced growth rates and reduced swimming abilities which would increase their vulnerability to a host of potential stressors including, temperature, flow, predation, and inter- and intraspecific competition for food and cover. Reduced growth rates could also delay reproductive maturity and productivity. Because amphibian larvae must grow to a critical minimum body size before they can metamorphose to the terrestrial stage, reduced growth rates of the California tiger salamander could preclude emergence prior to pond drying, resulting in larval mortalities.

We anticipate that the estimated reductions in hatched eggs and larval survival would reduce the California tiger salamander's fertility rates substantially. The salamander's fertility rates would be reduced based upon (a) reductions in numbers of eggs hatched, (b) reductions in larvae surviving through the first year and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time between mating.

Reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the CCC. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section of Population Level Effects, we noted that

reduction in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to reproduce, like California tiger salamanders. Lifetime reproductive success for California and other tiger salamanders is low. One study found the average female bred 1.4 times and produced 8.5 young that survived to metamorphosis per reproductive effort. This resulted in roughly 11 metamorphic offspring over the lifetime of a female. Most California tiger salamanders in this study did not reach sexual maturity until four or five years old. While individuals may survive for more than 10 years, many breed only once, and one study estimated that less than five percent of metamorphic juveniles survive to become breeding adults. The mechanisms for recruitment are clearly dependent on a number of factors such as migration, terrestrial survival, and population turnover, whose interaction is not well understood.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the California tiger salamander's reproduction by reducing the number of eggs laid by females, the hatchability of laid eggs, and a reduction of survivorship of larval salamanders. Salamanders may also experience effects on growth, locomotion, condition, and development. The California tiger salamander has a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily terrestrial. Although exposure to cyanide concentrations at the chronic criterion may be isolated to the aquatic phases of the salamander's life cycle (i.e. breeding activities and egg and larval development), the long-term effects of exposure could result in reduced adult growth, survival and fitness. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected California tiger salamander population's decline could stabilize at a reduced absolute population number or could continue to decline until it's extirpated. For extant populations supported by a single breeding pond, reproductive failure at that site could result in the extirpation of the population and even those populations with multiple breeding sites may have difficulty recolonizing after local extirpations due to physiological, morphological, and behavioral constraints. Based upon the magnitude of effect we anticipate could occur, we conclude that ultimately, California tiger salamanders are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the California Tiger salamander.

Critical Habitat: Critical habitat has been established in Santa Barbara County for this population segment of the California tiger salamander in six separate locations east of Vandenberg Air Force Base. The physical and biological features of critical habitat essential to the conservation of the California tiger salamander includes standing bodies of fresh water (ponds, vernal pools, dune ponds, or other ephemeral or permanent water bodies) sufficient for the aquatic portion of the salamander's life cycle. In order for these aquatic areas to provide the necessary conditions for successful breeding and survival they must include water of sufficient quality to allow the species to carry out normal feeding, movement, sheltering, and reproductive behaviors and to experience individual and population growth. Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentrations. This approval could adversely affect the quality of water to the degree

that it would impair individual reproduction and survival of California tiger salamanders and cause salamanders to experience adverse effects to growth, locomotion, condition, and development. Based on data for the rainbow trout, our surrogate species for assessing impacts to amphibians, we estimate the reduction in number of hatched eggs could be as much as, but is not likely to be greater than 52% and the reduction in larval survival to metamorphosis could be as much as, but is not likely to be greater than 61%. These effects are estimated to be of a magnitude great enough to reduce numbers of California tiger salamanders and result in the extirpation of local populations. Approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the species.

CALIFORNIA TIGER SALAMANDER

Ambystoma californiense
Sonoma County population

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. Based on multiple lines of evidence, including the relative sensitivity of amphibians to other pollutants, their relative sensitivity to rainbow trout and the sensitivity of rainbow trout to cyanide we conclude that California tiger salamanders exposed to cyanide at the CCC could experience substantial reductions in recruitment due to reductions in fecundity, hatching success, and larval development and survival. Compared to control populations, we estimate rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than 52% (Appendix E). We estimate that rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than 61% (Appendix E). Based on the relative sensitivity of amphibians as compared to rainbow trout we estimate the effects of cyanide at the CCC for amphibians could be of similar or greater magnitude than effects estimated for rainbow trout. As such, California tiger salamanders exposed to cyanide at the CCC could experience a substantial reduced egg hatching and decreased survival to metamorphosis of larval salamanders.

Although no comparable data are available for amphibians, cyanide may also adversely effect growth, locomotion, condition, and development as described above for fish in the Effects Overview. The effects of chronic toxicity could also be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

The Sonoma population appears to have been geographically isolated from the remainder of the California tiger salamander population by distance, mountains and major waterway barriers for more than 700,000 years. It occurs only in association with vernal pool

ecosystems and stock ponds remaining on the Santa Rosa Plain of Sonoma County, California. There are 8 known breeding sites within the Santa Rosa Plain. The life history of the California tiger salamander can be characterized as a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily terrestrial. Adult California tiger salamanders mate in vernal pools and similar water bodies, and the females lay their eggs in the water. Females attach their eggs singly or, in rare circumstances, in groups of two to four, to twigs, grass stems, vegetation, or debris. In ponds with little or no vegetation, females may attach eggs to objects, such as rocks and boards on the bottom. After breeding, adults leave the pool and return to small mammal burrows in surrounding uplands, although they may continue to come out nightly for approximately the next two weeks to feed. In drought years, the seasonal pools may not form and the adults may not breed. The eggs hatch in 10 to 14 days with newly hatched salamanders (larvae) ranging in size from 11.5 to 14.2 mm (0.5 to 0.6 in) in total length. The larval stage of the California tiger salamander usually lasts three to six months, because most seasonal ponds and pools dry up during the summer, although some larvae in Contra Costa and Alameda Counties may remain in their breeding sites over the summer. Amphibian larvae must grow to a critical minimum body size before they can metamorphose (change into a different physical form) to the terrestrial stage. One study found larvae metamorphosed and left the breeding pools 60 to 94 days after the eggs had been laid, with larvae developing faster in smaller, more rapidly drying pools. The longer the inundation period, the larger the larvae and metamorphosed juveniles are able to grow, and the more likely they are to survive and reproduce. The larvae perish if a site dries before they complete metamorphosis. There was a strong positive correlation between inundation period and total number of metamorphosing juvenile amphibians, including tiger salamanders. Size at metamorphosis is positively correlated with stored body fat and survival of juvenile amphibians, and negatively correlated with age at first reproduction.

Exposure to cyanide at the CCC could result in a substantial reduction in egg hatching and survival to metamorphosis. Larval salamanders that do survive could experience reduced growth rates and reduced swimming abilities which would increase their vulnerability to a host of potential stressors including, temperature, flow, predation, and inter- and intraspecific competition for food and cover. Reduced growth rates could also delay reproductive maturity and productivity. Because amphibian larvae must grow to a critical minimum body size before they can metamorphose to the terrestrial stage, reduced growth rates of the California tiger salamander could preclude emergence prior to pond drying, resulting in larval mortalities.

We anticipate that the estimated reductions in hatched eggs and larval survival would reduce the California tiger salamander's fertility rates substantially. The salamander's fertility rates would be reduced based upon (a) reductions in numbers of eggs hatched, (b) reductions in larvae surviving through the first year and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time between mating.

Reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the CCC. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). In the section of Population Level Effects, we noted that reduction in fecundity and juvenile survival would have a greater population-level effect on species that have fewer opportunities to reproduce, like California tiger salamanders. Lifetime reproductive success for California and other tiger salamanders is low. One study found the average female bred 1.4 times and produced 8.5 young that survived to metamorphosis per reproductive effort. This resulted in roughly 11 metamorphic offspring over the lifetime of a female. Most California tiger salamanders in this study did not reach sexual maturity until four or five years old. While individuals may survive for more than 10 years, many breed only once, and one study estimated that less than five percent of metamorphic juveniles survive to become breeding adults. The mechanisms for recruitment are clearly dependent on a number of factors such as migration, terrestrial survival, and population turnover, whose interaction is not well understood.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the California tiger salamander's reproduction by reducing the number of eggs laid by females, the hatchability of laid eggs, and a reduction of survivorship of larval salamanders. Salamanders may also experience effects on growth, locomotion, condition, and development. The California tiger salamander has a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily terrestrial. Although exposure to cyanide concentrations at the chronic criterion may be isolated to the aquatic phases of the salamander's life cycle (i.e. breeding activities and egg and larval development), the long-term effects of exposure could result in reduced adult growth, survival and fitness. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected California tiger salamander population's decline could stabilize at a reduced absolute population number or could continue to decline until it's extirpated. For extant populations supported by a single breeding pond, reproductive failure at that site could result in the extirpation of the population and even those populations with multiple breeding sites may have difficulty recolonizing after local extirpations due to physiological, morphological, and behavioral constraints. Based upon the magnitude of effect we anticipate could occur, we conclude that ultimately, California tiger salamanders are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the California Tiger salamander.

SONORA TIGER SALAMANDER

Ambystoma tigrinum stebbinsi

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. Based on multiple lines of evidence, including the relative sensitivity of amphibians to other pollutants, their relative sensitivity to rainbow trout and the sensitivity of rainbow trout to cyanide we

conclude that Sonora tiger salamanders exposed to cyanide at the CCC could experience substantial reductions in recruitment due to reductions in fecundity, hatching success, and larval development and survival. Compared to control populations, we estimate rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than 52% (Appendix E). We estimate that rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than 61% (Appendix E). Based on the relative sensitivity of amphibians as compared to rainbow trout we estimate the effects of cyanide at the CCC for amphibians could be of similar or greater magnitude than effects estimated for rainbow trout. As such, Sonora tiger salamanders exposed to cyanide at the CCC could experience a substantial reduced egg hatching and decreased survival to metamorphosis of larval salamanders.

Although no comparable data are available for amphibians, cyanide may also adversely effect growth, locomotion, condition, and development as described above for fish in the Effects Overview. The effects of chronic toxicity could also be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

This species probably once inhabited springs, streams, backwaters, and cienegas that held permanent or nearly permanent water sources in the San Rafael Valley, Arizona, and Sonora, Mexico. Cattle ponds or tanks are now the primary habitat for Sonora tiger salamanders. Terrestrial salamanders likely spend much of the year in rodent burrows, rotted logs, and other moist cover sites. The life history of the Sonora tiger salamander can be characterized as a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that may be fully aquatic or primarily terrestrial. Sonora tiger salamanders begin their life as jelly-coated eggs laid in water. They hatch and grow as aquatic larvae with gills, and then either mature as gilled aquatic adults called branchiate adults, neotenes, or paedomorphs, or metamorphose into terrestrial salamanders without gills. Sonora tiger salamanders begin breeding as early as January, and eggs can be found in ponds as late as early May. Breeding after monsoon rains in July and August is rare. Courtship takes place under water, and after fertilization, female tiger salamanders lay 200 to 2000 eggs attaching them to aquatic vegetation, sticks, rocks, or substrate individually or in clumps of up to 50. Eggs take from 2-4 weeks to hatch; the colder the water, the longer the eggs take to develop. Following hatching, Sonora tiger salamander larvae can develop to the minimum size necessary to metamorphose in as little as two months. However, because many sites with Sonora salamanders hold water all year, larvae often remain in the water longer before metamorphosing, or develop into branchiate adults instead of metamorphosing. The proportion of larvae that metamorphose depends heavily on pond permanence. In ponds that dry, all larvae that are large enough metamorphose. In ponds that do not dry, approximately 17 percent of larvae that are large enough metamorphose (Collins *et al.* 1988).

Exposure to cyanide at the CCC could result in a substantial reduction in egg hatching and survival to metamorphosis. Larval salamanders that do survive could experience reduced growth rates and reduced swimming abilities which would increase their

vulnerability to a host of potential stressors including, temperature, flow, predation, and inter- and intraspecific competition for food and cover. Reduced growth rates could also delay reproductive maturity and productivity. Because amphibian larvae must grow to a critical minimum body size before they can metamorphose to the terrestrial stage, reduced growth rates of the Sonora tiger salamander could preclude emergence prior to pond drying, resulting in larval mortalities.

We anticipate that the estimated reductions in hatched eggs and larval survival would reduce the Sonora salamander's fertility rates substantially. The salamander's fertility rates would be reduced based upon (a) reductions in numbers of eggs hatched, (b) reductions in larvae surviving through the first year and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rate delay maturity or lengthen the time between mating.

In permanent water bodies, approximately 83% of the Sonora salamander larvae develop into branchiate adults instead of metamorphosing. In these cases, individuals could experience increased exposure to cyanide at the CCC due to their continued use of aquatic habitats throughout all stages of their life cycle. Whereas metamorphosed terrestrial adults may experience reduced adult growth, survival and fitness due to the long-term effects of cyanide exposure during egg and larval development, fully aquatic adults will be at greater risk of such adverse and compounded effects due to continued exposure to cyanide at CCC as adults.

Reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the CCC. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). Metamorphs are the only life history stage that can disperse from pond to pond and establish new populations. Data suggest that only a small proportion of salamanders in a pond are likely to have dispersed from another pond, so salamanders in each pond are referred to as a population. For those populations consisting primarily of branchiate adults, habitat destruction associated with cyanide at CCC levels may adversely effect Sonora tiger salamander reproduction and survival to the extent that the population supported by that breeding pond becomes extirpated. Due to the low occurrence or dispersal amongst terrestrial adults, habitat destruction associated with cyanide at CCC levels may also result in population extirpation.

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Sonora tiger salamander's reproduction by reducing the number of eggs laid by females, the hatchability of laid eggs, and a reduction of survivorship of larval salamanders. Salamanders may also experience effects on growth, locomotion, condition, and development. The Sonora tiger salamander has a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are either wholly aquatic or primarily terrestrial. Although exposure to cyanide concentrations at the chronic criterion may be isolated to the aquatic phases of the salamander's life cycle (i.e.

breeding activities and egg and larval development), the long-term effects of exposure could result in reduced adult growth, survival and fitness. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Sonora tiger salamander population's decline could stabilize at a reduced absolute population number or could continue to decline until it's extirpated. For extant populations supported by a single breeding pond, reproductive failure at that site could result in the extirpation of the population and even those populations with multiple breeding sites may have difficulty recolonizing after local extirpations due to physiological, morphological, and behavioral constraints. Based upon the magnitude of effect we anticipate could occur, we conclude that ultimately, Sonora tiger salamanders are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Sonora tiger salamander.

SANTA CRUZ LONG-TOED SALAMANDER

Ambystoma macrodactylum croceum

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. Based on multiple lines of evidence, including the relative sensitivity of amphibians to other pollutants, their relative sensitivity to rainbow trout and the sensitivity of rainbow trout to cyanide we conclude that Santa Cruz long-toed salamanders exposed to cyanide at the CCC could experience substantial reductions in recruitment due to reductions in fecundity, hatching success, and larval development and survival. Compared to control populations, we estimate rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than 52% (Appendix E). We estimate that rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than 61% (Appendix E). Based on the relative sensitivity of amphibians as compared to rainbow trout we estimate the effects of cyanide at the CCC for amphibians could be of similar or greater magnitude than effects estimated for rainbow trout. As such, Santa Cruz long-toed salamanders exposed to cyanide at the CCC could experience a substantial reduced egg hatching and decreased survival to metamorphosis of larval salamanders.

Although no comparable data are available for amphibians, cyanide may also adversely effect growth, locomotion, condition, and development as described above for fish in the Effects Overview. The effects of chronic toxicity could also be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

The life history of the Santa Cruz long-toed salamander can be characterized as a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that may be fully aquatic or primarily terrestrial. Santa Cruz long-toed salamanders spend most of their lives underground in small mammal burrows and along the root systems of

plants in upland chaparral and woodland areas of coast live oak (*Quercus agrifolia*) or Monterey pine (*Pinus radiata*) as well as riparian strips of arroyo willows (*Salix lasiolepis*) and other species. Ideal breeding locations appear to be shallow, temporary, freshwater ponds that lack fishes and hold water at least through the spring months. Eggs are laid singly on submerged stalks of spike rush (*Eleocharis* spp.) or other vegetation about two to three centimeters apart. Free floating, unattached, and clustered eggs have also been observed. Each female lays about 300 (range 215 to 411) eggs per year. Eggs usually hatch in 15 to 30 days. The larvae remain in the pond environment for 90 to 145 days. Larvae metamorphose when they reach a minimum size of about 32 mm snout to vent length. Metamorphosis can be accelerated by adverse pond conditions, such as reduction in food resources, water pollution, increased temperatures, and drying of the pond environment. Metamorphosed salamanders leave the pond. These salamanders become sexually mature in 3-4 years and do not return to the pond except to breed.

As noted above, exposure to cyanide at the CCC could result in a substantial reduction in egg hatching and survival to metamorphosis. Larval salamanders that do survive could experience reduced growth rates and reduced swimming abilities which would increase their vulnerability to a host of potential stressors including, temperature, flow, predation, and inter- and intraspecific competition for food and cover. Reduced growth rates could also delay reproductive maturity and productivity. Because amphibian larvae must grow to a critical minimum body size before they can metamorphose to the terrestrial stage, reduced growth rates of the Santa Cruz long-toed salamander could preclude emergence prior to pond drying, resulting in larval mortalities.

We anticipate that the estimated reductions in hatched eggs and larval survival would reduce the Santa Cruz long-toed salamander's fertility rates substantially. The salamander's fertility rates would be reduced based upon (a) reductions in numbers of eggs hatched, (b) reductions in larvae surviving through the first year and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time between mating.

Reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the CCC. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation). Santa Cruz long-toed salamanders are known from 3 metapopulations, each comprised of one or more subpopulations, located in Santa Cruz and Monterey Counties. Habitat destruction associated with cyanide at CCC levels may result in a reduction of available breeding sites and the loss of subpopulations that could lead to a reduction in population size and range and an increased vulnerability to catastrophic events that may adversely affect breeding and survival. Pollution, siltation, and the degradation of water quality in breeding ponds resulting from nearby development and agriculture was cited as one of the primary threats to the Santa Cruz long-toed salamander at the time of listing and is continues to be a threat to species survival

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Santa Cruz long-toed salamander's reproduction by reducing the number of eggs laid by females, the hatchability of laid eggs, and a reduction of survivorship of larval salamanders. Salamanders may also experience effects on growth, locomotion, condition, and development. The Santa Cruz long-toed salamander has a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily terrestrial. Although exposure to cyanide concentrations at the chronic criterion may be isolated to the aquatic phases of the salamander's life cycle (i.e. breeding activities and egg and larval development), the long-term effects of exposure could result in reduced adult growth, survival and fitness. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Santa Cruz long-toed salamander population's decline could stabilize at a reduced absolute population number or could continue to decline until it's extirpated. For extant populations supported by a single breeding pond, reproductive failure at that site could result in the extirpation of the population and even those populations with multiple breeding sites may have difficulty recolonizing after local extirpations due to physiological, morphological, and behavioral constraints. Based upon the magnitude of effect we anticipate could occur, we conclude that ultimately, Santa Cruz long-toed salamanders are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Santa Cruz long-toed salamander.

Plethodontidae

SAN MARCOS SALAMANDER

Eurycea nana

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. Based on multiple lines of evidence, including the relative sensitivity of amphibians to other pollutants, their relative sensitivity to rainbow trout and the sensitivity of rainbow trout to cyanide we conclude that San Marcos salamanders exposed to cyanide at the CCC could experience substantial reductions in recruitment due to reductions in fecundity, hatching success, and larval development and survival. Compared to control populations, we estimate rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than 52% (Appendix E). We estimate that rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than 61% (Appendix E). Based on the relative sensitivity of amphibians as compared to rainbow trout we estimate the effects of cyanide at the CCC for amphibians could be of similar or greater magnitude than effects estimated for rainbow trout. As such, San Marcos salamanders exposed to cyanide at the CCC could experience a substantial reduced egg hatching and decreased survival to metamorphosis of larval salamanders.

Formal Draft Biological Opinion.

Although no comparable data are available for amphibians, cyanide may also adversely effect growth, locomotion, condition, and development as described above for fish in the Effects Overview. The effects of chronic toxicity could also be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

The San Marcos salamander is aquatic throughout its life cycle. Information on the reproduction of these salamanders is limited. It is found in rocky spring openings and rocky areas downstream from the dams at Spring Lake in San Marcos, TX, as well as in shallow spring areas on the northernmost portion of Spring Lake on a limestone shelf. Six essential elements are required for the San Marcos salamander: thermally constant water, flowing water, clean and clear water, sand, gravel, and rock substrates with little mud or detritus, vegetation for cover, and finally an adequate food supply. Males and females are sexually mature at 19 to 23.5 and 21 mm, respectively. No eggs have been found in nature, but the presence of gravid females and small larvae throughout the year suggests year round breeding. Artificial habitat studies have indicated that the average clutch size is 20, and that eggs are laid in standing pools with thick vegetation. Larvae emerge from the jelly covered eggs after 24 days.

The recovery plan lists a number of major threats to the San Marcos salamander, including decreases in water quantity and quality (including dissolved ions, trace elements, pH, nutrients, dissolved oxygen, and organic contaminants). As noted above, exposure to cyanide at the CCC could result in a substantial reduction in egg hatching and larval survival. Larval salamanders that do survive could experience reduced growth rates and reduced swimming abilities which would increase their vulnerability to a host of potential stressors including, temperature, flow, predation, and inter- and intraspecific competition for food and cover. Reduced growth rates could also delay reproductive maturity and productivity. Because the San Marcos salamander is aquatic throughout its entire life cycle, adults could also be at risk of increased adverse and compounded affects due to continued exposure to cyanide at CCC.

Little is known about the salamander's reproduction or longevity in the wild. We anticipate that the estimated reductions in hatched eggs and larval survival would reduce the San Marcos Salamander's fertility rates substantially. The salamander's fertility rates would be reduced based upon (a) reductions in numbers of eggs hatched, (b) reductions in larvae surviving through the first year and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rated delay maturity or lengthen the time between mating.

The San Marcos Salamander consists of a single population that is found only in Spring Lake and downstream in the San Marcos River below Spring Lake for 150 m. The San Marcos salamander population is estimated to exceed 53,000 individuals. Reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the CCC. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured

in terms of changes in population growth rates and changes in risk of extinction (or extirpation).

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the San Marcos salamander's reproduction by reducing the number of eggs laid by females, the hatchability of laid eggs, and a reduction of survivorship of larval salamanders. Salamanders may also experience effects on growth, locomotion, condition, and development. Although exposure to cyanide concentrations at the chronic criterion may be isolated to the aquatic phases of the salamander's life cycle (i.e. breeding activities and egg and larval development), the long-term effects of exposure could result in reduced adult growth, survival and fitness. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected San Marcos salamander population's decline could stabilize at a reduced absolute population number or could continue to decline until it's extirpated. For extant populations supported by a single breeding pond, reproductive failure at that site could result in the extirpation of the population and even those populations with multiple breeding sites may have difficulty recolonizing after local extirpations due to physiological, morphological, and behavioral constraints. Based upon the magnitude of effect we anticipate could occur, we conclude that ultimately, San Marcos salamanders are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the San Marcos salamander.

Critical Habitat: Critical habitat has been established for the San Marcos salamander in Hays County, Texas. It includes Spring Lake and its outflow and the San Marcos River downstream for 50 meters from the Spring Lake Dam. Primary constituent elements were not identified in the final rule designating critical habitat, but in order for these aquatic areas to provide the necessary conditions for successful breeding and survival they must include water of sufficient quality to allow the species to carry out normal feeding, movement, sheltering, and reproductive behaviors and to experience individual and population growth. Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentrations. This approval could adversely affect the quality of water to the degree that it would impair individual reproduction and survival of San Marcos salamanders and cause salamanders to experience adverse effects to growth, locomotion, condition, and development. Based on data for the rainbow trout, our surrogate species for assessing impacts to amphibians, we estimate the reduction in number of hatched eggs could be as much as, but is not likely to be greater than 52% and the reduction in larval survival to metamorphosis could be as much as, but is not likely to be greater than 61%. These effects are estimated to of a magnitude great enough to reduce numbers of San Marcos salamanders and result in the extirpation of local populations. Approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the species.

BARTON SPRINGS SALAMANDER

Eurycea sosorum

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. Based on multiple lines of evidence, including the relative sensitivity of amphibians to other pollutants, their relative sensitivity to rainbow trout and the sensitivity of rainbow trout to cyanide we conclude that Barton Springs salamanders exposed to cyanide at the CCC could experience substantial reductions in recruitment due to reductions in fecundity, hatching success, and larval development and survival. Compared to control populations, we estimate rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than 52% (Appendix E). We estimate that rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than 61% (Appendix E). Based on the relative sensitivity of amphibians as compared to rainbow trout we estimate the effects of cyanide at the CCC for amphibians could be of similar or greater magnitude than effects estimated for rainbow trout. As such, Barton Springs salamanders exposed to cyanide at the CCC could experience a substantial reduced egg hatching and decreased survival to metamorphosis of larval salamanders.

Although no comparable data are available for amphibians, cyanide may also adversely effect growth, locomotion, condition, and development as described above for fish in the Effects Overview. The effects of chronic toxicity could also be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

The Barton Springs salamander has been found only at the four spring outlets that make up Barton Springs within the City of Austin's Zilker Park in Travis County, Texas. The salamander requires stable aquatic environmental conditions, including perennially flowing spring water which is clear, clean, near neutral, varies very little in temperature (annual average must be 21 to 22 degrees Celsius) and proper flowing conditions to maintain dissolved oxygen content. The salamander also prefers clean, loose gravel substrates. The Barton Springs salamanders retain their larval gills throughout their lives, becoming mature and reproducing underwater. Known longevity for Barton Springs salamanders in captivity is at least 10 Years. Gravid females, eggs, and larvae are typically found throughout the year in the Barton Springs, which indicates that the salamander can reproduce year-round. Captive salamanders indicate that females are sexually mature at 11 to 17 months. During courtship the male deposits a spermatophore, which becomes attached to a plant, rock, or other substrate. Females can store the spermatophore in a specialized portion of the cloaca, known as the spermatheca for a month or longer. Females of some salamander species may store spermatophores for up to 2.5 years before ovulation and fertilization occur. In most salamanders, fertilization is internal and occurs during egg-laying whereby sperm are released onto eggs as they pass through the female's cloaca. Clutch sizes range from 5 to 39 eggs with an average of 22

eggs. Hatching of eggs in captivity occurred within 16 to 39 days after the eggs were laid, and the first three months following hatching were a critical period for juvenile survival.

Both the listing notice and the recovery plan for the Barton Springs salamander cite diminished water quality as a critical threat to the species. Analysis has shown that the water quality at Barton Springs has decreased. Dissolved oxygen has decreased (16 percent over 25 years) and conductivity (which has shown levels nearing the rate at which 100% mortality can be expected in 24 hours), sulfates, turbidity, nitrate-nitrogen, and total organic carbon have all increased. While data is limited concerning the Barton Springs salamander's vulnerability to contaminants, its semipermeable skin and reproductive processes suggest that it may be similar to other amphibians. As noted above, exposure to cyanide at the CCC could result in a substantial reduction in egg hatching and larval survival. Larval salamanders that do survive could experience reduced growth rates and reduced swimming abilities which would increase their vulnerability to a host of potential stressors including, temperature, flow, predation, and inter- and intraspecific competition for food and cover. Reduced growth rates could also delay reproductive maturity and productivity. Because the Barton Springs salamander is aquatic throughout its entire life cycle, adults could also be at risk of increased adverse and compounded affects due to continued exposure to cyanide at CCC.

Little is known about the salamander's reproduction or longevity in the wild. We anticipate that the estimated reductions in hatched eggs and larval survival would reduce the Barton Springs Salamander's fertility rates substantially. The salamander's fertility rates would be reduced based upon (a) reductions in numbers of eggs hatched, (b) reductions in larvae surviving through the first year and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time between mating.

The Barton Springs Salamander consists of a single population that is found only within Barton Springs in Austin, TX. Total population estimates for the entire species are difficult because of the challenge posed by population surveys. Previous surveys would indicate a number in the hundreds at most, especially as some surveys have found no salamanders even in years with sufficient water flow. Reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the CCC. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation).

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Barton Springs salamander's reproduction by reducing the number of eggs laid by females, the hatchability of laid eggs, and a reduction of survivorship of larval salamanders. Salamanders may also experience effects on growth, locomotion, condition, and development. Although exposure to cyanide concentrations at

the chronic criterion may be isolated to the aquatic phases of the salamander's life cycle (i.e. breeding activities and egg and larval development), the long-term effects of exposure could result in reduced adult growth, survival and fitness. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Barton Springs salamander population's decline could stabilize at a reduced absolute population number or could continue to decline until it's extirpated. For extant populations supported by a single breeding pond, reproductive failure at that site could result in the extirpation of the population and even those populations with multiple breeding sites may have difficulty recolonizing after local extirpations due to physiological, morphological, and behavioral constraints. Based upon the magnitude of effect we anticipate could occur, we conclude that ultimately, Barton Springs salamanders are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Barton Springs salamander.

TEXAS BLIND SALAMANDER

Typhlomolge rathbuni

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. Based on multiple lines of evidence, including the relative sensitivity of amphibians to other pollutants, their relative sensitivity to rainbow trout and the sensitivity of rainbow trout to cyanide we conclude that Texas blind salamanders exposed to cyanide at the CCC could experience substantial reductions in recruitment due to reductions in fecundity, hatching success, and larval development and survival. Compared to control populations, we estimate rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than 52% (Appendix E). We estimate that rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than 61% (Appendix E). Based on the relative sensitivity of amphibians as compared to rainbow trout we estimate the effects of cyanide at the CCC for amphibians could be of similar or greater magnitude than effects estimated for rainbow trout. As such, Texas blind salamanders exposed to cyanide at the CCC could experience a substantial reduced egg hatching and decreased survival to metamorphosis of larval salamanders.

Although no comparable data are available for amphibians, cyanide may also adversely effect growth, locomotion, condition, and development as described above for fish in the Effects Overview. The effects of chronic toxicity could also be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

The Texas blind salamander lives only underground in the Edwards Aquifer in Hays County, Texas. It is neotenic (non-transforming) and aquatic throughout its life. It lives in water filled caverns in the aquifer, and is well adapted to the environment, but is believed to be sensitive to temperature changes, as the aquifer has a near constant

temperature of 21 degrees Celsius. It is likely that the Texas blind salamander is sexually active year round, which results from the very little seasonal change in the conditions of the aquifer. Gravid females have been found each month of the year. This species reproduced in captivity at the Cincinnati Zoo. In two months, three spawning events occurred. Clutch sizes ranged from 8 to 21 eggs. The unpigmented eggs were attached in ones, twos, and threes to pieces of gravel. Temperatures of close to 21 degrees Celsius are required for proper egg development.

As noted above, exposure to cyanide at the CCC could result in a substantial reduction in egg hatching and larval survival. Larval salamanders that do survive could experience reduced growth rates and reduced swimming abilities which would increase their vulnerability to a host of potential stressors including, temperature, flow, predation, and inter- and intraspecific competition for food and cover. Reduced growth rates could also delay reproductive maturity and productivity. Because the Texas blind salamander is aquatic throughout its entire life cycle, adults could also be at risk of increased adverse and compounded effects due to continued exposure to cyanide at CCC.

The Texas blind salamander has been found only in Hays County, Texas and the total distribution of the species may be as small as 10 square kilometers. Population estimates have not been established. Little is known about the salamander's reproduction or longevity in the wild. We anticipate that the estimated reductions in hatched eggs and larval survival would reduce the Texas blind salamander's fertility rates substantially. The salamander's fertility rates would be reduced based upon (a) reductions in numbers of eggs hatched, (b) reductions in larvae surviving through the first year and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time between mating.

Reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the CCC. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation).

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Texas blind salamander's reproduction by reducing the number of eggs laid by females, the hatchability of laid eggs, and a reduction of survivorship of larval salamanders. Salamanders may also experience effects on growth, locomotion, condition, and development. Although exposure to cyanide concentrations at the chronic criterion may be isolated to the aquatic phases of the salamander's life cycle (i.e. breeding activities and egg and larval development), the long-term effects of exposure could result in reduced adult growth, survival and fitness. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An affected Texas blind salamander population's decline could stabilize at a reduced absolute population number or could continue to decline until it's extirpated. For extant populations supported by a single breeding pond, reproductive

failure at that site could result in the extirpation of the population and even those populations with multiple breeding sites may have difficulty recolonizing after local extirpations due to physiological, morphological, and behavioral constraints. Based upon the magnitude of effect we anticipate could occur, we conclude that ultimately, Texas blind salamanders are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Texas blind salamander.

Bufonidae

WYOMING TOAD

Bufo baxteri

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. Based on multiple lines of evidence, including the relative sensitivity of amphibians to other pollutants, their relative sensitivity to rainbow trout and the sensitivity of rainbow trout to cyanide we conclude that Wyoming toads exposed to cyanide at the CCC could experience substantial reductions in recruitment due to reductions in fecundity, hatching success, and larval development and survival. Compared to control populations, we estimate rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than 52% (Appendix E). We estimate that rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than 61% (Appendix E). Based on the relative sensitivity of amphibians as compared to rainbow trout we estimate the effects of cyanide at the CCC for amphibians could be of similar or greater magnitude than effects estimated for rainbow trout. As such, Wyoming toads exposed to cyanide at the CCC could experience a substantial reduced egg hatching and decreased survival to metamorphosis of tadpoles.

Although no comparable data are available for amphibians, cyanide may also adversely effect growth, locomotion, condition, and development as described above for fish in the Effects Overview. The effects of chronic toxicity could also be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

The life history of the Wyoming can be characterized as a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily terrestrial. Wyoming toads inhabit floodplains, ponds, and the margins of small seepage lakes in the shortgrass communities of the Laramie Basin. The toad is known to exist in an area between 10 and 20 miles west of Laramie, Wyoming. Sightings since 1987 have come only from a 2 square mile area around Mortenson Lake and its associated meadows. Breeding occurs in shallow water typically less than six inches deep. Vegetated margins and bays of lakes, ponds, and irrigated meadows are preferred breeding areas. Breeding sites are often dry by late summer. Adult toads appear at breeding sites in May after

daytime temperatures reach 70 degrees Fahrenheit. Males appear first and attract females with their calls. Breeding congregations are not large, usually consisting of half a dozen males and a few females gathering at a pond or lake margin. Breeding takes place from mid-May to mid-June depending upon weather conditions in any given year. Eggs are deposited in gelatinous strings containing 2,000 to 5,000 eggs and strands are often intertwined among vegetation. Eggs hatched in less than 1 week in water temperatures ranging between 77 degrees Fahrenheit during the day and 50 degrees at night. Tadpoles transformed into toadlets by 4 to 6 weeks following egg deposition.

As noted above, exposure to cyanide at the CCC could result in a substantial reduction in egg hatching and survival to metamorphosis. Tadpoles that do survive could experience reduced growth rates and reduced swimming abilities which would increase their vulnerability to a host of potential stressors including, temperature, flow, predation, and inter- and intraspecific competition for food and cover. Because amphibian larvae must grow to a critical minimum body size before they can metamorphose to the terrestrial stage, reduced growth rates of the Wyoming toad could delay reproductive maturity and productivity.

No long-term studies have been performed to determine the maximum longevity of Wyoming toads in the wild. Corn (1993a) observed that few adult toads lived >2 yr at Mortenson Lake, but that was in a population afflicted with chytrid fungus. In captivity, one female toad with an estimated birth date in 1989 lived in captivity from 1994 until its death in 1997 (Callaway, 1998) and produced large numbers of healthy young from 1994 - 96. Little else is known about the toad's reproduction or longevity in the wild. We anticipate that the estimated reductions in hatched eggs and larval survival would reduce the Wyoming toad's fertility rates substantially. The toad's fertility rates would be reduced based upon (a) reductions in numbers of eggs hatched, (b) reductions in larvae surviving through the first year and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time between mating.

Reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the CCC. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation).

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Wyoming toad's reproduction by reducing the number of eggs laid by females, the hatchability of laid eggs, and a reduction of survivorship of tadpoles. Toads may also experience effects on growth, locomotion, condition, and development. The Wyoming toad has a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily terrestrial. Although exposure to cyanide concentrations at the chronic criterion may be isolated to the aquatic phases of the toad's life cycle (i.e. breeding activities and egg and larval development), the long-term effects

of exposure could result in reduced adult growth, survival and fitness. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected Wyoming toad population's decline could stabilize at a reduced absolute population number or could continue to decline until it's extirpated. For extant populations supported by a single breeding pond, reproductive failure at that site could result in the extirpation of the population and even those populations with multiple breeding sites may have difficulty recolonizing after local extirpations due to physiological, morphological, and behavioral constraints. Based upon the magnitude of effect we anticipate could occur, we conclude that ultimately, Wyoming toads are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Wyoming toad.

ARROYO TOAD

Bufo californicus

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. Based on multiple lines of evidence, including the relative sensitivity of amphibians to other pollutants, their relative sensitivity to rainbow trout and the sensitivity of rainbow trout to cyanide we conclude that arroyo toads exposed to cyanide at the CCC could experience substantial reductions in recruitment due to reductions in fecundity, hatching success, and larval development and survival. Compared to control populations, we estimate rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than 52% (Appendix E). We estimate that rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than 61% (Appendix E). Based on the relative sensitivity of amphibians as compared to rainbow trout we estimate the effects of cyanide at the CCC for amphibians could be of similar or greater magnitude than effects estimated for rainbow trout. As such, arroyo toads exposed to cyanide at the CCC could experience a substantial reduced egg hatching and decreased survival to metamorphosis of tadpoles.

Although no comparable data are available for amphibians, cyanide may also adversely effect growth, locomotion, condition, and development as described above for fish in the Effects Overview. The effects of chronic toxicity could also be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

The arroyo toad occurs principally along coastal drainages, but it has been recorded at several locations on the desert slopes of the of the Transverse and Peninsular Mountain ranges south of the Santa Clara River, Los Angeles County, California. The life history of the Wyoming toad can be characterized as a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily terrestrial. The arroyo toad requires shallow, slow-moving stream habitats, and riparian habitats that are disturbed naturally on a regular basis, primarily by flooding. In the northern portion of their range,

arroyo toads are found in foothill canyons and intermountain valleys where medium- to large-sized rivers are bordered closely by low hills, riverbed gradients are low, and the surface stream flows frequently pool or are intermittent for at least a few months of the year. In southern California (central portion of the arroyo toad's range), they also occur on the coastal plain and on a few desert slopes. For breeding, adult arroyo toads use open sites such as overflow pools, and old flood channels which are less than 30 cm deep with clear water. Breeding sites usually have flow rates less than 5 cm per second. The breeding period lasts from late January or February to early July, although weather can extend the period. If conditions are unsuitable, females may not obtain sufficient resources for egg production and will forgo breeding during that year. Anywhere from 2000-10,000 eggs are laid in two parallel gelatinous strings on substrates of sand, gravel, cobble, or mud generally located away from vegetation in the shallow margins of the pool. Embryos usually hatch in 4 to 6 days at water temperatures of 12 to 16 degrees Celsius (54 to 59 degrees Fahrenheit). The larval period for arroyo toads lasts about 65 to 85 days, depending on water Temperatures. Newly metamorphosed juveniles remain on sparsely vegetated sand and gravel bars bordering the natal pool for 3–5 wk (Sweet, 1992). Male arroyo toads can reach sexual maturity in 1 year, if conditions are favorable, but females require 2 or 3 years. Mark–recapture studies suggest that few arroyo toads survive into their fifth year, and that these are predominantly females (Sweet, 1993). In the absence of American bullfrogs, adult arroyo toads have a high survivorship during the active season, but suffer 55–80% mortality as they overwinter (Sweet, 1993).

As noted above, exposure to cyanide at the CCC could result in a substantial reduction in egg hatching and survival to metamorphosis. Tadpoles that do survive could experience reduced growth rates and reduced swimming abilities which would increase their vulnerability to a host of potential stressors including, temperature, flow, predation, and inter- and intraspecific competition for food and cover. Because amphibian larvae must grow to a critical minimum body size before they can metamorphose to the terrestrial stage, reduced growth rates of the arroyo toad could delay reproductive maturity and productivity.

We anticipate that the estimated reductions in hatched eggs and larval survival would reduce the arroyo toad's fertility rates substantially. The toad's fertility rates would be reduced based upon (a) reductions in numbers of eggs hatched, (b) reductions in larvae surviving through the first year and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time between mating.

Reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the CCC. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation).

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the arroyo toad's reproduction by reducing the number of eggs laid by females, the hatchability of laid eggs, and a reduction of survivorship of tadpoles. Toads may also experience effects on growth, locomotion, condition, and development. The arroyo toad has a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily terrestrial. Although exposure to cyanide concentrations at the chronic criterion may be isolated to the aquatic phases of the toad's life cycle (i.e. breeding activities and egg and larval development), the long-term effects of exposure could result in reduced adult growth, survival and fitness. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An effected arroyo toad population's decline could stabilize at a reduced absolute population number or could continue to decline until it's extirpated. For extant populations supported by a single breeding pond, reproductive failure at that site could result in the extirpation of the population and even those populations with multiple breeding sites may have difficulty recolonizing after local extirpations due to physiological, morphological, and behavioral constraints. Based upon the magnitude of effect we anticipate could occur, we conclude that ultimately, arroyo toads are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the arroyo toad.

Critical Habitat: Critical habitat consists of 11,695 acres in five counties in California: Riverside, San Bernardino, Los Angeles, Ventura, Santa Barbara. The physical and biological features of critical habitat essential to the conservation of the arroyo toad include rivers or streams with hydrologic regimes that supply water to provide space, food, and cover needed to sustain eggs, tadpoles, metamorphosing juveniles, and adult breeding toads. In order for these aquatic areas to provide the necessary conditions for successful breeding and survival they must include water of sufficient quality to allow the species to carry out normal feeding, movement, sheltering, and reproductive behaviors and to experience individual and population growth. Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentrations. This approval could adversely affect the quality of water to the degree that it would impair individual reproduction and survival of arroyo toads and cause toads to experience adverse effects to growth, locomotion, condition, and development. Based on data for the rainbow trout, our surrogate species for assessing impacts to amphibians, we estimate the reduction in number of hatched eggs could be as much as, but is not likely to be greater than 52% and the reduction in larval survival to metamorphosis could be as much as, but is not likely to be greater than 61%. These effects are estimated to of a magnitude great enough to reduce numbers of arroyo toads and result in the extirpation of local populations. Approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the species.

HOUSTON TOAD

Bufo houstonensis

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. Based on multiple lines of evidence, including the relative sensitivity of amphibians to other pollutants, their relative sensitivity to rainbow trout and the sensitivity of rainbow trout to cyanide we conclude that Houston toads exposed to cyanide at the CCC could experience substantial reductions in recruitment due to reductions in fecundity, hatching success, and larval development and survival. Compared to control populations, we estimate rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than 52% (Appendix E). We estimate that rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than 61% (Appendix E). Based on the relative sensitivity of amphibians as compared to rainbow trout we estimate the effects of cyanide at the CCC for amphibians could be of similar or greater magnitude than effects estimated for rainbow trout. As such, Houston toads exposed to cyanide at the CCC could experience a substantial reduced egg hatching and decreased survival to metamorphosis of tadpoles.

Although no comparable data are available for amphibians, cyanide may also adversely effect growth, locomotion, condition, and development as described above for fish in the Effects Overview. The effects of chronic toxicity could also be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Houston toads are associated with forest ecosystems and sandy soils. The life history of the Houston toad can be characterized as a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily terrestrial. Adult Houston toads respond to cold or summer heat by burrowing into moist sand or hiding under rocks, leaf litter, logs, or in abandoned animal burrows. Breeding occurs in ephemeral, rain-fed pools, flooded fields, and permanent ponds from late January to June with a peak from February to March. Reported egg-laying dates range from February 18 through June 26 and clutch sizes range from 512 to 6,199. Depending on environmental conditions, eggs may hatch within a week and tadpoles develop into toadlets within 40-80 days. Mortality rates are high, and only 1% of eggs laid are believed to survive to adulthood. Captive raised males are sexually mature at 1 year and females at 1 to 2 years.

As noted above, exposure to cyanide at the CCC could result in a substantial reduction in egg hatching and survival to metamorphosis. Tadpoles that do survive could experience reduced growth rates and reduced swimming abilities which would increase their vulnerability to a host of potential stressors including, temperature, flow, predation, and inter- and intraspecific competition for food and cover. Because amphibian larvae must grow to a critical minimum body size before they can metamorphose to the terrestrial stage, reduced growth rates of the Houston toad could delay reproductive maturity and productivity. For tadpoles developing in ephemeral ponds, reduced growth rates resulting in a prolonged time to metamorphosis could preclude full development prior to pond drying, resulting in larval mortalities.

We anticipate that the estimated reductions in hatched eggs and larval survival would reduce the Houston toad's fertility rates substantially. The toad's fertility rates would be reduced based upon (a) reductions in numbers of eggs hatched, (b) reductions in larvae surviving through the first year and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rate delay maturity or lengthen the time between mating.

Reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the CCC. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation).

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Houston toad's reproduction by reducing the number of eggs laid by females, the hatchability of laid eggs, and a reduction of survivorship of tadpoles. Toads may also experience effects on growth, locomotion, condition, and development. The Houston toad has a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily terrestrial. Although exposure to cyanide concentrations at the chronic criterion may be isolated to the aquatic phases of the toad's life cycle (i.e. breeding activities and egg and larval development), the long-term effects of exposure could result in reduced adult growth, survival and fitness. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An affected Houston toad population's decline could stabilize at a reduced absolute population number or could continue to decline until it's extirpated. For extant populations supported by a single breeding pond, reproductive failure at that site could result in the extirpation of the population and even those populations with multiple breeding sites may have difficulty recolonizing after local extirpations due to physiological, morphological, and behavioral constraints. Based upon the magnitude of effect we anticipate could occur, we conclude that ultimately, Houston toads are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Houston toad.

Critical Habitat: Critical habitat has been established in Bastrop, Burleson and Harris County, Texas. Although not described when critical habitat was designated, physical and biological features essential to the conservation of the Houston toad include seasonally-flooded breeding ponds, deep sandy soils, and forest or woodlands. In order for these aquatic areas to provide the necessary conditions for successful breeding and survival they must include water of sufficient quality to allow the species to carry out normal feeding, movement, sheltering, and reproductive behaviors and to experience individual and population growth. Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentrations. This approval could adversely affect the quality of water to the degree

that it would impair individual reproduction and survival of Houston toads and cause toads to experience adverse effects to growth, locomotion, condition, and development. Based on data for the rainbow trout, our surrogate species for assessing impacts to amphibians, we estimate the reduction in number of hatched eggs could be as much as, but is not likely to be greater than 52% and the reduction in larval survival to metamorphosis could be as much as, but is not likely to be greater than 61%. These effects are estimated to be of a magnitude great enough to reduce numbers of Houston toads and result in the extirpation of local populations. Approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the species.

Eleutherodactylidae

GUAJON

Eleutherodactylus cooki

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. Based on multiple lines of evidence, including the relative sensitivity of amphibians to other pollutants, their relative sensitivity to rainbow trout and the sensitivity of rainbow trout to cyanide we conclude that guajons exposed to cyanide at the CCC could experience substantial reductions in recruitment due to reductions in fecundity, hatching success, and larval development and survival. Compared to control populations, we estimate rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than 52% (Appendix E). We estimate that rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than 61% (Appendix E). Based on the relative sensitivity of amphibians as compared to rainbow trout we estimate the effects of cyanide at the CCC for amphibians could be of similar or greater magnitude than effects estimated for rainbow trout. As such, guajons exposed to cyanide at the CCC could experience a substantial reduced egg hatching and decreased survival to metamorphosis of tadpoles.

Although no comparable data are available for amphibians, cyanide may also adversely effect growth, locomotion, condition, and development as described above for fish in the Effects Overview. The effects of chronic toxicity could also be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

The guajon is associated with the granitic rocks found in the Cuchilla de Panduras mountain range in southeastern Puerto Rico where it inhabits caves formed by large boulders of granite rock known as “guajonales,” but can also be found in associated streams with patches of rock without caves systems. In streams, the guajón has been found only in patches of rock in the streambed. The streams can be perennial, or ephemeral formed

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during heavy rain and are surrounded by secondary forest. Rocks in the streambed form crevices and grottoes. The guajón deposits eggs on humid boulders within grottoes and on flat surfaces. Eggs are guarded by males. The mean clutch size of the guajón is 17.35 eggs, the developmental time of eggs is 20 to 29 days, and parental care contributes to hatching success. Hatching success of this species is 85 percent, with hatchlings remaining together as a group in the nest for several days before dispersing.

As noted above, exposure to cyanide at the CCC could result in a substantial reduction in egg hatching and survival to metamorphosis. Tadpoles that do survive could experience reduced growth rates and reduced swimming abilities which would increase their vulnerability to a host of potential stressors including, temperature, flow, predation, and inter- and intraspecific competition for food and cover. Because amphibian larvae must grow to a critical minimum body size before they can metamorphose to the terrestrial stage, reduced growth rates of the guajón could delay reproductive maturity and productivity. For tadpoles developing in ephemeral ponds, reduced growth rates resulting in a prolonged time to metamorphosis could preclude full development prior to pond drying, resulting in larval mortalities.

Little else is known about the frog's reproduction or longevity in the wild. We anticipate that the estimated reductions in hatched eggs and larval survival would reduce the guajón's fertility rates substantially. The frog's fertility rates would be reduced based upon (a) reductions in numbers of eggs hatched, (b) reductions in larvae surviving through the first year and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time between mating.

Reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the CCC. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation).

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the guajón's reproduction by reducing the number of eggs laid by females, the hatchability of laid eggs, and a reduction of survivorship of tadpoles. Frogs may also experience effects on growth, locomotion, condition, and development. The guajón has a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily terrestrial. Although exposure to cyanide concentrations at the chronic criterion may be isolated to the aquatic phases of the frog's life cycle (i.e. breeding activities and egg and larval development), the long-term effects of exposure could result in reduced adult growth, survival and fitness. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An affected guajón population's decline could stabilize at a reduced absolute population number or could continue to decline until it's extirpated. For extant populations supported by a single breeding pond, reproductive failure at that site could

result in the extirpation of the population and even those populations with multiple breeding sites may have difficulty recolonizing after local extirpations due to physiological, morphological, and behavioral constraints. Based upon the magnitude of effect we anticipate could occur, we conclude that ultimately, guajons are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the guajon.

Critical Habitat: Critical habitat units were established for Humacao, Las Piedras, Maunabo, Patillas, and Yabucoa, Puerto Rico. The physical and biological features of critical habitat essential to the conservation of the guajon includes plutonic, granitic, or sedimentary rocks/boulders that form caves, crevices, and grottoes (interstitial spaces) in a streambed, and that are in proximity, or connected, to a permanent, ephemeral, or subterranean clear-water stream or water source. The interstitial spaces between or underneath rocks provide microenvironments characterized by generally higher humidity and cooler temperatures than outside the rock formations. In order for these aquatic areas to provide the necessary conditions for successful breeding and survival they must include water of sufficient quality to allow the species to carry out normal feeding, movement, sheltering, and reproductive behaviors and to experience individual and population growth. Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentrations. This approval could adversely affect the quality of water to the degree that it would impair individual reproduction and survival of guajon and cause these frogs to experience adverse effects to growth, locomotion, condition, and development. Based on data for the rainbow trout, our surrogate species for assessing impacts to amphibians, we estimate the reduction in number of hatched eggs could be as much as, but is not likely to be greater than 52% and the reduction in larval survival to metamorphosis could be as much as, but is not likely to be greater than 61%. These effects are estimated to of a magnitude great enough to reduce numbers of California red-legged frogs and result in the extirpation of local populations. Approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the species.

Ranidae

CALIFORNIA RED-LEGGED FROG

Rana aurora draytonii

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. Based on multiple lines of evidence, including the relative sensitivity of amphibians to other pollutants, their relative sensitivity to rainbow trout and the sensitivity of rainbow trout to cyanide we conclude that California red-legged frogs exposed to cyanide at the CCC could experience substantial reductions in recruitment due to reductions in fecundity, hatching success, and larval development and survival. Compared to control populations, we

estimate rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than 52% (Appendix E). We estimate that rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than 61% (Appendix E). Based on the relative sensitivity of amphibians as compared to rainbow trout we estimate the effects of cyanide at the CCC for amphibians could be of similar or greater magnitude than effects estimated for rainbow trout. As such, California red-legged frogs exposed to cyanide at the CCC could experience a substantial reduced egg hatching and decreased survival to metamorphosis of tadpoles.

Although no comparable data are available for amphibians, cyanide may also adversely effect growth, locomotion, condition, and development as described above for fish in the Effects Overview. The effects of chronic toxicity could also be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

California red-legged frogs have been documented in 46 counties in California, but now remain in only 238 streams or drainages in 31 counties. The life history of the California red-legged frog can be characterized as a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily aquatic but use a variety of aquatic, riparian, and upland habitats. The California red-legged frog's larvae, tadpoles, and metamorphs can be found in streams, deep pools, backwaters, creeks, ponds, marshes, sag ponds, dune ponds, and lagoons. Breeding adults are commonly associated with water deeper than 0.7 m (2 feet) which is slow moving and choked by shrubby riparian or emergent vegetation. California red-legged frogs breed from November to April. Males appear at breeding sites 2-4 weeks before females. Once a pair of frogs has moved into the breeding position, they move to where the eggs are laid and fertilized. The 2,000 to 5,000 eggs float near the surface, attached to emergent vegetation, roots, or twigs. Eggs hatch within 6 to 14 days depending on water temperatures and require approximately 20 days to develop into tadpoles. Tadpoles in turn require anywhere between 11 to 20 weeks to develop into terrestrial frogs. At some locations, larvae may overwinter before metamorphosing. Water bodies suitable for tadpole rearing must remain watered at least until the tadpoles metamorphose into adults, typically between July and September. Adult California red-legged frogs can survive in moist upland areas after breeding habitat has dried, and can live several years to make new breeding attempts. Therefore, aquatic breeding habitat need not be available every year, but it must be available often enough and for appropriate hydroperiods to maintain a California red-legged frog population during most years.

As noted above, exposure to cyanide at the CCC could result in a substantial reduction in egg hatching and survival to metamorphosis. Tadpoles that do survive could experience reduced growth rates and reduced swimming abilities which would increase their vulnerability to a host of potential stressors including, temperature, flow, predation, and inter- and intraspecific competition for food and cover. Because amphibian larvae must grow to a critical minimum body size before they can metamorphose to the terrestrial stage, reduced growth rates of the California red-legged frog could delay reproductive

maturity and productivity. For tadpoles developing in ephemeral ponds, reduced growth rates resulting in a prolonged time to metamorphosis could preclude full development prior to pond drying, resulting in larval mortalities.

We anticipate that the estimated reductions in hatched eggs and larval survival would reduce the California red-legged frog's fertility rates substantially. The frog's fertility rates would be reduced based upon (a) reductions in numbers of eggs hatched, (b) reductions in larvae surviving through the first year and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time between mating.

Reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the CCC. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation).

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the California red-legged frog's reproduction by reducing the number of eggs laid by females, the hatchability of laid eggs, and a reduction of survivorship of tadpoles. Frogs may also experience effects on growth, locomotion, condition, and development. The California red-legged frog has a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily terrestrial. Although exposure to cyanide concentrations at the chronic criterion may be isolated to the aquatic phases of the frog's life cycle (i.e. breeding activities and egg and larval development), the long-term effects of exposure could result in reduced adult growth, survival and fitness. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An affected California red-legged frog population's decline could stabilize at a reduced absolute population number or could continue to decline until it's extirpated. For extant populations supported by a single breeding pond, reproductive failure at that site could result in the extirpation of the population and even those populations with multiple breeding sites may have difficulty recolonizing after local extirpations due to physiological, morphological, and behavioral constraints. Based upon the magnitude of effect we anticipate could occur, we conclude that ultimately, California red-legged frogs are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the California red-legged frog.

Critical Habitat: California red-legged frog critical habitat has been established in 34 locations in Alameda, Butte, Contra Costa, El Dorado, Kern, Los Angeles, Marin, Merced, Monterey, Napa, Nevada, San Benito, San Luis Obispo, San Mateo, Santa Barbara, Santa Clara, Santa Cruz, Solano, Ventura and Yuba Counties, California, and includes aquatic breeding and non-breeding habitats that provide the physical and biological features of critical habitat essential to the conservation of the California red-

legged frog. Aquatic breeding habitat is essential for providing space, food, and cover necessary to sustain the early life history stages of larval and juvenile California red-legged frogs. It consists of low-gradient fresh water bodies, including natural and manmade (e.g., stock) ponds, backwaters within streams and creeks, marshes, lagoons, and dune ponds. It does not include deep lacustrine water habitat (e.g., deep lakes and reservoirs 50 ac (20 ha) or larger in size). To be considered essential breeding habitat, the aquatic feature must have the capability to hold water for a minimum of 20 weeks in all but the driest of years.

Nonbreeding aquatic habitat consists of those aquatic elements identified above, and also includes, but is not limited to, other wetland habitats such as intermittent creeks, seeps, and springs. California red-legged frogs can use large cracks in the bottom of dried ponds as refugia to maintain moisture and avoid heat and solar exposure. Without these non-breeding aquatic features, California red-legged frogs would not be able to survive drought periods, or be able to disperse to other breeding habitat.

In order for these aquatic areas to provide the necessary conditions for successful breeding and survival they must include water of sufficient quality to allow the species to carry out normal feeding, movement, sheltering, and reproductive behaviors and to experience individual and population growth. Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentrations. This approval could adversely affect the quality of water to the degree that it would impair individual reproduction and survival of California red-legged frogs and cause frogs to experience adverse effects to growth, locomotion, condition, and development. Based on data for the rainbow trout, our surrogate species for assessing impacts to amphibians, we estimate the reduction in number of hatched eggs could be as much as, but is not likely to be greater than 52% and the reduction in larval survival to metamorphosis could be as much as, but is not likely to be greater than 61%. These effects are estimated to of a magnitude great enough to reduce numbers of California red-legged frogs and result in the extirpation of local populations. Approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the species.

CHIRICAHUA LEOPARD FROG

Rana chiricahuensis

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. Based on multiple lines of evidence, including the relative sensitivity of amphibians to other pollutants, their relative sensitivity to rainbow trout and the sensitivity of rainbow trout to cyanide we conclude that Chiricahua leopard frogs exposed to cyanide at the CCC could experience substantial reductions in recruitment due to reductions in fecundity, hatching success, and larval development and survival. Compared to control populations, we estimate rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in the

number of hatched eggs and that reduction could be as much as, but is not likely to be greater than 52% (Appendix E). We estimate that rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than 61% (Appendix E). Based on the relative sensitivity of amphibians as compared to rainbow trout we estimate the effects of cyanide at the CCC for amphibians could be of similar or greater magnitude than effects estimated for rainbow trout. As such, Chiricahua leopard frogs exposed to cyanide at the CCC could experience a substantial reduced egg hatching and decreased survival to metamorphosis of tadpoles.

Although no comparable data are available for amphibians, cyanide may also adversely effect growth, locomotion, condition, and development as described above for fish in the Effects Overview. The effects of chronic toxicity could also be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

The range of the Chiricahua leopard frog is split into two disjunct parts - northern populations along the Mogollon Rim in Arizona east into the mountains of west-central New Mexico, and southern populations in southeastern Arizona, southwestern New Mexico, and Mexico. The life history of the Chiricahua leopard frog can be characterized as a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily aquatic. It is an inhabitant of montane and river valley cienegas, springs, pools, cattle tanks, lakes, reservoirs, streams, and rivers. It is a habitat generalist that historically was found in a variety of aquatic habitat types, but is now limited to the comparatively few aquatic systems that support few or no non-native predators (e.g. American bullfrogs, fishes, and crayfishes). The species also requires permanent or semi-permanent pools for breeding, water characterized by low levels of contaminants and moderate pH, and may be excluded or exhibit periodic die-offs where a pathogenic chytridiomycete fungus is present. Egg masses of Chiricahua leopard frogs have been reported in all months except January, November, and December, but reports of oviposition in June are uncommon. Hatching time of egg masses in the wild has not been studied in detail. Eggs of the Ramsey Canyon leopard frog hatch in approximately 14 days depending on temperature, and hatching time may be as short as eight days in geothermally influenced springs. Tadpoles metamorphose in three to nine months (Jennings 1988, 1990), and may overwinter.

As noted above, exposure to cyanide at the CCC could result in a substantial reduction in egg hatching and survival to metamorphosis. Tadpoles that do survive could experience reduced growth rates and reduced swimming abilities which would increase their vulnerability to a host of potential stressors including, temperature, flow, predation, and inter- and intraspecific competition for food and cover. Because amphibian larvae must grow to a critical minimum body size before they can metamorphose to the terrestrial stage, reduced growth rates of the Chiricahua leopard frog could delay reproductive maturity and productivity. For tadpoles developing in ephemeral ponds, reduced growth rates resulting in a prolonged time to metamorphosis could preclude full development prior to pond drying, resulting in larval mortalities.

Little is known about age and size at reproductive maturity or the longevity of the Chiricahua leopard frog. We anticipate that the estimated reductions in hatched eggs and larval survival would reduce the Chiricahua leopard frog's fertility rates substantially. The frog's fertility rates would be reduced based upon (a) reductions in numbers of eggs hatched, (b) reductions in larvae surviving through the first year and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time between mating.

Reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the CCC. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation).

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the Chiricahua leopard frog's reproduction by reducing the number of eggs laid by females, the hatchability of laid eggs, and a reduction of survivorship of tadpoles. Frogs may also experience effects on growth, locomotion, condition, and development. The Chiricahua leopard frog has a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily aquatic. Although exposure to cyanide concentrations at the chronic criterion may be isolated to the aquatic phases of the frog's life cycle (i.e. breeding activities and egg and larval development), the long-term effects of exposure could result in reduced adult growth, survival and fitness. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An affected Chiricahua leopard frog population's decline could stabilize at a reduced absolute population number or could continue to decline until it's extirpated. For extant populations supported by a single breeding pond, reproductive failure at that site could result in the extirpation of the population and even those populations with multiple breeding sites may have difficulty recolonizing after local extirpations due to physiological, morphological, and behavioral constraints. Based upon the magnitude of effect we anticipate could occur, we conclude that ultimately, Chiricahua leopard frogs are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the Chiricahua leopard frog.

MOUNTAIN YELLOW-LEGGED FROG

Rana muscosa

Southern California Population

Relatively few studies were available for estimating the magnitude of effects that could occur following exposure to cyanide at criterion concentrations. Based on multiple lines of evidence, including the relative sensitivity of amphibians to other pollutants, their relative sensitivity to rainbow trout and the sensitivity of rainbow trout to cyanide we conclude that mountain yellow-legged frogs exposed to cyanide at the CCC could

experience substantial reductions in recruitment due to reductions in fecundity, hatching success, and larval development and survival. Compared to control populations, we estimate rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in the number of hatched eggs and that reduction could be as much as, but is not likely to be greater than 52% (Appendix E). We estimate that rainbow trout exposed to cyanide at the CCC could experience a substantial reduction in survival of young fish through the first year and that reduction could be as much as, but is not likely to be greater than 61% (Appendix E). Based on the relative sensitivity of amphibians as compared to rainbow trout we estimate the effects of cyanide at the CCC for amphibians could be of similar or greater magnitude than effects estimated for rainbow trout. As such, mountain yellow-legged frogs exposed to cyanide at the CCC could experience a substantial reduced egg hatching and decreased survival to metamorphosis of tadpoles.

Although no comparable data are available for amphibians, cyanide may also adversely effect growth, locomotion, condition, and development as described above for fish in the Effects Overview. The effects of chronic toxicity could also be influenced by other stressors such as dissolved oxygen concentration, temperature, and the presence of other pollutants in the water column.

Currently the mountain yellow-legged frog is known from only seven locations in southern California in portions of the San Gabriel, San Bernardino, and San Jacinto Mountains. The life history of the mountain yellow-legged frog can be characterized as a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily aquatic. Water depth, persistence, and configuration (*i.e.* gently sloping shorelines and margins) appear to be important for mountain yellow-legged frogs, allowing for shelter from predators along shores or in deeper waters, and habitat for breeding, foraging, egg-laying, thermoregulation (to regulate the body temperature through behavior), and overwintering. Breeding activity typically begins in April at lower elevations, to June or July at upper elevations and continues for approximately a month. Egg masses vary in size from as few as 15 eggs to 350 eggs per mass, which is considered low, relative to a range of several hundred to several thousand for other true frogs. Egg masses are normally deposited in shallow waters where they may be attached to rocks, gravel, vegetation, or similar substrates. As larvae develop, they tend to gravitate towards warmer waters to elevate body temperatures which may facilitate larval and metamorphic development by allowing for a higher metabolic rate. Even with this behavior, larvae apparently must overwinter at least two times for 6 to 9 month intervals before attaining metamorphosis because the active season is short and the aquatic habitat maintains warm temperatures for only brief intervals. Time to develop from fertilization to metamorphosis appears to be variable, ranging up to 3.5 years, with reproductive maturity reached from 3 to 4 years following metamorphosis. Little is known about adult longevity, but the species is presumed to be long-lived due to adult survivorship.

As noted above, exposure to cyanide at the CCC could result in a substantial reduction in egg hatching and survival to metamorphosis. Tadpoles that do survive could experience reduced growth rates and reduced swimming abilities which would increase their vulnerability to a host of potential stressors including, temperature, flow, predation, and inter- and intraspecific competition for food and cover. Because amphibian larvae must

grow to a critical minimum body size before they can metamorphose to the terrestrial stage, reduced growth rates of the mountain yellow-legged frog could delay reproductive maturity and productivity.

We anticipate that the estimated reductions in hatched eggs and larval survival would reduce the mountain yellow-legged frog's fertility rates substantially. The frog's fertility rates would be reduced based upon (a) reductions in numbers of eggs hatched, (b) reductions in larvae surviving through the first year and (c) the additive effect of reduced fecundity and survival. The combined effect could be less than additive if the eggs that hatch in the presence of cyanide represent individuals that are less sensitive. These reductions in reproduction could be exacerbated further if reduced growth rates delay maturity or lengthen the time between mating.

Reductions in reproductive performance and survival represent reductions in the fitness of the individuals exposed to cyanide at the CCC. Changes in the fitness of the individuals in a population will affect the population as a whole. Effects to populations can be measured in terms of changes in population growth rates and changes in risk of extinction (or extirpation).

In summary, exposure to cyanide concentrations at the chronic criterion could substantially reduce the mountain yellow-legged frog's reproduction by reducing the number of eggs laid by females, the hatchability of laid eggs, and a reduction of survivorship of tadpoles. Frogs may also experience effects on growth, locomotion, condition, and development. The mountain yellow-legged frog has a complex life cycle, consisting of eggs and larvae that are entirely aquatic and adults that are primarily terrestrial. Although exposure to cyanide concentrations at the chronic criterion may be isolated to the aquatic phases of the frog's life cycle (i.e. breeding activities and egg and larval development), the long-term effects of exposure could result in reduced adult growth, survival and fitness. Because of the high magnitude of effects, we would expect density-dependent compensatory mechanisms, if they exist, to be overwhelmed. An affected mountain yellow-legged frog population's decline could stabilize at a reduced absolute population number or could continue to decline until it's extirpated. For extant populations supported by a single breeding pond, reproductive failure at that site could result in the extirpation of the population and even those populations with multiple breeding sites may have difficulty recolonizing after local extirpations due to physiological, morphological, and behavioral constraints. Based upon the magnitude of effect we anticipate could occur, we conclude that ultimately, mountain yellow-legged frogs are likely to become extirpated from waters where they are exposed to cyanide toxicity at the chronic criterion concentration. Continued approval of the chronic criterion could reduce the reproduction, numbers, and distribution of the mountain yellow-legged frog.

Critical Habitat: The physical and biological features of critical habitat essential to the conservation of the mountain yellow-legged frog include (1) Water source(s) found between 1,214 to 7,546 feet (370 to 2,300 meter) in elevation that are permanent. Water sources include, but are not limited to, streams, rivers, perennial creeks (or permanent plunge pools within intermittent creeks), pools (i.e., a body of impounded water that is

contained above a natural dam) and other forms of aquatic habitat. Aquatic habitats that are used by mountain yellow-legged frog for breeding purposes must maintain water during the entire tadpole growth phase, which can last for up to 2 years. During periods of drought, or less than average rainfall, these breeding sites may not hold water long enough for individuals to complete metamorphosis, but they would still be considered essential breeding habitat in wetter years. Further, the aquatic includes: a.) Bank and pool substrates consisting of varying percentages of soil or silt, sand, gravel cobble, rock, and boulders; b.) Open gravel banks and rocks projecting above or just beneath the surface of the water for sunning posts; c.) Aquatic refugia, including pools with bank overhangs, downfall logs or branches, and/or rocks to provide cover from predators; and d.) Streams or stream reaches between known occupied sites that can function as corridors for movement between aquatic habitats used as breeding and/or foraging

In order for these aquatic areas to provide the necessary conditions for successful breeding and survival they must include water of sufficient quality to allow the species to carry out normal feeding, movement, sheltering, and reproductive behaviors and to experience individual and population growth. Approval of the CCC in State and Tribal water quality standards would authorize States and Tribes to continue to manage cyanide in waters to the criteria concentrations. This approval could adversely affect the quality of water to the degree that it would impair individual reproduction and survival of California red-legged frogs and cause frogs to experience adverse effects to growth, locomotion, condition, and development. Based on data for the rainbow trout, our surrogate species for assessing impacts to amphibians, we estimate the reduction in number of hatched eggs could be as much as, but is not likely to be greater than 52% and the reduction in larval survival to metamorphosis could be as much as, but is not likely to be greater than 61%. These effects are estimated to be of a magnitude great enough to reduce numbers of California red-legged frogs and result in the extirpation of local populations. Approval of the CCC would adversely affect the quality of water to the degree that normal population growth is likely to be impacted and could result in extirpation from critical habitat containing cyanide at the CCC. Impacts to water quality resulting from management of cyanide to the CCC would diminish the ability of critical habitat to provide for conservation of the species.

8.0 CUMULATIVE EFFECTS

Cumulative effects include the effects of future State, tribal, local or private actions that are reasonably certain to occur in the action area considered in this biological opinion. Future Federal actions that are unrelated to the proposed action are not considered in this section because they require separate consultation pursuant to section 7 of the ESA.

As noted in Section 5.0 above, the action area for this consultation consists of all waters of the United States, including territorial seas, which extend seaward a distance of three miles from the coast (CWA section 502), where federally listed endangered, threatened, and proposed species reside. The action area includes such waters within and surrounding Indian country, the 50 States, and all U.S. territories. Given the size of the action area, it is not practical to specifically evaluate cumulative effects in this biological opinion.

In general, the threatened and endangered aquatic species and designated critical habitats considered in this biological opinion are likely to be adversely affected by non-federal activities that affect the quantity, quality, and hydrographic patterns of water, waterways, and habitats important to these species and critical habitats. These activities could include changes in land and water use and management patterns in ways that increase erosion and sedimentation, increase introduction of pollutants into waterways, and result in introductions and spread of non-native invasive species that directly or indirectly affect listed species and critical habitats. These species and their critical habitats could also be affected by illegal harvest. States or private entities may also engage in activities to restore, enhance, and improve water quality and quantity and restore more natural hydrographic patterns that benefit listed species and their habitats. All of the species and critical habitats considered in this document are likely to be subject to these types of activities in the future to varying extents. The final listing and designation rules, recovery plans, and 5-year reviews for these species and critical habitats are good sources of information, in part, on the threats and benefits associated with these types of activities. These documents are cited in Appendix A.

9.0 CONCLUSION

After reviewing the current status of the following listed species, the environmental baseline for the action area, the effects of EPA's continuing approval of state water quality standards that rely on their nationally recommended criteria for cyanide, and cumulative effects, it is the Service's biological opinion that the action, as proposed, is likely to jeopardize the continued existence of the following species:

Gulf sturgeon, Kootenai River white sturgeon, Pallid sturgeon, Alabama sturgeon, Ozark cavefish, Alabama cavefish, Waccamaw silverside, Modoc sucker, Santa Ana sucker, Warner sucker, Shortnose sucker, Cui-ui, June sucker, Lost River sucker, Razorback sucker, Pygmy sculpin, Blue shiner, Beautiful shiner, Devils River minnow, Spotfin chub, Slender chub, Mojave tui chub, Owens tui chub, Borax Lake chub, Humpback chub, Sonora chub, Bonytail chub, Gila chub, Yaqui chub, Pahrnagat roundtail chub, Virgin River Chub, Rio Grande silvery minnow, Big Spring spinedace, Little Colorado spinedace, Spikedace, Moapa dace, Palezone shiner, Cahaba shiner, Arkansas River shiner, Cape Fear shiner, Pecos bluntnose shiner, Topeka shiner, Oregon chub, Blackside dace, Woundfin, Colorado pikeminnow (=squawfish), Ash Meadows speckled dace, Kendall Warm Springs dace, Loach minnow, Unarmored threespine stickleback, Tidewater goby, White River springfish, Hiko White River springfish, Railroad Valley springfish, Delta smelt, Slackwater darter, Vermilion darter, Relict darter, Etowah darter, Fountain darter, Niangua darter, Watercress darter, Okaloosa darter, Duskytail darter, Bayou darter, Cherokee darter, Maryland darter, Bluemask darter, Boulder darter, Amber darter, Goldline darter, Conasauga logperch, Leopard darter, Roanoke logperch, Snail darter, Big Bend gambusia, San Marcos gambusia, Clear Creek gambusia, Pecos gambusia, Gila topminnow, Bull trout, Little Kern Golden trout, Apache trout, Lahontan Cutthroat trout, Paiute Cutthroat trout, Greenback Cutthroat Mountain trout, Gila trout, Atlantic salmon, Illinois cave amphipod,

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Noel's amphipod, Cumberland elktoe, Dwarf wedgemussel, Appalachian elktoe, Fat three-ridge, Ouachita rock pocketbook, Birdwing pearlymussel, Fanshell, Dromedary pearlymussel, Chipola slabshell, Tar River spiny mussel, Purple bankclimber, Cumberlandian combshell, Oyster mussel, Curtis pearlymussel, Yellow blossom, Tan riffleshell, Upland combshell, Catspaw, White catspaw, Southern acornshell, Southern combshell, Green blossom, Northern riffleshell, Tubercled blossom, Turgid blossom, Shiny pigtoe, Finerayed pigtoe, Cracking pearlymussel, Pink mucket, Fine-lined pocketbook, Higgins eye, Orangenacre mucket, Arkansas fatmucket, Speckled pocketbook, Shinyrayed pocketbook, Alabama lampmussel, Carolina heelsplitter, Scaleshell mussel, Louisiana pearlshell, Alabama moccasinshell, Coosa moccasinshell, Gulf moccasinshell, Ochlockonee moccasinshell, Ring pink, Littlewing pearlymussel, White wartyback pearlymussel, Orangefoot pimpleback, Clubshell, James spiny mussel, Black clubshell, Southern clubshell, Dark pigtoe, Southern pigtoe, Cumberland pigtoe, Flat pigtoe, Ovate clubshell, Rough pigtoe, Oval pigtoe, Heavy pigtoe, Fat pocketbook, Alabama heelsplitter, Triangular kidneyshell, Rough rabbitsfoot, Winged mapleleaf, Cumberland monkeyface, Appalachian monkeyface, Stirrupshell, Pale Lilliput, Purple bean, Cumberland bean, reticulated flatwoods salamander, frosted flatwoods salamander, California tiger salamander (central California DPS), California tiger salamander (Santa Barbara County DPS), California tiger salamander (Sonoma DPS), Santa Cruz long-toed salamander, Sonora Tiger salamander, San Marcos salamander, Barton Springs salamander, Texas blind salamander, Wyoming toad, arroyo toad, Houston toad, guajon, California red-legged frog, Chiricahua leopard frog, mountain yellow-legged frog

Exposure of the above listed fish species to cyanide at the proposed chronic criterion concentration is likely to substantially reduce their reproduction by reducing the number of eggs spawned by females, reducing the hatchability of spawned eggs, and by reducing the survivorship of young fish in their first year. These fish may also experience effects on growth, swimming performance, condition, and development. In addition, Fountain darters, Bull trout, Apache trout, and Lahontan cutthroat trout exposed to cyanide at the acute criterion are likely to experience substantial reductions in survival. Based upon the magnitude of adverse effects caused by the exposure of these listed species to cyanide at the proposed acute and/or chronic criteria concentrations, these fish species are likely to become extirpated from waters where they are exposed to cyanide toxicity at the CMC and/or CCC. Continued approval of the acute and/or chronic criteria at the rangewide scale of these listed species is likely to reduce their reproduction, numbers, and distribution.

Exposure of the Illinois cave amphipod and Noel's amphipod to cyanide at the chronic criterion concentration is likely to result in the loss of individuals, especially in situations when these amphipods are subject to interspecific resource competition or predation. Because both amphipod species exist in populations that are geographically isolated from one another, the ability of amphipods to recolonize perturbed habitats is limited. Thus, cyanide exposure may result in the elimination of a population unit that cannot rebound. The loss of a population unit for either amphipod species would substantially reduce the reproduction, numbers, or distribution of these species.

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For the above mentioned mussel species, exposure of their host fish to cyanide at criterion concentrations is likely to reduce the abundance of fish hosts for glochidia, thereby decreasing the likelihood that glochidia will survive because they will be unable to attach to suitable host. Since attachment of glochidia to a suitable host is a rare and necessary event in the mussel reproductive cycle, reductions in host fish abundance are likely to negatively impact mussel reproductive output and ultimately population numbers. Thus, host fish abundance for these species is anticipated to substantially decline to levels that will reduce reproduction, numbers, or distribution of these mussel species.

After reviewing the current status of critical habitat, the environmental baseline for the action area, the effects of EPA's continuing approval of state water quality standards that rely on their nationally recommended criteria for cyanide, and cumulative effects, it is the Service's biological opinion that the action, as proposed, is likely to result in the destruction or adverse modification of critical habitat that has been designated for the following species:

Gulf sturgeon, Kootenai River white sturgeon, Alabama sturgeon, Alabama cavefish, Waccamaw silverside, Modoc sucker, Santa Ana sucker, Warner sucker, June sucker, Razorback sucker, Beautiful shiner, Devils River minnow, Spotfin chub, Slender chub, Owens tui chub, Borax Lake chub, Humpback chub, Sonora chub, Bonytail chub, Gila chub, Yaqui chub, Virgin River Chub, Rio Grande silvery minnow, Big Spring spinedace, Little Colorado spinedace, Spikedace, Arkansas River shiner, Cape Fear shiner, Pecos bluntnose shiner, Topeka shiner, Woundfin, Colorado pikeminnow (=squawfish), Loach minnow, Tidewater goby, White River springfish, Hiko White River springfish, Railroad Valley springfish, Delta smelt, Slackwater darter, Fountain darter, Niangua darter, Maryland darter, Amber darter, Conasauga logperch, Leopard darter, Snail darter, San Marcos gambusia, Bull trout, Little Kern Golden trout, Cumberland elktoe, Appalachian elktoe, Fat three-ridge, Chipola slabshell, Purple bankclimber, Cumberlandian combshell, Oyster mussel, Upland combshell, Southern acornshell, Fine-lined pocketbook, Orangenacre mucket, Shinyrayed pocketbook, Carolina heelsplitter, Alabama moccasinshell, Coosa moccasinshell, Gulf moccasinshell, Ochlockonee moccasinshell, Southern clubshell, Dark pigtoe, Southern pigtoe, Ovate clubshell, Oval pigtoe, Triangular kidneyshell, Rough rabbitsfoot, Purple bean, reticulated flatwoods salamander, frosted flatwoods salamander, California tiger salamander (central California DPS), California tiger salamander (Santa Barbara County DPS), San Marcos salamander, arroyo toad, Houston toad, guajon, California red-legged frog, mountain yellow-legged frog

The physical and biological features of critical habitat essential to the conservation of these listed species include water of sufficient quality for species to carry out normal feeding, movement, sheltering, and reproductive behaviors, and to experience individual and population growth. Approval of the CCC in state water quality standards would authorize states to manage cyanide in waters to these levels. This approval and cyanide in waters to these levels is likely to adversely affect the quality of water to the degree that it would impair individual reproduction and survival of these listed fish species as well as fish species that are hosts for the above mentioned listed mussels. Approval of the CCC

and cyanide in waters to these levels is likely to adversely affect the quality of water to the degree that it would impair normal population growth and likely cause the extirpation of these listed fish from their critical habitat containing cyanide at the CCC. In addition, the majority of fish hosts identified for the above mentioned mussel species are likely to exhibit population declines at cyanide criteria concentrations. For these reasons, impacts to water quality resulting from cyanide in waters to the level of the CCC would diminish the intended conservation function of critical habitat for these listed fishes and mussels.

10.0 REASONABLE AND PRUDENT ALTERNATIVES

The regulations (50 CFR 402.02) implementing section 7 of the ESA define reasonable and prudent alternatives (RPAs) as alternative actions, identified during formal consultation, that: (1) can be implemented in a manner consistent with the intended purpose of the action; (2) can be implemented consistent with the scope of the action agency’s legal authority and jurisdiction; (3) are economically and technologically feasible; and (4) would, the Service believes, avoid the likelihood of jeopardizing the continued existence of listed species or the destruction or adverse modification of critical habitat.

The Service has developed the following RPAs to the EPA’s proposed action:

1. By December 1, 2012, EPA shall, subject to Service approval, review the geographic ranges of the listed species and designated critical habitats addressed in this biological opinion and insure that the water bodies or water body segments within those ranges include: a) a designated use for which aquatic life criteria apply; b) aquatic life cyanide criteria at least as stringent as described below; and, c) appropriate language within any EPA-approved general policies requiring State coordination with local Service field offices on implementation of general policies. The Service recommends the EPA consider the following criteria for water bodies and water body segments:

Fish: The acute and chronic cyanide criteria for protection of listed fish species are shown in Table 21. These values are based on the acute and chronic Assessment Effects Concentrations (EC_A) which represent the *the highest* concentration of cyanide where the effects on listed species are expected to be insignificant (see Appendix B for details).

Table 21. Freshwater acute and chronic cyanide criteria for the protection of listed fish species (NC, no change).

Listed Species		Order/Family	Surrogate Taxa	Recommended Acute Criteria [Acute EC _A (ug CN/L)]	Recommended Chronic Criteria [Chronic EC _A (ug CN/L)]
Gulf sturgeon	<i>Acipenser oxyrinchus desotoi</i>	Acipenseriformes Acipenseridae (sturgeon)	Actinopterygii (class)	NC	2.86
Kootenai River white sturgeon	<i>Acipenser transmontanus</i>				
Pallid sturgeon	<i>Scaphirhynchus albus</i>				

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Alabama sturgeon	<i>Scaphirhynchus suttkusi</i>								
Waccamaw silverside	<i>Menidia extensa</i>	Atheriniformes Atherinopsidae							
Modoc sucker	<i>Catostomus micorps</i>	Cypriniformes Catosdomidae (suckers)	Cypriniformes (order)	NC	3.64				
Santa Anna sucker	<i>Catostomus santaanae</i>								
Warner sucker	<i>Catostomus warnerensis</i>								
Shortnose sucker	<i>Chasmistes brevirostris</i>								
Cui ui	<i>Chasmistes cujus</i>								
June sucker	<i>Chasmistes liorus</i>								
Lost River sucker	<i>Deltistes luxatus</i>								
Razorback sucker	<i>Xyrauchen texanus</i>								
Spotfin chub	<i>Cyprinella monacha</i>						<i>Xyrauchen texanus</i>	NC	3.61
Blue shiner	<i>Cyprinella caerulea</i>						<i>Cyprinella monacha</i>	NC	1.58
Beautiful shiner	<i>Cyprinella formosa</i>	Cypriniformes Cyprinidae	Cyprinidae (family)	NC	4.38				
Devils River minnow	<i>Dionda diaboli</i>								
Slender chub	<i>Erimystax cahni</i>								
Mohave tui chub	<i>Gila bicolor mohavensis</i>								
Owens tui chub	<i>Gila bicolor snyderi</i>								
Hutton tui chub	<i>Gila bicolor ssp.</i>								
Borax Lake chub	<i>Gila boraxobius</i>								
Humpback chub	<i>Gila cypha</i>								
Sonora chub	<i>Gila ditaenia</i>								
Gila chub	<i>Gila intermedia</i>								
Yaqui chub	<i>Gila purpurea</i>								
Pahrnagat roundtail chub	<i>Gila robusta jordani</i>								
Virgin River chub	<i>Gila robusta seminuda</i>								
Rio Grand silvery minnow	<i>Hybognathus amarus</i>								
Big Spring spinedace	<i>Lepidomeda mollispinis pratensis</i>								
Little Colorado spinedace	<i>Lepidomeda vittata</i>								
Spikedace	<i>Meda fulgida</i>								
Moapa dace	<i>Moapa coriacea</i>								
Palezone shiner	<i>Notropis albizonatus</i>								
Cahaba shiner	<i>Notropis cahabae</i>								
Arkansas River shiner	<i>Notropis girardi</i>								
Pecos bluntnose shiner	<i>Notropis simus pecosensis</i>								
Topeka shiner	<i>Notropis Topeka</i>								
Oregon chub	<i>Oregonichthys crameri</i>								
Blackside dace	<i>Phoxinus cumberlandensis</i>								
Woundfn	<i>Plagopterus agrentissimus</i>								
Ash Meadows speckled dace	<i>Rhinichthys osculus nevadensis</i>								
Kendall Warm Springs dace	<i>Rhinichthys osculus thermalis</i>								
Foskett speckled dace	<i>Rhinichthys osculus ssp.</i>								
Loach minnow	<i>Tiaroga cobitis</i>								
Bonytail chub	<i>Gila elegans</i>						<i>Gila elegans</i>	NC	2.19

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Cape Fear shiner	<i>Notropis mekistochohas</i>		<i>Notropis mekistochohas</i>	NC	2.09				
Colorado pikeminnow	<i>Ptychocheilus lucis</i>		<i>Ptychocheilus lucis</i>	NC	1.87				
White River springfish	<i>Crenichthys baileyi baileyi</i>	Cyprinodontiformes Goodeidae	Actinopterygii (class)	NC	2.86				
Hiko White River springfish	<i>Crenichthys baileyi grandis</i>								
Railroad Valley springfish	<i>Crenichthys nevadae</i>								
Big Bend gambusia	<i>Gambusia gaigei</i>	Cyprinodontiformes Poeciliidae							
San Marcos gambusia	<i>Gambusia georgei</i>								
Clear Creek gambusia	<i>Gambusia heterochir</i>								
Pecos gambusia	<i>Gambusia nobilis</i>								
Gila topminnow	<i>Poeciliopsis occidentalis occidentalis</i>								
Yaqui topminnow	<i>Poeciliopsis occidentalis sonoriensis</i>								
Unarmored threespine stickleback	<i>Gasterosteus aculeatus williamsoni</i>	Gasterosteiformes Gasterosteidae							
Delta smelt	<i>Hypomesus transpacificus</i>	Osmeriformes Osmeridae							
Tidewater goby	<i>Eucyclogobius newberryi</i>	Perciformes Gobiidae				Perciformes (order)	NC	3.91	
Slackwater darter	<i>Etheostoma boschungii</i>	Perciformes Percidae				<i>Etheostoma</i> (genus)	NC	1.72	
Vermilion darter	<i>Etheostoma chermockii</i>								
Relict darter	<i>Etheostoma chienense</i>								
Etowah darter	<i>Etheostoma etowahae</i>								
Niangua darter	<i>Etheostoma nianguae</i>								
Watercress darter	<i>Etheostoma nuchale</i>								
Okaloosa darter	<i>Etheostoma okaloosae</i>								
Duskytail darter	<i>Etheostoma percnurum</i>								
Bayou darter	<i>Etheostoma rubrum</i>								
Cherokee darter	<i>Etheostoma scotti</i>								
Maryland darter	<i>Etheostoma sellare</i>								
Bluemask darter	<i>Etheostoma sp.</i>								
Boulder darter	<i>Etheostoma wapiti</i>								
Fountain darter	<i>Etheostoma fonticola</i>			<i>Etheostoma fonticola</i> (species)	17.2				0.93
Amber darter	<i>Percina antesella</i>		Percidae (family)	Percidae (family)	NC				1.82
Goldline darter	<i>Percina aurolineata</i>								
Conasauga logperch	<i>Percina jenkinsi</i>								
Leopard darter	<i>Percina pantherina</i>								
Roanoke logperch	<i>Percina rex</i>								
Snail darter	<i>Percina tanasi</i>								
Ozark cavefish	<i>Amblyopsis rosae</i>	Percopsiformes Amblyopsidae				Actinopterygii (class)	NC	2.86	
Alabama cavefish	<i>Spleoplatyrhinus poulsoni</i>								
Little Kern golden trout	<i>Oncorhynchus aguabonita whitei</i>	Salmoniformes Salmonidae	<i>Oncorhynchus</i> (genus)	NC	2.02				
Paiute cutthroat trout	<i>Oncorhynchus clarki seleniris</i>								

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Greenback cutthroat trout	<i>Oncorhynchus clarki stomias</i>				
Gila trout	<i>Oncorhynchus gilae</i>				
Apache trout	<i>Oncorhynchus apache</i>		<i>Oncorhynchus apache</i> (species)	14.47	0.71
Lahontan cutthroat trout	<i>Oncorhynchus clarki henshawi</i>		<i>Oncorhynchus clarki henshawi</i> (species)	20.00	0.98
Atlantic salmon	<i>Salmo salar</i>		<i>Salmo salar</i> (species)	NC	3.87
Bull trout	<i>Salvelinus confluentus</i>		<i>Salvelinus</i> (genus)	13.77	0.68
Pygmy sculpin	<i>Cottus paulus</i>	Scorpaeniformes Cottidae	Actinopterygii (class)	NC	2.86

Freshwater mussels: The acute and chronic criteria for listed mussels are based on the protection of their host fish. For mussels with obligate host fish, the RPA is based on the acute and chronic Assessment Effects Concentrations (EC_A) which represent *the highest* concentration of cyanide where the effects on host fish species are expected to be insignificant (Table 22, see Appendix B for details).

Table 22. Freshwater acute and chronic cyanide criteria for the protection of listed mussels with known obligate host fish species (NC, no change).

Listed Species	Host Fish	Surrogate Taxa	Recommended Acute Criteria [Acute EC _A (ug CN/L)]	Recommended Chronic Criteria [Chronic EC _A (ug CN/L)]
Fat Pocketbook <i>Potamilus capax</i>	Freshwater drum	Perciformes (order)	NC	2.86
Scaleshell Mussel <i>Leptodea leptodon</i>	Freshwater drum	Perciformes (order)	NC	2.86

For mussels with multiple host fish (non-obligates), or for which host fish are unknown, the recommended acute and chronic criteria for listed mussels are based on the protection of fish from the genus *Etheostoma*, family Percidae (Table 23). Percids make up approximately one-third of all known host fish species for listed mussels. For non-obligate listed mussels for which percids have not been identified as hosts, there is a reasonable possibility that these species serve as hosts where they occur. Protection of fish from the genus *Etheostoma* is expected to protect for the majority of species in this family.

For listed mussels either occurring in areas that do not support percids, or are known not to transform on these species, we recommend acute and chronic criteria for listed mussels based on the protection of fish from the class Actinopterygii (Table 23).

Table 23. Freshwater acute and chronic cyanide criteria for the protection of listed mussels with multiple host fish or for which host fish are unknown (NC, no change).

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Listed Species	Host Fish	Surrogate Taxa	Recommended Acute Criteria [Acute EC _A (ug CN/L)]	Recommended Chronic Criteria [Chronic EC _A (ug CN/L)]
Mussels no known obligate host fish	Multiple or unknown	<i>Etheostoma</i> (genus)	NC	1.72
Mussels with non-percid hosts ¹	Non-percid hosts	Actinopterygii (class)	NC	2.86

¹ Habitat is known to not support percid species or mussels species are known to not transform on percids.

Amphipods: The acute and chronic cyanide criteria for protection of listed amphipod species are shown in Table 24. These values are based on the acute and chronic Assessment Effects Concentrations (EC_A) which represent the *the highest* concentration of cyanide where the effects on listed species are expected to be insignificant (see Appendix B for details).

Table 24. Freshwater acute and chronic cyanide criteria for the protection of listed amphipod species (NC, no change).

Listed Species	Order/Family	Surrogate Taxa	Recommended Acute Criteria [Acute EC _A (ug CN/L)]	Recommended Chronic Criteria [Chronic EC _A (ug CN/L)]
Illinois cave amphipod	Amphipoda Cambaridae	<i>Gammarus</i> (genus)	NC	3.33
Noel's Amphipod			NC	3.33

Amphibians: EPA shall implement RPA Alternative #2 for Amphibians.

and/or,

2. In place of RPA 1(b), the EPA shall, subject to the Service's approval, develop and implement the research necessary to replace modeled estimates of species sensitivities to cyanide with direct evidence, using listed species or more closely related surrogates, as the basis for defining cyanide criteria to insure an appropriate level of protection is afforded to listed species and critical habitats addressed by this RPA. This RPA shall be implemented for all amphibians addressed in this biological opinion, and is optional for all other taxa. This task shall be completed by December 1, 2012.

Because this biological opinion has found jeopardy and adverse modification, the EPA is required to notify the Service of its final decision on implementation of the reasonable and prudent alternatives.

11.0 INCIDENTAL TAKE STATEMENT

The Service has developed the following Incidental Take Statement based on the premise that the RPA will be implemented.

Section 9 of the ESA and Federal regulations pursuant to section 4(d) of the ESA prohibit the take of endangered and threatened species, respectively, without special exemption. Take is defined as harass, harm, pursue, hunt, shoot, wound, kill, trap, capture or collect, or to attempt to engage in any such conduct. Harass is defined by the Service as an intentional or negligent act or omission which creates the likelihood of injury to a listed species by annoying it to such an extent as to significantly disrupt normal behavioral patterns which include, but are not limited to, breeding, feeding or sheltering. Harm is defined by the Service to include significant habitat modification or degradation that results in death or injury to listed species by impairing behavioral patterns including breeding, feeding, or sheltering. Incidental take is defined as take that is incidental to, and not the purpose of, the carrying out of an otherwise lawful activity. Under the terms of section 7(b)(4) and section 7(o)(2), taking that is incidental to and not intended as part of the agency action is not considered to be a prohibited taking under the ESA, provided that such taking is in compliance with the terms and conditions of this Incidental Take Statement.

The Service anticipates that with implementation of the RPA, incidental take of the listed fish, mussel, amphipod, and amphibian species considered in this biological opinion is not likely to occur from exposure to cyanide at revised criteria concentrations. However, other elements of water quality standards could allow for exceedance of criteria concentrations and may contribute to incidental take. The other elements of water quality standards will be the focus of subsequent tiered consultations on individual State and Tribal water quality standards. Therefore, no incidental take exemptions are provided in this biological opinion.

12.0 CONSERVATION RECOMMENDATIONS

Section 7(a)(1) of the ESA directs Federal agencies to utilize their authorities to further the purposes of the Act by carrying out conservation programs for the benefit of endangered and threatened species. Conservation recommendations are discretionary agency activities to minimize or avoid adverse effects of a proposed action on listed species or critical habitat, to help implement recovery plans, or to develop information. We recommend that EPA implement the following actions:

1. In consultation with the Service, develop a conservation program for threatened and endangered species and, in collaboration with States and Tribes, develop conservation plans that specifically addresses threats to listed species and how implementation of Clean Water Act programs can ameliorate those threats;
2. Work with the Service and the National Marine Fisheries Service to reinvigorate implementation of the 2001 MOA on ESA and the CWA, especially to address local and regional water quality concerns, research needs, and revisions to the criteria derivation process.

In order for the Service to be kept informed of actions minimizing or avoiding adverse effects or benefitting listed species or their habitats, the Service requests notification of the implementation of any conservation recommendations.

13.0 REINITIATION STATEMENT

This concludes formal consultation on EPA's continuing programmatic approval of cyanide criteria in state and tribal water quality standards. As provided in 50 CFR 402.16, reinitiation of formal consultation is required where discretionary Federal agency involvement or control over the action has been retained (or is authorized by law) and if: (1) the amount or extent of incidental take is exceeded; (2) new information reveals effects of the action that may affect listed species or critical habitat in a manner or to an extent not considered in this opinion; (3) the agency action is subsequently modified in a manner that causes an effect to the listed species or critical habitat not considered in this opinion; or (4) a new species is listed or critical habitat designated that may be affected by the action.

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