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Multiple stress response of lowland stream benthic macroinvertebrates depends on habitat type

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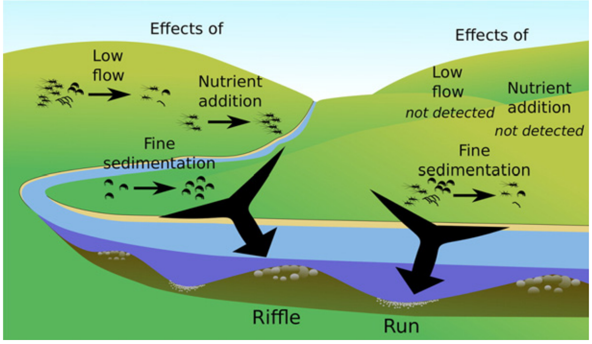
HIGHLIGHTS

- Macroinvertebrates in runs are affected by fine sediment, not by low flow.
- Strong response to low flow in riffles, mitigated by fine sediment and nutrients
- Fast reaction of macroinvertebrates to low flow and fine sediment, if responses detected
- Habitat dependency of effects advises habitat restoration measures.

GRAPHICAL ABSTRACT



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ABSTRACT

Worldwide, lowland stream ecosystems are exposed to multiple anthropogenic stress due to the combination of water scarcity, eutrophication, and fine sedimentation. The understanding of the effects of such multiple stress on stream benthic macroinvertebrates has been growing in recent years. However, the interdependence of multiple stress and stream habitat characteristics has received little attention, although single stressor studies indicate that habitat characteristics may be decisive in shaping the macroinvertebrate response. We conducted an experiment in large outdoor flumes to assess the effects of low flow, fine sedimentation, and nutrient enrichment on the structure of the benthic macroinvertebrate community in riffle and run habitats of lowland streams. For most taxa, we found a negative effect of low flow on macroinvertebrate abundance in the riffle habitat, an effect which was mitigated by fine sedimentation for overall community composition and the dominant shredder species (*Gammarus pulex*) and by nutrient enrichment for the dominant grazer species (*Baetis rhodani*). In contrast, fine sediment in combination with low flow rapidly affected macroinvertebrate composition in the run habitat, with decreasing abundances of many species. We conclude that the effects of typical multiple stressor scenarios on lowland stream benthic macroinvertebrates are highly dependent on habitat conditions and that high habitat diversity needs to be given priority by stream managers to maximize the resilience of stream macroinvertebrate communities to multiple stress.

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1. Introduction

Worldwide, stream benthic macroinvertebrates are facing a plethora of anthropogenic environmental stressors. Altered precipitation patterns (Arnell, 1999) and water abstraction (Vanneuville and Uhel, 2012) create periods with critical low flow that result in loss of macroinvertebrate species typical for stream ecosystems (Graeber et al., 2013; Hille et al., 2014; Lorenz et al., 2016). At the same time, increased loads of fine sediments and elevated nutrient concentrations (Kronvang et al., 2005; Pacheco et al., 2014; Townsend et al., 2008) adversely impact the structure of stream macroinvertebrate communities (Piggott et al., 2015; Townsend et al., 2008; Wagenhoff et al., 2013). Although the interactive effect of these stressors is difficult to predict, stream mesocosm experiments consistently document that the effects of flow reduction on macroinvertebrate community structure are stronger when combined with fine sedimentation than with nutrient enrichment (Piggott et al., 2012; Townsend et al., 2008). Furthermore, these experiments have shown that subsidy effects of nutrient enrichment commonly appear in macroinvertebrates feeding on benthic biofilms (i.e. scrapers; Gruner et al., 2008) and that the effects of nutrient enrichment may be counteracted by fine sedimentation (Wagenhoff et al., 2012).

Field-based studies further document that the susceptibility of stream macroinvertebrate communities to different environmental stressors depends on physical habitat characteristics (Dewson et al., 2007; Rasmussen et al., 2012). For example, the effects of low flow on macroinvertebrate communities depend on the availability of suitable habitats acting as refugia, and this refugial capacity is intrinsically linked to other habitat-specific characteristics such as fine sediment cover and availability of food resources (Lancaster and Hildrew, 1993). Conversely, the effects of fine sediment cover on macroinvertebrate community composition depend on habitat type (Roy et al., 2003). Therefore, evaluating the effects of different stressor combinations in different habitat types is a fundamental prerequisite for robust quantification of the summed impacts of typical anthropogenic stress scenarios on stream macroinvertebrate communities.

In the present study, we explored how habitat-specific characteristics shape the response of macroinvertebrates to flow reduction, nutrient enrichment, and increased coverage of fine sediment by applying multiple combinations of these three stressors in a controlled experimental setup. We used twelve 12-m long outdoor flumes to assess the effects of the stressor combinations on the benthic macroinvertebrate community in contrasting habitat types typical for lowland streams (riffle and run habitats, Pedersen, 2003). In more detail, we assessed the effects of nutrient enrichment during a normal-flow phase followed by a low-flow phase with a reduction in discharge representative of summer time low-flow periods in lowland streams (Graeber et al., 2015). Within the low-flow phase, fine sediment collected from a nearby stream was added to half of the outdoor flumes. During both the normal- and low-flow phases, macroinvertebrate samples were collected with weekly intervals in both habitat types to assess the temporal development of the benthic macroinvertebrate community induced by the selected multiple stressors. Based on these samples, we tested the following hypotheses with specific focus on the temporality of responses:

1. The combined effects of low flow and fine sediment addition on macroinvertebrate composition depend on habitat characteristics, with the strongest influence in run habitats compared with riffle habitats as fine sediment accumulation in run habitats is more pronounced than in riffle habitats.
2. Nutrient enrichment changes the structure of the benthic macroinvertebrate community in both run and riffle habitats towards increasing abundance of grazers due to a stimulating effect of nutrients on the biomass of epibenthic algae. This effect is reduced by addition of fine sediment, which diminishes the algae biomass available to grazers.

2. Materials and methods

2.1. Experimental setup

We conducted the experiment in twelve outdoor flumes during summer 2015 in Denmark (56°4′ N, 9°31′ E). The flumes consisted of rectangular 12 m long, 60 cm wide, and 30 cm deep channels. In each flume, four run-riffle sequences were created, resembling natural habitat conditions in lowland streams (Pedersen, 2003). Each of the four riffle and run habitats covered 1.5 m of the stream flume length with an average sediment depth of 5.5 cm (0.5–2, 2–4, 4–8, and 8–16 mm grain-sized sediment at a volume ratio of 3:1:1:1) in the runs and 15 cm (4–8, 8–16, 16–32, 32–64, and 64–120 mm grain-sized sediment in equal volumes) in the riffles. The stream flumes were continuosly supplied with water from a nearby source stream (Lemming stream) using a central stream feeder pump. To obtain the desired discharge volume, part of the water volume was recycled from twelve water reservoirs. With this setup, invertebrate drift into the flumes was allowed in order to simulate the natural drift within the source stream (refer to Neif et al. (2016) for further details on the stream flume setup).

2.2. Experimental phases

2.2.1. Pre-treatment phase

The pre-treatment phase was initiated on 15 June 2015 and lasted eight weeks. During this phase, the average discharge of the flumes was 5.4 L s^{-1} (±0.4 1SD, $n = 12$), which is comparable with discharges typical for hydrologically undisturbed small lowland streams (Pedersen, 2003).

After two weeks, benthic macroinvertebrates were collected from the source stream using kick sampling along a reach of approximately 500 m to be introduced to the flumes. In total, 120 kick samples were transferred to each flume, corresponding to a sampled source stream bed area of approximately 7.5 m^2, which is of a size similar to one flume (7.2 m^2). The colonization was successful as the source stream and the stream flume contained similar macroinvertebrate communities (see Appendix A for detailed comparisons of the macroinvertebrate communities in stream flumes and source stream and Appendix B for species lists).

2.2.2. Normal-flow phase

The normal-flow phase lasted four weeks and was initiated immediately after the pre-treatment phase. Six randomly chosen flumes were subjected to nutrient enrichment by adding fertilizer (SweDane NPK 21-3-10 and GrowHow NS 24-6, DLG, Copenhagen, Denmark) (NP treatment). The nutrients were mixed in a 600 L tank and continuously transferred to the respective flumes using a peristaltic pump (BVP-Process with a 12-channel CA pump head, Ismatec, Wertheim, Germany). In brief, target concentrations of nitrate-N, ammonium-N, and phosphate-P were elevated by a factor of 2, 20, and 4 in the NP treatment (Table 1, see Appendix A for methods used to quantify nutrient concentrations). These increases in dissolved inorganic nitrogen and phosphate represented concentrations in lowland streams draining catchments with intensive agriculture (Larsen et al., 1999).

The cover of fine sediment was low during the normal-flow phase (see Appendix A for estimation methods of fine sediment cover), and the flow was therefore sufficient to avoid precipitation of suspended matter from the water column (Table 1).

Twelve leaf litter bags with coarse mesh size (10 mm), allowing macroinvertebrates to access the leaf material, were deployed in each flume between the riffle and run habitats at the beginning of the normal-flow phase. Each bag contained 1 (±0.01) g DW leaves of beech (*Fagus sylvatica* L.), which is the dominant broad-leaf tree species in the area.

Table 1
Mean ± 1SD of discharge, current velocity, water depth, temperature, nutrient concentrations, and fine sediment cover during the normal-flow and low-flow phase. FS = fine-sediment treatments, NP = nutrient-enrichment treatments.

	Normal-flow phase (NF)	Low-flow phase (LF)
Discharge (L s^{-1}, n = 48)	4.65 ± 0.28	1.05 ± 0.12
Current velocity (cm s^{-1}, n = 12)		
Run, below surface	0.15 ± 0.05	0.02 ± 0.01
Run, half depth	0.11 ± 0.04	0.02 ± 0.01
Run, above sediment	0.07 ± 0.04	0.02 ± 0.01
Riffle	0.46 ± 0.04	0.04 ± 0.02
Water depth (cm, n = 12), run[a]	10.4 ± 0.9	12.4 ± 1.0
Temperature (°C, NF n = 31,667, LF n = 33,408)	12.9 ± 1.3	12.1 ± 1.1
NO_3^- (mg N L^{-1}, NF n = 42, LF n = 54)		
non-NP treatments	1.22 ± 0.14	1.19 ± 0.23
NP treatments	2.96 ± 0.79	2.77 ± 0.75
NH_4^+ (mg N L^{-1}, NF n = 42, LF n = 54)		
non-NP treatments	0.09 ± 0.15	0.13 ± 0.18
NP treatments	1.81 ± 0.75	1.84 ± 0.81
PO_4^{2-} (mg P L^{-1}, NF n = 42, LF n = 54)		
non-NP treatments	0.012 ± 0.005	0.014 ± 0.004
NP treatments	0.049 ± 0.019	0.042 ± 0.016
Fine sediment cover, run (%, NF n = 48, LF n = 24)[b]		
non-FS treatments	9.3 ± 10.7	22.2 ± 22.0
FS treatments	–	79.6 ± 19.3

[a] Water depth was measured in detail in the run habitat to check for too low water depths during the low-flow phase; it was always >3 cm in the riffle.

[b] No fine sediment cover was detectable in the riffle habitat; during normal flow the fine sediment treatment was not applied.

2.2.3. Low-flow phase

Immediately after the normal-flow phase, the low-flow phase was initiated by reducing discharge but keeping the water depth and water temperature stable (Table 1). Current velocity was reduced by 90% compared with the normal-flow phase (Table 1; see Appendix A for methods used to measure current velocity).

The NP treatment was continued in the low-flow phase to maintain stable eutrophic conditions.

After initiating the low-flow phase, fine sediment was added to six randomly chosen flumes, hereby creating four treatments (n = 3): no treatment, NP, fine sediment addition (FS), and combined nutrient enrichment and fine sediment addition (NP + FS). The organic-rich fine sediment was collected from the source stream (Lemming stream) and introduced manually into the flumes until >90% fine sediment cover was reached (consult Neif et al. (2016) for details). On average, the fine sediment treatments (FS) were characterized by an increase in the fine sediment cover by a factor of 4 compared with the flumes with no sediment addition (Table 1).

Similar to the normal-flow phase, leaf litter bags with coarse mesh size (10 mm) and each containing 1 (±0.01) g DW of beech leaves were deployed at the beginning of the low-flow phase. In total, 12 leaf bags were deployed in each flume and positioned between the riffle and run habitats.

2.3. Macroinvertebrate sampling and identification

One week before the start of the normal-flow phase and weekly during the normal-and low-flow phases (9 sampling occasions), macroinvertebrates were sampled using a surber sampler (area = 195 cm^2, mesh size = 200 μm). For each flume and on each sampling occasion, one Surber sample was collected in an upstream run or riffle habitat and one in a downstream run or riffle habitat. The samples were pooled habitat-wise (riffle or run, resulting in 216 samples in total). The purpose of restricting the number of samples per sampling occasion was to avoid removing an excess number of individuals from the stream flumes in the repeated samplings.

All macroinvertebrate samples were preserved in 95% ethanol in the field. All macroinvertebrate taxa were identified to species level except for Chironomidae (sub-family), Oligochaeta (class), as well as Empididae, Tipulidae, and Simuliidae (family). In cases where individuals were too small to be identified to species level, they were identified to genus level, and at this level all individuals of the same genus were used for further statistical computations. Please refer to Appendix B for the species data used in our study.

2.4. Statistics

2.4.1. Community response to multiple stress

We used principal response curves (PRC) to analyse the temporal development in macroinvertebrate community composition in the stream flumes (Van den Brink and Ter Braak, 1999). The PRC model is based on the first axis of a principal coordinate analysis using Bray-Curtis similarity to generate site and species scores using the "capscale" function of the "vegan" R package (Oksanen et al., 2015) in R (version 3.3.2, R Core Team, 2016). The PRC consists of treatment scores and species weights. The treatment scores can be interpreted as the principal response of the community to a treatment (Van den Brink and Ter Braak, 1999). The species weights allow determination of taxon-specific reactions since higher species weights indicate stronger responses of the species to the treatment patterns in the PRC (Van den Brink and Ter Braak, 1999). Taxa with near zero species weights show either no response or one that is unrelated to the patterns represented in the PRC. Moreover, the direction of the species weights determines the direction of the response of the species to the treatments (Van den Brink and Ter Braak, 1999).

To investigate habitat-specific stressor-induced effects on macroinvertebrate communities, separate PRCs were performed for riffle and run habitats and for the normal-flow phase and the low-flow phase as the control differed between the phases. The control for the habitat-specific PRCs of the normal-flow phase was the habitat-specific (riffle or run) species composition in the channels in the last week of the pre-experimental phase, one week before the start of the normal-flow phase. For the low-flow phase, the control was the habitat-specific average of macroinvertebrate species-specific densities across all weeks of the normal-flow phase for the six channels without nutrient enrichment. An ANOVA-like permutation test (999 iterations) was used to assess the significance of the PRC model using the "anova.cca" function of the "vegan" package (Oksanen et al., 2015) in R (R Core Team, 2016). All PRC models significantly explained the data ($F > 2.8$, $p < 0.001$), except for the PRC of the run habitat during normal flow, which was only marginally significant ($F = 1.8$, $p = 0.06$), however.

To assess habitat-specific effects of low flow with and without fine sediment addition, the benthic macroinvertebrate community structure was analysed with separate permutational multivariate analyses of variance (PERMANOVA, 999 iterations, Bray-Curtis dissimilarity) (adonis function, vegan package in R; Oksanen et al., 2015) for each of the two habitats within the flumes with FS, FS + NP and without FS or NP. Here, we compared the last week of the normal-flow phase with the first week of the low-flow phase to minimize potential temporal effects interfering with the effect of low flow. Within the PERMANOVAs, the phase (normal or low flow) was used as main factor and the flumes were used as strata.

PERMANOVAs (999 iterations, Bray-Curtis dissimilarity) were also used to assess the effects of the NP treatment on the macroinvertebrate community composition during the normal-flow phase and the effects of the FS, NP, and FS + NP treatments and their interaction during the low-flow phase. Weeks were used as strata for the permutations due to the repeated nature of the sampling.

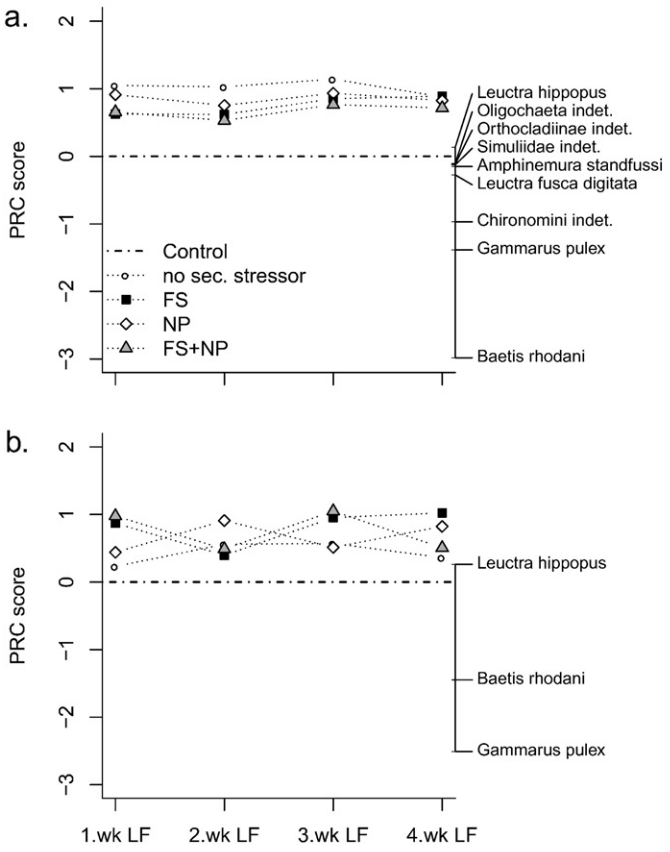

Fig. 1. Principal response curve of macroinvertebrate composition during each week of the low-flow phase in the riffle (a) and run (b) habitats with or without fine sediment (FS), the nutrient enrichment treatment (NP), the combination of FS and NP (FS + NP) or without any secondary stressor (no sec. stressor). The control is the average of taxa-specific densities during all weeks of the normal-flow phase. Only means of the treatment scores are shown for each point ($n = 3$) and only taxa with species weights > 0.1 are plotted (see the Statistics section for details on the meaning of the treatment scores and species weights).

2.4.2. Species-specific response to multiple stress

We investigated the species-specific response to the FS, NP, and FS + NP treatments within the riffle and run habitats based on the abundances of *Gammarus pulex* L. and *Baetis rhodani* Pictet. We used linear mixed-effects models with channel as random intercept (lme function, nlme package in R, Pinheiro et al., 2015) to test the effects of the NP treatment during the normal-flow phase and the FS, NP, and FS + NP treatments during the low-flow phase. We ran the linear-mixed effects models separately for the two phases and the two habitats to obtain phase- and habitat-specific responses. The above to species were selected because they constituted 73% of the macroinvertebrate density in the experiment, and their abundances thus strongly affected community statistics and assisted in their interpretation. Furthermore, *B. rhodani* is a grazer presumably influenced by the hypothesized nutrient enrichment-induced increase in algal biomass, whereas *G. pulex* is a shredder and therefore less likely to be influenced by this (Colling and Schmedtje, 1996).

3. Results

3.1. Effects of low flow and fine sediment

During the low-flow phase, we found a significant mitigating effect of fine sedimentation in the riffle habitat; hence the flumes with FS treatment were not as strongly altered in their macroinvertebrate composition relative to the control as flumes without FS treatment (Fig.1a, Table 2). Due to the mitigating effect of fine sedimentation in the riffle habitat, the change from normal to low flow was not significant in combination with the FS treatment (Table 3). Without application of the FS treatment, we detected a significant effect of the change from normal to low flow in the riffle habitat (Table 3).

In contrast, in the run habitat, the flumes with FS treatment showed the strongest deviation from the control, indicating that the FS treatment created the strongest alteration in macroinvertebrate composition (Fig. 1b, Table 2). Consequently, there was a significant effect of the change from normal to low flow in the run habitat, when this change was combined with the FS treatment (Table 3).

For *G. pulex*, we also detected habitat-specific effects of the FS treatment. Significantly higher abundances occurred with than without the FS treatment in the riffle habitats (Fig. 2a), whereas no effect of the FS treatment appeared in the run habitats (Fig. 2b). For *B. rhodani*, we found no effect of the FS treatment, but a strong habitat-specific abundance decline occurred after the flow reduction (Fig. 3), this being more pronounced in the riffle (Fig. 3a) than in the run habitats (Fig. 3b).

3.2. Effects of nutrient enrichment during normal and low flow

The NP treatment did not significantly affect the macroinvertebrate community composition during either normal or low flow (Table 3, Figs. 1, 4). However, the densities of *B. rhodani* were higher in riffle habitats in flumes with NP treatment under both normal-and low-flow conditions (Fig. 3a) which was not the case in run habitats (Fig. 3b).

4. Discussion

4.1. Habitat-specific effects of low flow and fine sediment

We found that the combined effects of low flow and fine sediment on the macroinvertebrate community were habitat dependent. This is in accordance with empirical evidence from other field studies also reporting habitat-specific effects of these two stressors (Lancaster and Hildrew, 1993; Roy et al., 2003). Furthermore, we observed that the response of the macroinvertebrate community to fine sedimentation and low flow occurred rapidly, being detectable after just one week with no or only little further development over time.

A combination of different mechanisms likely contributed to the stronger response of the macroinvertebrate community to fine sedimentation in the run habitat than in the riffle habitat. Fine sediment has been shown to affect benthic macroinvertebrates in different

Table 2
Effects of the secondary stressor treatments and their interactions on macroinvertebrate composition during the normal-flow and low-flow phase. Secondary stressor treatments were nutrient enrichment (NP) during the normal-flow phase and fine sedimentation (FS) and NP during the low-flow phase. The results from permutational multivariate analyses of variance (PERMANOVA) with sampling weeks as strata are shown ($n = 48$). $^{ns}p > 0.05$, $^{*}p < 0.05$, $^{***}p < 0.001$.

Habitat	Flow phase	NP	FS	NP × FS
Riffle	Normal flow	$F = 1.7, R^2 = 0.04^{ns}$	–	–
Riffle	Low flow	$F = 1.6, R^2 = 0.02^{ns}$	$F = 9.3, R^2 = 0.17^{***}$	$F = 0.8, R^2 = 0.01^{ns}$
Run	Normal flow	$F = 1.0, R^2 = 0.02^{ns}$	–	–
Run	Low flow	$F = 1.6, R^2 = 0.03^{ns}$	$F = 2.3, R^2 = 0.05^{*}$	$F = 1.0, R^2 = 0.02^{ns}$

ways: i) by reducing the stability and thereby the suitability of habitats, creating increased drift (Wood and Armitage, 1997) and limiting the access to food sources (Matthaei et al., 2010), ii) by clogging of interstitial spaces, reducing the availability of suitable habitats (Wood and Armitage, 1997), and iii) by increasing benthic respiration, hereby decreasing the daily minimum concentrations of oxygen (García and Pardo, 2016; Wood and Armitage, 1997). In our study, the effect of clogging of interstitial spaces was probably limited in the run habitat as we mainly used sand as original sediment to simulate typical Danish conditions (Pedersen, 2003). We found that the deployed beech leaves were partly covered with fine sediment and that the applied fine sediment was relatively rich in organic matter. Consequently, the combined effect of loss of food sources, reduced habitat stability, and increased benthic respiration probably caused the strong response of the benthic macroinvertebrates to fine sedimentation in the run habitat.

In the riffle habitats, fine sediment seemed to counteract the effects of low flow on the overall macroinvertebrate community composition. Furthermore, the abundance of *G. pulex* was higher in the riffle than in the run habitats in the flumes with fine sediment treatment. However, we did not observe cover by fine sediment, a commonly used indicator of the level of fine sedimentation (e.g. Matthaei et al., 2010; Piggott et al., 2012; Wagenhoff et al., 2012), indicating that the fine sediment may have accumulated at deeper sites, clogging of the interstitial spaces. This likely reduced the ability of *G. pulex* and other taxa to migrate vertically into the sediment (Vadher et al., 2015) and probably increased the vulnerability of *G. pulex* and other species to lower water depth (Vadher et al., 2015), and higher water temperatures (Vorste et al., 2016).

We found relatively constant effects of fine sedimentation and low flow on macroinvertebrate composition, although invertebrate drift from the source stream occurred. Therefore, despite the potential for recovery by drift, the ecosystem conditions were not sufficiently favourable to allow recovery of the macroinvertebrate community from the effects of fine sedimentation or low flow.

4.2. Habitat-specific effects of nutrient enrichment

Table 3
Effects on macroinvertebrate composition by the change from normal to low flow with or without additional fine-sediment treatment. Results of permutational multivariate analyses of variance (PERMANOVA) with stream flumes as strata are shown (n = 6).*p < 0.05, FS = fine-sediment treatment.

Habitat	FS	F	R^2
Riffle	No	4.3	0.30*
Run	No	1.1	0.10
Riffle	Yes	2.8	0.22
Run	Yes	5.9	0.37*

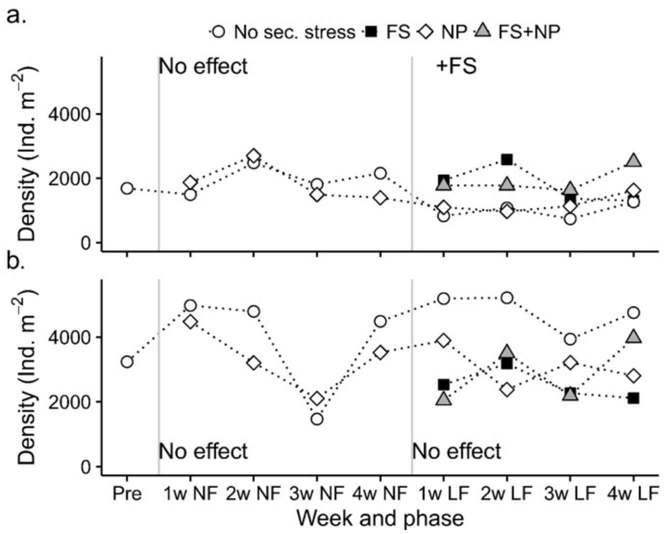

Fig. 2. Densities of *Gammarus pulex* during the normal-flow and low-flow phase in the riffle (a) and run (b) habitats. NP = nutrient-enrichment treatment, FS = fine-sediment treatment, FS + NP = both FS and NP, no sec. stressor = neither FS nor NP. Significant positive or negative effects ($p < 0.05$) of the secondary stressors within the phases were assessed with linear mixed-effects models and are depicted as + or − followed by the stressors, or as "No effect" if no secondary stressor effect was found. Only means are shown for each point (pre-experimental phase n = 12, normal-flow phase n = 6, low-flow phase n = 3) for clearer presentation.

In contrast to our second hypothesis, we found no significant overall response of macroinvertebrate community composition to nutrient enrichment. This is likely related to the already high background concentrations of dissolved inorganic nitrogen, where a further enrichment would either yield no or even negative toxic effects on benthic macroinvertebrates (Camargo et al., 2005; Camargo and Alonso, 2006; Wagenhoff et al., 2011). This may also explain why our findings contradict those obtained in earlier studies under less nitrogen-rich conditions (Bourassa and Cattaneo, 2000; Piggott et al., 2012; Townsend et al., 2008; Wagenhoff et al., 2011, 2012). Additionally, the dominant species within our experiment likely fed on sources of terrestrial organic matter, such as CPOM and fine detritus (*G. pulex*, *Leuctra fusca*, *Leuctra hippopus*, Chironomini) (Colling and Schmedtje, 1996; López-Rodríguez et al., 2012) and less on autochthonous sources, such as epibenthic algae. Therefore, their response may not have been dependent on the influence of nutrient enrichment on biofilm development. In line with this, we observed an increase in the abundance of the grazer *B. rhodani* in the riffle habitat, probably as a consequence of the increased benthic algae growth (Dudley and D'Antonio, 1991). The missing effect of nutrient enrichment on *B. rhodani* within the run habitat implies that benthic algal growth was not affected by nutrient enrichment. This is in accordance with an earlier study conducted within the same stream flumes under the same experimental conditions, which did not reveal any effect of nutrient enrichment on the benthic algae

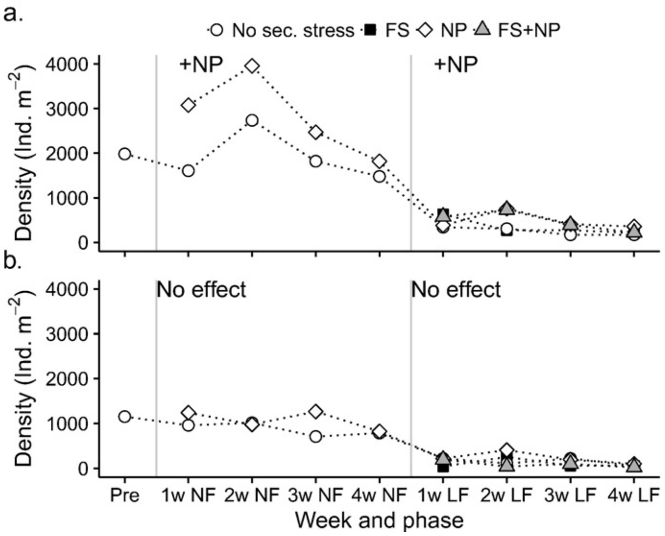

Fig. 3. Densities of *Baetis rhodani* during the normal-flow and low-flow phase in the riffle (a) and run (b) habitats. NP = nutrient-enrichment treatment, FS = fine-sediment treatment, FS + NP = both FS and NP, no sec. stressor = neither FS nor NP. Significant positive or negative effects ($p < 0.05$) of the secondary stressors within the phases were assessed with linear mixed-effects models and are depicted as + or − followed by the stressors, or as "No effect" if no secondary stressor effect was found. Only means are shown for each point (pre-experimental phase n = 12, normal-flow phase n = 6, low-flow phase n = 3) for clearer presentation.

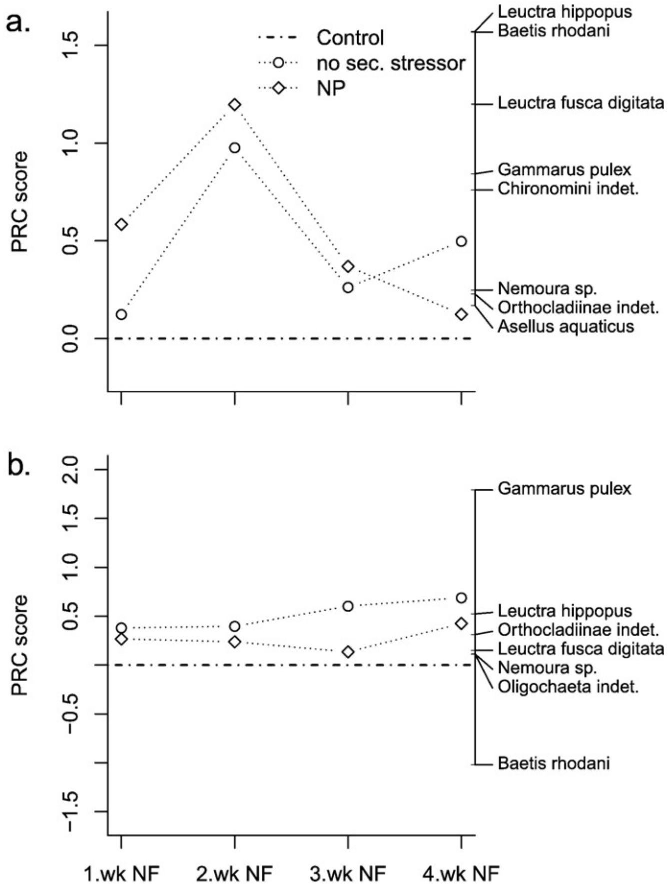

Fig. 4. Principal response curve of macroinvertebrate composition during each week of the normal-flow phase in the riffle (a) and run (b) habitats with or without the nutrient enrichment treatment (NP). The control is the macroinvertebrate composition determined for the last week of the pre-treatment phase. Only means of the treatment scores are shown for each point ($n = 6$) and only taxa with species weights > 0.1 are plotted (please see the Statistics section for details on the meaning of the treatment scores and species weights).

biovolume in the run habitat (Neif et al., 2016). However, in Neif et al. (2016) the riffle habitat was not investigated, and therefore our claim of increased benthic algae growth with nutrient enrichment in the riffle habitat remains to be supported by direct evidence.

4.3. Implications

Benthic macroinvertebrates are sensitive indicators of ecosystem status and health and are therefore used as such within monitoring programs and legislation (Friberg, 2014). Our findings showed a rapid response of the macroinvertebrate community to low flow, fine sedimentation, and nutrient enrichment, being highly dependent on habitat characteristics. This suggests a rapid ecosystem-wide response that may alter with habitat type and implies that more diverse physical stream conditions may enhance the resilience of the benthic macroinvertebrate community to multiple stress effects in streams. This notion is supported by an earlier finding in a field study with flow and sedimentation as the main stressors (Lorenz et al., 2016). Hence, maintenance or restoration of a diverse, natural range of habitats seems to be an obvious measure to apply in order to mitigate multiple stress effects in streams. Furthermore, our discovery that even short-term low-flow and fine-sedimentation events may strongly affect benthic macroinvertebrates in lowland agricultural streams clearly highlights the need for reducing the number of short multiple stress events. For example, fine sediment influx into stream ecosystems due to river bank and catchment erosion (Stutter et al., 2012) could be diminished through the use of vegetated sediment filters (buffer zones) with special focus on areas with high erosion risk (Gumiere et al., 2011). Management options to reduce the number of low-flow events created by water abstraction may be decreased cultivation of water-demanding crops and/or more efficient irrigation (e.g. droplet irrigation) in areas with intensive agriculture and dry summers (Vanneuville and Uhel, 2012). In the long term, however, the number of low-flow events can only be minimized by abating the anthropogenic climate change that not only affects stream hydrology directly via the predicted reduction in summer precipitation in coastal Europe but also increases the need for water abstraction during dry periods (Vanneuville and Uhel, 2012).

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Appendix A. Supplementary data

Supplementary data to this article can be found online at http://dx.doi.org/10.1016/j.scitotenv.2017.05.102.

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