

APPLIED ISSUES

# Assessment of European streams with diatoms, macrophytes, macroinvertebrates and fish: a comparative metric-based analysis of organism response to stress

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## SUMMARY

1. Periphytic diatoms, macrophytes, benthic macroinvertebrates and fish were sampled with standard methods in 185 streams in nine European countries to compare their response to degradation. Streams were classified into two main stream type groups (i.e. lowland, mountain streams); in addition, the lowland streams were grouped into four more specific stream types.
2. Principal components analysis with altogether 43 environmental parameters was used to construct complex stressor gradients for physical–chemical, hydromorphological and land use data. About 30 metrics were calculated for each sample and organism group. Metric responses to different stress types were analysed by Spearman Rank Correlation.
3. All four organism groups showed significant response to eutrophication/organic pollution gradients. Generally, diatom metrics were most strongly correlated to eutrophication gradients (85% and 89% of the diatom metrics tested correlated significantly in mountain and lowland streams, respectively), followed by invertebrate metrics (91% and 59%).
4. Responses of the four organism groups to other gradients were less strong; all organism groups responded to varying degrees to land use changes, hydromorphological degradation on the microhabitat scale and general degradation gradients, while the response to hydromorphological gradients on the reach scale was mainly limited to benthic macroinvertebrates (50% and 44% of the metrics tested correlated significantly in mountain and lowland streams, respectively) and fish (29% and 47%).
5. Fish and macrophyte metrics generally showed a poor response to degradation gradients in mountain streams and a strong response in lowland streams.
6. General recommendations on European bioassessment of streams were derived from the results.

*Keywords:* eutrophication, hydromorphology, land use, metrics, microhabitats, perturbation

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## Introduction

Streams are among the most threatened habitat types. Although the type and severity of human-generated pressures affecting the integrity of streams varies across Europe, the major drivers can be summarised as multiple use (such as fisheries, navigation and drinking water extraction), nutrient enrichment and organic pollution, acidification and alterations of hydrology and morphology (Stanner & Bordeau, 1995; Malmqvist & Rundle, 2002). Awareness of the deleterious effects of human pressures on streams has resulted in a long history of monitoring using biological indicators (Hellowell, 1986; De Pauw & Hawkes, 1993; Rosenberg & Resh, 1993; Knobon, Roos & van Oirschot, 1995). For example, the use of invertebrates in bioassessment began as early as the early 1900s (Kolkwitz & Marson, 1909). Building on this tradition, the European Commission recently passed legislation mandating the use of different organism groups to monitor the integrity of inland waters and coastal regions (Council of the European Communities, 2000). The Water Framework Directive advocates the use of different organism groups such as benthic diatoms, macrophytes, invertebrates and fish to be used either singly or together in assessing the ecological integrity of stream ecosystems.

A number of studies have evaluated the effects of different stressors on stream assemblages and many assessment systems have been developed including one or more taxonomic groups. *Benthic diatoms* are often used for assessing nutrient enrichment (e.g. Kelly, Penny & Whitton, 1995; Rott *et al.*, 1997, 1999; Coring, 1999), salinity (Ziemann, 1999) and acidity (Coring, 1993; Battarbee *et al.*, 1997; van Dam, 1997). Consequently, several studies have addressed the tolerances and preferences of diatoms along a number of environmental gradients (e.g. salinity, pH, trophy, saprobity and current preference; e.g. Denys, 1991a,b; van Dam, Mertens & Sinkeldam, 1994; Rott *et al.*, 1997). *Perennial macrophytes* (flowering plants, vascular cryptogams, bryophytes and macro-algae) are regarded as good indicators of long-lasting habitat changes that can integrate the temporal effects of disturbances (Westlake, 1975; Tremp & Kohler, 1995). Stream assessment methods using macrophytes are relatively scarce compared with benthic diatoms, but more recently standard methods have been proposed for monitoring (e.g. in Denmark, Svendsen &

Rebsdorf, 1994; UK, Holmes *et al.*, 1999; Germany, Schneider, Krumpholz & Melzer, 2000; France, Haury *et al.*, 2002). Macrophyte-based methods are mainly focussed on assessing nutrient enrichment (e.g. Holmes *et al.*, 1999; Schneider *et al.*, 2000; Haury *et al.*, 2002) and acidification (e.g. Tremp & Kohler, 1995), although recently assessment systems have addressed general stream degradation (Passauer *et al.*, 2002; Schaumburg *et al.*, 2004). *Benthic macroinvertebrates* are used for assessing the effects of multiple stressor types such as organic pollution (e.g. Zelinka & Marvan, 1961; Armitage *et al.*, 1983; Statzner, Bis & Usseglio-Polatera, 2001), hydromorphological degradation (e.g. Statzner *et al.*, 2001; Buffagni *et al.*, 2004; Lorenz *et al.*, 2004a), acidification (e.g. Townsend, Hildrew & Francis, 1983; Sandin, Dahl & Johnson, 2004) and general stress (e.g. Barbour *et al.*, 1998; Dolédec, Statzner & Bournard, 1999; Karr & Chu, 1999). *Fishes* have traditionally been used as indicators for habitat degradation (e.g. Gorman & Karr, 1978) and flow regulation (e.g. Bain, Finn & Booke, 1988), although some studies have used fish to assess water pollution (Belpaire *et al.*, 2000) and land use influences (Snyder *et al.*, 2003). Moreover, as fishes are migratory they are considered as sensitive organisms for continuum disruptions (Northcote, 1998). According to a recent study of European river assessment methods (<http://starwp3.eu-star.at/>), benthic macroinvertebrates are frequently used in European biomonitoring programmes, as they are relatively easy to sample and to identify and taxon traits are well known; most river assessment systems presently applied rely on this taxonomic group. Benthic diatoms are also often used, because of a strong response to eutrophication, while the use of macrophytes and fish in biomonitoring is comparatively recent.

Because of recent European legislation (Council of the European Communities, 2000), these four organism groups will constitute the backbone of future stream biomonitoring programmes (Heiskanen *et al.*, 2004). Similarly, in North America benthic diatoms, macroinvertebrates and fish are frequently used together to assess the integrity of stream ecosystems (Barbour *et al.*, 1998). Although the response of individual organism groups to different stress types is putatively well established, surprisingly few studies have compared the suitability and performance of different organism groups in biomonitoring (e.g. Lenat & Crawford, 1994; Lancaster *et al.*, 1996; Hall

*et al.*, 1999; Sonneman *et al.*, 2001; Triest *et al.*, 2001; Walsh *et al.*, 2001; Sawyer *et al.*, 2004; Mazor *et al.*, 2006) and, with the exception of the study by Ormerod *et al.* (1994) in Nepal, we are not aware of a single study comparing the concurrent response of diatoms, macrophytes, macroinvertebrates and fish.

Based on a large dataset from Central and Northern Europe, this study aimed to compare and contrast the response of diatoms, macrophytes, macroinvertebrates and fishes to different stressors (eutrophication/organic pollution, catchment land use, hydromorphological degradation on the reach and microhabitat scales, 'general degradation') and to test if these organism-specific responses vary between stream types. In brief, from the above mentioned studies, we hypothesised that all four organism groups would be capable of indicating eutrophication/organic pollution intensity because of increased nutrient availability and/or because of temporary oxygen depletion. However, the speed of response of

each four organism groups should differ, as longevity of each group varies strongly e.g. between diatoms and fish. We anticipated that the impact of habitat degradation would be strongest on fishes, particularly on the reach scale, as impairment may affect spawning and feeding habitats, followed by macroinvertebrates and macrophytes, which may be most affected by habitat degradation on the microhabitat scale (Passauer *et al.*, 2002; Buffagni *et al.*, 2004; Lorenz *et al.*, 2004a), and weakest on benthic diatoms. Nutrient enrichment was expected to show the reverse, as it might affect autotrophic organism most strongly.

## Methods

### Stream types and sites

In total, 185 streams were sampled in 2002 and 2003 (Fig. 1; Table 1) as part of the European project STAR (standardisation of river classifications: framework

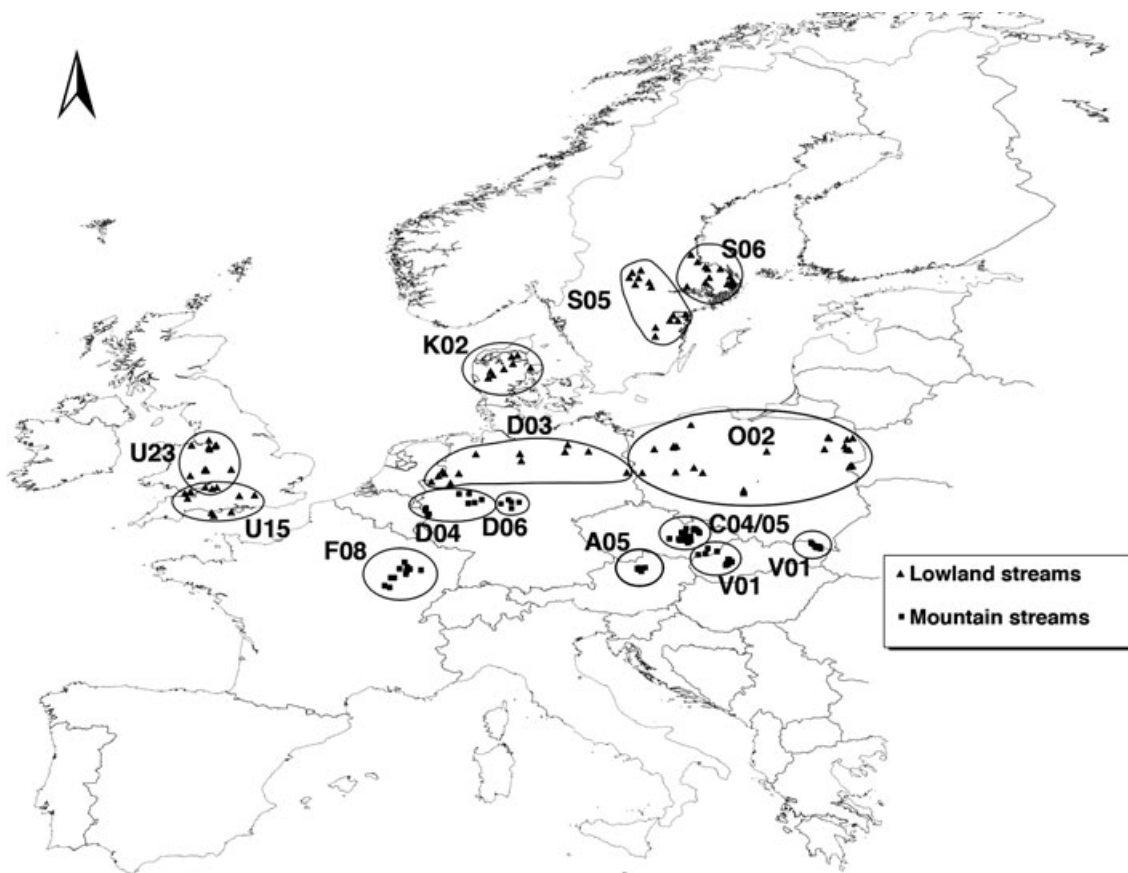


Fig. 1 Location of the sampling sites. For type numbers see Table 1.

**Table 1** Selected variables and number of samples collected in the stream types investigated. Stream type code according to Furse *et al.* (2006). Ecoregion name and number according to Illies (1978).

Stream type no.	Country	Ecoregion name and number	Altitude (m.a.s.l.) range	Number of sampling sites	Diatom samples	Macrophyte samples	Macroinvertebrate samples	Fish samples
Lowland streams								
D03	Germany	Central Lowlands (14)	16–66	10	10	10	10	10
K02	Denmark	Central Lowlands (14)	1–60	11	11	11	11	11
O02	Poland	Central Lowlands (14), Eastern Lowlands (16)	40–190	25	25	25	24	24
S05	Sweden	Central Lowlands (14)	3–261	16	14	15	16	16
S06	Sweden	Central Lowlands (14)	2–28	11	11	11	11	11
U15	United Kingdom	England (18)	5–75	13	13	13	13	13
U23	United Kingdom	England (18)	0–120	12	12	12	12	12
Lowland streams sum				98	96	97	97	97
Mountainous streams								
A05	Austria	Central Sub-alpine Mountains (9)	454–529	11	11	8	10	9
C04	Czech Republic	Central Sub-alpine Mountains (9)	244–485	14	14	13	14	12
C05	Czech Republic	Central Sub-alpine Mountains (9)	253–481	10	10	10	10	9
D04	Germany	Central Sub-alpine Mountains (9)	174–490	10	10	9	10	9
D06	Germany	Central Sub-alpine Mountains (9)	140–275	6	6	6	6	6
F08	France	Western Sub-alpine Mountains (8)	260–417	12	12	12	12	12
V01	Slovakia	Central Sub-alpine Mountains (9), Carpathian (10)	220–534	24	24	9	24	24
Mountain streams sum				87	87	67	86	81
Overall sum				185	183	164	183	178

method for calibrating different biological survey results against ecological quality classifications to be developed for the Water Framework Directive; Furse *et al.*, 2006). An earlier ordination-based analysis of these data showed that community composition differed among the mountain and lowland streams (Verdonschot, 2006), hence two main stream groups were analysed here: (i) small mountain (87 sites) and (ii) medium-sized lowland (98 sites) streams. In addition, these two groups were further divided into seven subgroups or 'stream types'. Lowland streams were situated at mean altitudes of  $69 \pm 63$  m a.s.l., had larger catchments (mean =  $208 \pm 195$  km<sup>2</sup>) and were more nutrient rich (mean TP  $\approx 1 \pm 2.6$  mg L<sup>-1</sup>) than mountain streams ( $353 \pm 91$  m a.s.l.,  $55 \pm 72$  km<sup>2</sup>,  $0.19 \pm 0.27$  mg L<sup>-1</sup>, respectively) (Table 1). The percentage of catchments classified as cropland was similar between the two

main stream-groups (mountain streams: mean = 23%; lowland streams: mean = 29%); however, mountain stream catchments generally had more forest (mean = 59%) compared with lowland streams (38%). Substratum particle size also differed markedly between the two groups, with lowland streams having a higher proportion of sand/silt (mean 40%), whilst mountain streams had more cobbles (mean 37%). TP ranged from being not detectable to 1270 µg L<sup>-1</sup> in mountain streams and to 15 430 µg L<sup>-1</sup> in lowland streams, with the most strongly enriched sites being part of stream types S05/S06 and O02. Substratum varied from microhabitats with 100% cobbles to those with 100% sand. These wide ranges in nutrients and substratum indicate that relatively broad environmental gradients were sampled to test the utility of the various organism groups to detect stress.

### Sampling design

The main stress types affecting the study streams were eutrophication/organic pollution and habitat degradation (i.e. alterations of stream morphology). Stress gradients were obtained by preclassifying the streams into five classes of ecological status using data on environmental variables and/or expert opinion:

- 1 High: no or only minimal disturbance;
- 2 Good: slight deviation from high status;
- 3 Moderate: median deviation from high status and significantly more disturbed than good;
- 4 Poor: major alteration from high status;
- 5 Bad: severe alteration from high status.

At least three streams each were sampled for classes 1 and 2 and at least two streams each for the three remaining classes. The preclassification was used only for the sampling design to ensure that a broad gradient was sampled and was replaced later on by stress gradients based on environmental data.

### Sampling

A large number of physical, chemical, hydromorphological and geographical variables were sampled or obtained [e.g. using geographical information system (GIS)] for each of the study streams using standardised site protocols (Hering *et al.*, 2004b). The variables included covered catchment classification of land use/type (obtained from the CORINE land cover system; <http://www.corine.dfd.dlr.de>), hydromorphological (e.g. the number of debris dams and the percentage of the shoreline covered with woody riparian vegetation in a 500 m stretch up- and downstream of the sampling site), habitat variables (e.g. substratum type and nutrient concentration) (Table 2) and variables describing the streams in a more general way (catchment size, distance to source and catchment geology).

Four organism groups (diatoms, macrophytes, benthic macroinvertebrates and fish) were sampled at each site in 2002 and/or 2003 using standardised sampling protocols (Hering *et al.*, 2004b; Furse *et al.*, 2006). Periphytic diatoms were sampled in summer from natural or artificial substrata, such as mineral (sand/silt, pebbles, cobbles and boulders) and vegetative material (macroalgae and macrophytes). Each sample consisted of a composite from more than one type of substratum (cobbles, macrophytes and sand).

If available, a minimum of five cobbles was selected arbitrarily, with a combined exposed surface area of ca 100 cm<sup>2</sup>. The stones were placed individually in a plastic tray and 100–200 mL of distilled or filtered water added to the tray. The upper part of the stone substratum was scrubbed with a toothbrush. The dislodged material was decanted into a sample bottle and preserved using formalin or Lugol's iodine solution. Submerged parts of macrophytes were collected, placed in a wide-mouth 1-L sample bottle, 100–200 mL of distilled/filtered water added, the container was shaken vigorously for about 60 s, and a 250 mL aliquot was decanted to a sample bottle and preserved as above. Sand was sampled by pushing a glass tube into the sediment and extracting sediment and interstitial water. Living and dead diatom cells were identified by light microscopy at 400× and 1000× magnification. For each sample a minimum of 300 diatom valves was counted.

Macrophytes were sampled once in late summer or early autumn. A 100 m stream length was assessed in each stream by wading or walking along the stream bank. All macrophyte species were recorded and the percent cover of the overall macrophyte growth was recorded visually, according to Holmes *et al.* (1999) and national methodologies (Dawson, 2002). Submerged vegetation was observed using a glass-bottom bucket. Species were normally identified in the field but, if identification was uncertain, a representative sample was collected for later identification.

Benthic macroinvertebrates were sampled in spring with the multi-habitat sampling technique described by Hering *et al.* (2004b). Generally, the sampling site covered 20–50 m stream length in the small mountainous streams (10–100 km<sup>2</sup> catchment area) and 50–100 m stream length in medium sized lowland streams (100–1000 km<sup>2</sup> catchment area). Each sampling site encompassed the whole width of the stream and was representative of a minimum area surveyed (i.e. 500 m of length or 100× average width). Multi-habitat samples reflecting the proportion of different microhabitat types present were taken from each stream site. The sampling site was classified according to the coverage of all microhabitats with at least 5% cover. Each sample comprised 20 sample units taken with a hand net (500 µm mesh size) from all microhabitats with >5% coverage. The 20 sample units resulted in about 1.25 m<sup>2</sup> of stream bottom sampled. Each composite sample was preserved with formalin

**Table 2** Physical and chemical variables used for calculating stressor gradients

Parameter	Transformation	Lowlands					Mountains				
		G	P	L	H	M	G	P	L	H	M
Eutrophication/organic pollution											
pH		×	×				×	×			
Conductivity	Log 10	×	×				×	×			
Biological oxygen demand in 5 days (BOD5)	Log 10						×	×			
Oxygen (mg L <sup>-1</sup> )	Log 10		×					×			
Ammonium (mg L <sup>-1</sup> )	Log 10	×	×				×	×			
Nitrite (mg L <sup>-1</sup> )	Log 10						×	×			
Nitrate (mg L <sup>-1</sup> )	Log 10	×	×				×	×			
Sortho-phosphate (µg L <sup>-1</sup> )	Log 10	×	×				×	×			
Total phosphate (µg L <sup>-1</sup> )	Log 10		×					×			
Source pollution (yes/no)								×			
Non-source pollution (yes/no)								×			
Eutrophication (yes/no)								×			
Land use											
Forest catchment (%)	Arcsin sq. root	×					×				
Urban sites catchment (%)	Arcsin sq. root	×		×			×		×		
Natural grassland catchment (%)	Arcsin sq. root	×					×				
Cropland catchment (%)	Arcsin sq. root	×		×			×		×		
Pasture catchment (%)	Arcsin sq. root	×					×				
Hydromorphology (reach scale)											
Shading at zenith (foliage cover)	Arcsin sq. root	×			×		×				×
Width woody rip. vegetation (m)	Arcsin sq. root	×			×		×				×
Number of debris dams		×			×		×				×
Number of logs		×			×		×				×
Shoreline covered with woody riparian vegetation (%)	Arcsin sq. root	×			×		×				×
No bank fixation (%)	Arcsin sq. root	×			×		×				×
No bed fixation (%)	Arcsin sq. root	×			×		×				×
Stagnation (yes/no)		×			×		×				×
Straightening (yes/no)		×			×		×				×
Hydromorphology (microhabitat scale)											
Hygropetric sites (%)	Arcsin sq. root					×					×
Megalithal (stones >40 cm) (%)	Arcsin sq. root					×					×
Macrolithal (stones >20 to 40 cm) (%)	Arcsin sq. root					×					×
Mesolithal (stones >6 to 20 cm) (%)	Arcsin sq. root					×					×
Microolithal (stones >2 to 6 cm) (%)	Arcsin sq. root					×					×
Akal (gravel >0.2 to 2 cm) (%)	Arcsin sq. root					×					×
Psammal/psammopelal (sand) (%)	Arcsin sq. root					×					×
Argyllal (clay) <6µm (%)	Arcsin sq. root					×					×
Macro-algae (%)	Arcsin sq. root					×					×
Micro-algae (%)	Arcsin sq. root					×					×
Submerged macrophytes (%)	Arcsin sq. root					×					×
Emergent macrophytes (%)	Arcsin sq. root					×					×
Living parts of ter. plants (%)	Arcsin sq. root					×					×
Xylal (%)	Arcsin sq. root					×					×
C POM (%)	Arcsin sq. root					×					×
FPOM (%)	Arcsin sq. root					×					×
debris (%)	Arcsin sq. root					×					×

G, general degradation gradient; P, organic pollution/eutrophication gradient; L, land use index; H, hydromorphology (reach scale) gradient; M, hydromorphology (microhabitat scale) gradient. For further explanations compare Furse *et al.* (2006).

(approximately 4% final concentration) or 95% ethanol (approximately 70% final concentration). Macro-invertebrates were sorted (subsampling with the

target of 700 individuals) and identified usually to species/genus. The target number of 700 individuals ensures comparability between samples and is appro-

priate for most metrics (Lorenz, Kirchner & Hering, 2004b).

Fish were sampled by electric fishing [CEN 14011 (CEN, 2003)] within a stop-netted stream section on a single occasion in late summer or early autumn. The recommended sampling area was  $10 \times$  the stream width, with a minimum of a 100 m stream length sampled. The fish were held in plastic tubs until the end of the sampling run after which they were identified to species and returned to the stream. Preliminary population estimates were obtained using the formula of Seber & LeCren (1967) as  $N = c12 / (c1 - c2)$ ;  $c =$  catch). If the proportion caught had a standard error  $>10\%$  then a third run was performed.

#### Definition of stressor gradients

The samples were analysed separately for the lowland and mountain streams, which are very different in terms of community composition (Verdonschot, 2006). Analysis of stream type subgroups was limited to the lowland streams, which are less homogeneous than mountain streams (Verdonschot & Nijboer, 2004; Verdonschot, 2006). To ascertain if the response of organism groups differed between regional, more homogeneous stream types, the lowland streams were subsequently divided into: (i) streams from the UK (stream types U15, U23,  $n = 25$  sites), (ii) streams from Germany (stream type D03) and Denmark (stream type K02)  $n = 21$  sites), (iii) streams from Sweden (stream types S05, S06,  $n = 27$  sites) and (iv) streams from Poland (stream types O02,  $n = 25$  sites).

A number of stress types were included in the STAR sampling protocol: eutrophication/organic pollution and habitat degradation on three different spatial scales (catchment, reach and microhabitat).

Principal components analysis (PCA) of physical-chemical, hydromorphological and land use/type data was used to reduce the dimensionality of the

dataset and construct orthogonal stressor gradients. Four PCA gradients were constructed separately for mountain and lowland streams; the gradients calculated for the lowland stream type group were also used for the subgroups of stream types. (i) general degradation gradients were derived by combining selected physical-chemical, hydromorphological and land use data (Table 2); to prevent the number of parameters exceeding the number of sites, only a subset of those parameters used for the separate stressor gradients was employed for this analysis. In addition, separate stressor gradients were constructed targeting (ii) water chemistry (PCA eutrophication/organic pollution), (iii) hydromorphological degradation (PCA hydromorphology – reach scale) and (iv) alteration of microhabitat composition (PCA hydromorphology – microhabitat scale). Lastly, a land use index was constructed using land use variables, calculated as ‘% urban sites +  $0.5 \times$  % cropland’, assuming that urban sites more strongly affect streams than cropland, while the impact of forest and pasture is minimal (formula derived from Böhrner *et al.*, 2004).

Before constructing the PCA stressor gradients, outlier sites that may have a disproportionate influence on the stress gradients were removed using predefined threshold values (Table 3). For example, sites that were strongly affected by a single stressor A (e.g. eutrophication/organic pollution) were excluded when calculating a gradient for stressor B (e.g. hydromorphological stress). Based on these thresholds and missing data in a few cases, the following numbers of sites were used to calculate the individual stressor gradients: 54 sites for mountain streams/eutrophication; 75 sites for mountain streams/land use; 74 sites for mountain streams/hydromorphology (reach scale); 71 sites for mountain streams/hydromorphology (microhabitat scale); 64 sites for mountain streams/general degradation; 84 sites for lowland

**Table 3** Threshold values for excluding sites from the gradient calculation for another stressor

Stressor	Parameter	Lowlands	Mountains
Eutrophication/organic pollution	Ortho P ( $\mu\text{g L}^{-1}$ )	$\geq 700$	$\geq 600$
Land use	Land use index	$> 40$	$> 30$
Hydromorphology	Hydromorphology index	$< 0$	$< 0$

Land use index = % urban sites +  $0.5 \times$  % cropland.

Hydromorphology index = % shoreline covered with woody riparian vegetation + % no bank fixation + % no bed fixation – (stagnation  $\times 100$ ) – (straightening  $\times 100$ ).

Threshold values taken from Hering *et al.* (2004a).

streams/eutrophication; 86 sites for lowland streams/land use; 78 sites for lowland streams/hydromorphology (reach scale); 70 sites for lowland streams/hydromorphology (microhabitat scale); 98 sites for lowland streams/general degradation.

#### *Metric calculation and selection*

A large number of candidate metrics was calculated for each of the four organism groups. Correlation was used to reduce the number of candidate metrics to a similar number for each organism group. If metric pairs had a Spearman Rank Correlation coefficient of greater than 0.8 or less than -0.8, one of the two metrics was excluded from further analyses (the metric with the lower average Spearman Rank Correlation coefficient to the other candidate metrics was kept). Removal of redundant metrics resulted in 25 to 32 metrics per organism group that were selected for assessing the response of the four organism groups to the stress gradients (Table 4).

#### *Correlation analysis*

Spearman Rank Correlation of metrics and the four PC stressor gradients was used to evaluate the response of the four organism groups to stress. If the first PCA axis of a stressor gradient analysis had an eigenvalue <0.4, correlation was carried out using both the first and second PCA axes; otherwise, only the first PCA axis was used. Each metric was separately correlated to each stressor gradient, at the level of the mountain and lowland streams, respectively, and at the level of the smaller, more homogeneous lowland stream types.

Three statistical parameters were used to evaluate the response of the four organism groups to different types of human-generated stress. To determine if at least a single metric per organism group was sensitive to the targeted stressor, we used the maximum  $r^2$  of all correlations between metrics calculated with the targeted organism group and the stressor gradient. Only metrics significantly correlated with the stressor gradient ( $P < 0.05$ ) were considered (significance was calculated for each correlation analysis separately). To determine if more than one metric of the four organism groups was sensitive to the targeted stressor we used the 75% percentile of the  $r^2$  values of all

correlations significantly indicating the stress gradient. This parameter takes the different number of metrics included into the analysis into account. Finally, to determine if the metrics of the four organism groups respond similarly to stress, we used the percentage of metrics significantly correlating to a stressor gradient, in relation to the total number of metrics selected for the organism group.

#### *Redundancy analysis*

Detrended correspondence analysis (DCA) of the 40 selected metrics (10 for diatoms, macrophytes, macroinvertebrates and fish, see below) gave gradient lengths <3 standard deviations implying that linear model would best fit the data. Consequently, redundancy analysis (RDA), a form of multivariate regression, was used to determine the response of selected metrics to stress gradients (PCA gradients and land use index). In the RDA, metrics were used as response variables and environmental gradients as predictor variables. Moreover, to avoid the number of metrics exceeding the number of sites and to lower the effect of comparing different metric numbers among the organism groups, this analysis was limited to 10 metrics for each organism group. Selection of the 10 metrics was carried out separately for the mountain and lowland stream type groups: the maximum  $r^2$  of all correlation analyses (all stressors, all stream type groups) was calculated for each metric. The 10 metrics with the highest maximum  $r^2$  were selected for each organism group. Both DCA and RDA were run using CANOCO 4.5 (ter Braak & Smilauer, 2002). All sites of the mountain and lowland stream type groups were included in the DCA and RDA analyses.

## **Results**

### *Eutrophication/organic pollution*

In general, all four organism groups responded more strongly to nutrient enrichment than to land use, hydromorphology (reach scale), hydromorphology (microhabitat scale) and general degradation (Table 5). For both the mountain and the lowland stream type groups and for three out of four more specific lowland stream types a diatom metric showed the strongest correlation to the eutrophication



**Table 4** Biological metrics included in the analysis

Code	Description	R	C	S	F	Source
Diatom metrics						
D_IPS	IPS (polluo-sensibilité, Zelinka and Marvan Index modified by Cemagref)			×		CEMAGREF (1982)
D_Slad	Zelinka and Marvan Index modified by Sladeczek			×		Sladeczek (1986)
D_Descy	Zelinka and Marvan Index modified by Descy			×		Descy (1979)
D_L&M	Zelinka and Marvan Index modified by Leclercq and Maquet			×		Leclercq & Maquet (1987)
D_She	Trophic conditions according to Steinberg and Schiefele			×		Steinberg & Schiefele (1988)
D_Wat	Saprobic conditions according to Watanabe			×		Watanabe <i>et al.</i> (1986)
D_TDI	Trophic Diatom Index			×		Kelly <i>et al.</i> (1995)
D_%PT	% Pollution tolerant taxa			×		Kelly <i>et al.</i> (1995)
D_EPI-D	Pollution index based on diatoms			×		Dell'Uomo (1996)
D_Rott(Om)	Rott index calculated with Omnidia			×		Rott <i>et al.</i> (1999)
D_IDG	Generic diatom index			×		Rumeau & Coste (1988)
D_CEE	Community for Economical Community index			×		Descy & Coste (1990)
D_IBD	IBD (Index Biologique Diatomique)			×		Lenoir & Coste (1996)
D_IDAP	IDAP (Index Diatom Artois Picardie)			×		Prygiel <i>et al.</i> (2002)
D_TDI_DVWK	Trophic Diatom Index according to Deutscher Verband für Wasserwirtschaft und Kulturbau			×		DVWK (Deutscher Verband für Wasserwirtschaft und Kulturbau e.V.) (1999)
D_TDI_Rott_G	Trophic Diatom Index according to Rott			×		Rott <i>et al.</i> (1999)
D_TDI_lakes	Trophic Diatom Index for lakes according to Hofmann			×		Hofmann (1994, 1999)
D_DI_Swi	Diatom index Switzerland			×		Hürlimann, Elber & Niederberger (1999)
D_SI_Rott	Saprobic Index Rott			×		Rott <i>et al.</i> (1997)
D_Halo	Halobienindex (targeting salinity)			×		Ziemann (1999)
D_Phylip_DI_1a	PHYLIP multimetric diatom index for German river type 1a (calcareous alpine rivers)		×	×		Schaumburg <i>et al.</i> (2004)
D_Phylip_DI_4	PHYLIP multimetric diatom index for German river type 4 (small silicious mountain streams)		×	×		Schaumburg <i>et al.</i> (2004)
D_Phylip_DI_7b	PHYLIP multimetric diatom index for German river type 7b (small to medium calcareous mountain rivers)		×	×		Schaumburg <i>et al.</i> (2004)
D_Phylip_DI_9	PHYLIP multimetric diatom index for German river type 9 (calcareous lowland rivers)		×	×		Schaumburg <i>et al.</i> (2004)
D_Phylip_RLI	PHYLIP multimetric diatom index for Germany (Red data book index)		×	×		Schaumburg <i>et al.</i> (2004)
Macrophyte metrics						
M_no_mo_li	Number of moss and liverworts species	×				
M_no_subm	Number of submerged species				×	
M_no_anch	Number of species anchored but with floating leaves or heterophyllus				×	
M_no_float	Number of free floating species				×	
M_no_amph	Number of amphibious species				×	
M_no_terr	Number of terrestrial species				×	
M_cov_mo_li	Cover mosses and liverworts		×			

Table 4 (Continued)

Code	Description	R	C	S	F	Source
M_cov_subm	Cover submerged species		×			
M_cov_anch	Cover species anchored but with floating leaves or heterophyllus		×			
M_cov_float	Cover floating free species		×			
M_cov_amph	Cover amphibious species		×			
M_cov_terr	Cover terrestrial species		×			
M_MTR	Mean Trophic Rank			×		Holmes <i>et al.</i> (1999)
M_IBMR	Macrophyte Biological Index for Rivers (IBMR)			×		Haurly <i>et al.</i> (2002)
M_Ellenberg_N	Ellenberg_N			×	×	Ellenberg <i>et al.</i> (1992)
M_Hemeroby index	Hemeroby index			×		Jalas (1955)
M_sp_no	Number of all occurring species	×				
M_ge_no	Number of all occurring genera	×				
M_fa_no	Number of all occurring families	×				
M_Div(SW)	Shannon–Wiener diversity all species	×				Shannon & Weaver (1949)
M_dom	Domination (all species)	×				McNaughton (1967)
M_Ellenberg_N*	Ellenberg_N, typical macrophytes only			×		Ellenberg <i>et al.</i> (1992)
M_ty_sp_no	Number of typical species	×				<a href="http://www.eu-star.at">http://www.eu-star.at</a>
M_ty_dom	Domination (typical species)	×				McNaughton (1967)
M_ty_evenn	Evenness (typical species)	×				Pielou (1966)
M_Ge_sp_no	Number of typical species (German list)	×				<a href="http://www.eu-star.at">http://www.eu-star.at</a>
M_Div(Si)_Ge	Simpson diversity, typical species (German list)	×				Simpson (1949)
M_Ge_dom	Domination (typical species, German list)	×				McNaughton (1967)
M_Ge_evenn	Evenness (typical species, German list)	×				Pielou (1966)
Invertebrate metrics						
I_Acid_Index	Acid Index			×		Henrikson & Medin (1986)
I_ASPT	Average Score Per Taxon			×		Armitage <i>et al.</i> (1983)
I_Div(Marg)	Margalef diversity	×				Margalef (1984)
I_Div(SW)	Shannon–Wiener diversity	×				Shannon & Weaver (1949)
I_DSFI	Danish Stream Fauna Index			×		Skriver, Friberg & Kirkegaard (2001)
I_epirhithral	Epirhithral preferring taxa (%)				×	Hering <i>et al.</i> (2004a)
I_EPT-taxa	Number of Ephemeroptera, Plecoptera and Trichoptera Taxa	×				
I_EPT-taxa (%)	Ephemeroptera, Plecoptera, Trichoptera (%)		×			
I_gatherers	Gatherers (%)				×	Hering <i>et al.</i> (2004a)
I_GFID01	German Fauna Index D01			×		Lorenz <i>et al.</i> (2004a)
I_GFID02	German Fauna Index D02			×		Lorenz <i>et al.</i> (2004a)
I_GFID03	German Fauna Index D03			×		Lorenz <i>et al.</i> (2004a)
I_GFID04	German Fauna Index D04			×		Lorenz <i>et al.</i> (2004a)
I_GFID05	German Fauna Index D05			×		Lorenz <i>et al.</i> (2004a)
I_grazers	Grazers (%)				×	Hering <i>et al.</i> (2004a)
I_IBE_Aqem	Indice Biotico Estesio, AQEM version			×		Ghetti (1997); Buffagni <i>et al.</i> (2004)
I_Index_BR	Index of Biocoenotic Region			×		Hering <i>et al.</i> (2004a)
I_littoral	Littoral preferring taxa (%)				×	Hering <i>et al.</i> (2004a)
I_MAS_IC	Mayfly Average Score Integrity Class			×		Buffagni (1997, 1999)
I_metarhithral	Metarhithral preferring taxa (%)				×	Hering <i>et al.</i> (2004a)
I_no_families	Number of families	×				
I_Oligochaeta	Number of Oligochaeta taxa	×				
I_Oligochaeta (%)	Oligochaeta (%)		×			
I_passive_filt	Passive filterers (%)				×	Hering <i>et al.</i> (2004a)
I_pelal	Pelal preferring taxa (%)				×	Hering <i>et al.</i> (2004a)

Table 4 (Continued)

Code	Description	R	C	S	F	Source
I_Plecoptera (%)	Plecoptera (%)		×			
I_RETI	Rhithron feeding type index				×	Schweder (1992); Podraza, Schuhmacher & Sommerhäuser (2000)
I_rheophile	Rheophile taxa (%)				×	Hering <i>et al.</i> (2004a)
I_shredders	Shredders (%)				×	Hering <i>et al.</i> (2004a)
I_SI(ZM)	Saprobic Index (Zelinka and Marvan)			×		Zelinka & Marvan (1961)
I_Trichoptera (%)	Trichoptera (%)		×			
I_xeno	Xenosaprobic taxa (%)			×	×	Zelinka & Marvan (1961)
Fish metrics						
F_n_sp_tol	Number of tolerant species	×		×		
F_n_sp_intol	Number of intolerant species	×		×		Karr (1981)
F_perc_sp_intol	Native intolerant species (% species)			×		
F_n_ha_hab_wc	Density of species preferring the water column ( $n \text{ ha}^{-1}$ )		×		×	
F_n_sp_hab_b	Number of native benthic species	×			×	
F_perc_sp_hab_b	Native benthic species (% species)		×		×	
F_n_ha_hab_b	Density of native benthic species ( $n \text{ ha}^{-1}$ )		×		×	
F_perc_nha_hab_b	Native benthic species (% individuals of density)		×		×	
F_n_sp_hab_rh	Number of rheophilic species	×			×	Oberdorff <i>et al.</i> (2001)
F_n_ha_hab_rh	Density of rheophilic species ( $n \text{ ha}^{-1}$ )		×		×	
F_perc_sp_hab_li	Native limnophilic species (% species)		×		×	
F_n_ha_hab_li	Density of limnophilic species ( $n \text{ ha}^{-1}$ )		×		×	
F_n_sp_hab_eury	Number of eurytopic species	×			×	
F_n_ha_hab_eury	Density of eurytopic species ( $n \text{ ha}^{-1}$ )		×		×	
F_n_sp_re_lith	Number of lithophilic species	×			×	Oberdorff <i>et al.</i> (2001)
F_perc_nha_re_lith	Lithophilic species (% individuals of density)		×		×	Pont <i>et al.</i> (2006)
F_n_ha_re_phyt	Density of phytophilic species ( $n \text{ ha}^{-1}$ )		×		×	Pont <i>et al.</i> (2006)
F_n_sp_lon_ll	Number of long living species	×			×	
F_perc_sp_lon_ll	Long living species (% species)		×		×	
F_n_sp_lon_sl	Number of short living species	×			×	
F_perc_sp_lon_sl	Short living species (% species)		×		×	
F_n_ha_lon_sl	Density of short living species ( $n \text{ ha}^{-1}$ )		×		×	
F_n_ha_fe_pisc	Density of piscivorous species ( $n \text{ ha}^{-1}$ )		×		×	
F_n_sp_fe_insev	Number of insectivorous species	×			×	
F_n_ha_fe_insev	Density of insectivorous species ( $n \text{ ha}^{-1}$ )		×		×	Pont <i>et al.</i> (2006)
F_n_ha_fe_omni	Density of omnivorous species ( $n \text{ ha}^{-1}$ )		×		×	Pont <i>et al.</i> (2006)
F_n_sp_mi_long	Number of long distance migrating species	×			×	Pont <i>et al.</i> (2006)
F_n_sp_mi_potad	Number of potamodromous species	×			×	Pont <i>et al.</i> (2006)

R, richness/diversity metric; C, composition/abundance metric; S, sensitivity/tolerance metric; F, functional metric.

gradient. Almost all diatom metrics were correlated to the nutrient gradient (85% and 89% of the metrics in the mountain and lowland stream types, respectively), albeit some weakly (Table 5; Appendix). The metric 'Trophic Diatom Index according to Rott' correlated most strongly to eutrophication intensity (stream type S05/S06;  $r^2 = 0.84$ ). Stream type S05/S06

is characterised by the strongest eutrophication gradient. Similar to diatoms, macrophyte metrics showed a consistent response to eutrophication/organic pollution gradients, with the 'best' metrics explaining between 30% and 82% of the variability. The 'number of occurring genera' correlated most strongly to eutrophication intensity (stream type D03/K02;  $r^2 =$

**Table 5** Correlation analyses of metrics and eutrophication/organic pollution gradients. max  $r^2$ : the maximum  $r^2$  of all metrics significantly correlating to a stressor gradient; 75 perc: the 75th percentile of the  $r^2$  values of all metrics significantly correlating to a stressor gradient; share sg: the share of metrics significantly correlating to a stressor gradient, in relation to the total number of metrics used for the organism group.

	No sites	Diatoms			Macrophytes			Invertebrates			Fish														
		max $r^2$	75 perc	share sg	max $r^2$	75 perc	share sg	max $r^2$	75 perc	share sg	max $r^2$	75 perc	share sg												
All mountain streams	87	●	0.50	●	0.35	●	85.2	●	0.45	●	0.27	●	17.2	●	0.49	●	0.30	●	90.6	●	0.31	●	0.14	●	60.7
All lowland streams	98	●	0.67	●	0.59	●	88.9	●	0.30	●	0.20	●	20.7	●	0.24	●	0.11	●	59.4	●	0.22	●	0.10	●	21.4
Lowland streams U15 U23	25	●	0.73	●	0.47	●	81.5	●	0.40	●	0.30	●	6.9	●	0.37	●	0.30	●	53.1	●	0.58	●	0.47	●	32.4
Lowland streams D03 K02	21	●	0.75	●	0.41	●	37.0	●	0.82	●	0.78	●	55.2	●	0.37	●	0.35	●	6.3	●	0.69	●	0.42	●	17.9
Lowland streams S05 S06	27	●	0.84	●	0.73	●	77.8	●	0.73	●	0.62	●	51.7	●	0.72	●	0.45	●	28.1	●	0.41	●	0.25	●	17.9
Lowland streams O02	25	●	0.24	●	0.24	●	3.7	●	0.45	●	0.42	●	13.8	●	0.47	●	0.44	●	18.8	●	0.45	●	0.35	●	14.3

**Correlation strength**      **% of metrics correlating**

$r^2 = 0.2-0.4$	●	●	20-40%
$r^2 < 0.6$	●	●	>40-60%
$r^2 < 0.8$	●	●	>60-80%
$r^2 < 1.0$	●	●	>80-100%

0.82). The share of significantly correlating metrics was <25% for the stream groups (all mountain streams, all lowland streams), while for some more homogeneous stream types (e.g. lowland streams D03/K02) the share was >50%, indicating that different metrics best reflect the eutrophication/organic pollution gradients in the individual stream types.

For mountain streams macroinvertebrate metrics responded more strongly to the eutrophication/organic pollution gradient than for all lowland streams. Conversely, for stream types S05/S06 and O02 some metrics were strongly correlated [average score per taxon (ASPT) in S05/S06:  $r^2 = 0.72$ ; the overall strongest correlation for invertebrate metrics and eutrophication/organic pollution intensity]. Similar to macrophytes (but not diatoms) this indicates the lack of generally applicable metrics (i.e. most metrics are best suited for individual stream types).

For mountain streams, fish metrics responded poorly to the eutrophication/organic pollution gradients, whilst in the individual lowland stream types one or more fish metrics explained from 40% to 70% of the variability, although none of the metrics were found to be applicable for the entire lowland stream type group. The metric 'density of native benthic species' correlated most strongly to the eutrophic-

ation/organic pollution gradient (stream type D03/K02;  $r^2 = 0.69$ ).

#### Land use

All organism groups responded less strongly to catchment land use than to eutrophication/organic pollution gradients (Table 6; Appendix). Most diatom metrics reflected catchment land use to some degree (>80% of the metrics tested for both all mountain and all lowland streams), but correlation coefficients were weak (maximum  $r^2 < 0.4$  in four of the six stream groups). The strongest correlation was found for the metric 'Rott index calculated with Omnidia' (stream type S05/S06;  $r^2 = 0.81$ ). At least one macrophyte metric correlated significantly to land use gradients in all stream type groups, although in five of the six stream groups the maximum  $r^2$  was <0.4. The 'mean trophic rank (MTR)' metric correlated most strongly (stream types S05/S06:  $r^2 = 0.58$ ), followed by 'Ellenberg\_N' (all lowland streams:  $r^2 = 0.25$ ). The majority (84%) of the macroinvertebrate metrics were significantly correlated to catchment land use in the mountain streams, albeit mostly weakly. For the entire lowland stream group no metrics were found to be significantly correlated to catchment land use with  $r^2 > 0.2$ , but at the individual stream type level several metrics were more strongly related to the catchment

**Table 6** Correlation analyses of metrics and land use gradients. See Table 5 for details.

	No sites	Diatoms			Macrophytes			Invertebrates			Fish		
		max $r^2$	75 perc	share sg	max $r^2$	75 perc	share sg	max $r^2$	75 perc	share sg	max $r^2$	75 perc	share sg
All mountain streams	87	• 0.37	• 0.24	● 85.2	• 0.36	• 0.20	● 62.1	• 0.31	• 0.23	● 84.4	0.16	0.09	● 46.4
All lowland streams	98	• 0.38	• 0.25	● 88.9	• 0.25	• 0.15	• 27.6	0.12	0.10	● 43.8	0.18	0.15	• 25.0
Lowland streams U15 U23	25	0.00	0.00	0.00	• 0.25	0.19	17.2	0.00	0.00	0.00	• 0.20	0.19	14.3
Lowland streams D03 K02	21	• 0.30	• 0.24	• 37.0	• 0.21	• 0.20	10.3	• 0.39	• 0.35	12.5	● 0.40	● 0.40	17.9
Lowland streams S05 S06	27	● 0.81	● 0.62	● 70.4	● 0.58	● 0.46	● 48.3	● 0.66	• 0.38	• 21.9	● 0.40	• 0.22	• 28.6
Lowland streams O02	25	● 0.56	● 0.40	● 70.4	• 0.38	• 0.38	13.8	● 0.44	● 0.41	• 31.3	• 0.20	• 0.20	3.6

land use gradient: 'ASPT' (stream types S05/S06,  $r^2 = 0.66$ ; stream types D03/K02,  $r^2 = 0.39$ ) and 'Plecoptera (%)' (stream type O02,  $r^2 = 0.44$ ). Fish metrics were weakly correlated to land use in mountain streams (all correlations with  $r^2 < 0.2$ ), while in lowland stream types the explanatory power of the fish metrics was similar to that found for invertebrate metrics. The strongest correlations were for the metrics 'number of lithophilic species' (stream types S05/S06,  $r^2 = 0.4$ ) and 'density of limnophilic species ( $n \text{ ha}^{-1}$ )' (stream types D03/K02,  $r^2 = 0.4$ ).

#### Hydromorphology (reach scale)

The impact of hydromorphological degradation (reach scale) on the biological metrics tested here was generally less obvious than the impact of eutrophication/organic pollution and land use (Table 7; Appendix). Diatoms showed almost no response to hydromorphological degradation on the reach scale. The metric 'IPS' (polluo-sensibilité) correlated most strongly (stream type O02;  $r^2 = 0.41$ ). Similarly, the response of macrophyte metrics to hydromorphological gradients was weak. In mountain streams, the

best response was found for the metric 'number of submerged species' ( $r^2 = 0.25$ ), while in the lowland streams the response was restricted to single stream types. Overall, the strongest correlation was found for the metric 'evenness (typical species)' (stream type D03/K02;  $r^2 = 0.4$ ).

In contrast to benthic diatoms and macrophytes, macroinvertebrates responded more strongly to hydromorphological (reach scale) gradients. In all stream types at least one metric was significantly correlated to this stress gradient, with a maximum  $r^2$  of 0.51 ('German Fauna Index D01' in stream type O02). In particular the metrics 'Ephemeroptera, Plecoptera and Trichoptera taxa (EPT)' and the 'German Fauna Indices' responded strongly. By contrast, the response of fish metrics to hydromorphological gradients was restricted to lowland streams, with the metric 'density of native benthic species ( $n \text{ ha}^{-1}$ )' correlated most strongly (stream type O02,  $r^2 = 0.46$ ) to impairment.

#### Hydromorphology (microhabitat scale)

Diatom metrics responded to alterations in microhabitat composition; however, responses were generally

**Table 7** Correlation analyses of metrics and hydromorphological (reach scale) gradients. See Table 5 for details.

	No sites	Diatoms			Macrophytes			Invertebrates			Fish		
		max $r^2$	75 perc	share sg	max $r^2$	75 perc	share sg	max $r^2$	75 perc	share sg	max $r^2$	75 perc	share sg
All mountain streams	87	0.09	0.07	12.1	• 0.25	0.19	● 48.2	• 0.23	0.13	● 50.0	0.13	0.12	• 28.6
All lowland streams	98	0.00	0.00	0.00	0.12	0.10	● 44.8	• 0.26	0.16	● 43.8	0.17	0.15	● 46.4
Lowland streams U15 U23	25	0.00	0.00	0.00	0.00	0.00	0.00	• 0.35	• 0.35	3.1	0.00	0.00	0.00
Lowland streams D03 K02	21	0.00	0.00	0.00	0.40	• 0.38	• 27.6	● 0.45	• 0.38	• 28.1	● 0.42	• 0.37	• 21.4
Lowland streams S05 S06	27	0.00	0.00	0.00	• 0.20	0.19	10.3	• 0.20	• 0.20	6.3	• 0.24	• 0.20	• 28.6
Lowland streams O02	25	● 0.41	• 0.25	• 25.9	• 0.30	• 0.27	6.9	● 0.51	● 0.41	• 37.5	● 0.46	• 0.35	• 28.6

**Table 8** Correlation analyses of metrics and hydromorphological (microhabitat scale) gradients. See Table 5 for details.

	No sites	Diatoms			Macrophytes			Invertebrates			Fish		
		max $r^2$	75 perc	share sg	max $r^2$	75 perc	share sg	max $r^2$	75 perc	share sg	max $r^2$	75 perc	share sg
All mountain streams	87	• 0.20	0.14	● 51.9	• 0.20	0.16	• 31.0	• 0.33	0.18	● 62.5	0.16	0.14	• 32.1
All lowland streams	98	• 0.36	• 0.27	● 47.1	• 0.31	• 0.23	● 72.4	• 0.38	• 0.21	• 37.5	0.19	0.12	• 35.7
Lowland streams U15 U23	25	0.76	● 0.49	● 55.6	● 0.70	● 0.65	• 24.1	• 0.35	• 0.35	3.1	0.00	0.00	0.0
Lowland streams D03 K02	21	0.00	0.00	0.0	0.00	0.00	0.0	0.62	● 0.45	12.5	● 0.64	● 0.44	• 21.4
Lowland streams S05 S06	27	0.18	0.18	3.7	● 0.41	• 0.34	• 20.7	• 0.30	• 0.28	18.8	• 0.29	• 0.27	• 21.4
Lowland streams O02	25	• 0.27	• 0.27	3.7	• 0.24	• 0.24	6.9	• 0.33	• 0.31	• 34.4	● 0.45	● 0.40	7.1

weak and differed among stream types (Table 8; Appendix). The strongest correlation was observed for the metric 'PHYLIP DI\_4' (stream type U15/U23;  $r^2 = 0.76$ ). In general, macrophyte metrics showed a stronger response to hydromorphology (microhabitat scale) gradients than benthic diatoms, but the response was mainly observed in stream types rather than stream groups. For example, in the British stream type (U15/U23) Shannon diversity was correlated to microhabitat diversity ( $r^2 = 0.7$ ; strongest correlation observed).

The correlation patterns of macroinvertebrates were more consistent across the stream types. In all but one stream type (D03/K02) at least one metric was significantly correlated to the hydromorphology (microhabitat scale) gradient; in most cases zonation measures, e.g. 'epirhithral (%)' (for example, stream types D03/K02,  $r^2 = 0.62$ ; all lowland streams,  $r^2 = 0.38$ ), or 'metarhithral (%)' (for example, all mountain streams,  $r^2 = 0.33$ ) were best correlated with the impairment gradient. As with reach-scale alterations in hydromorphology fish metrics were mainly correlated to the impairment gradient in lowland streams,

e.g. 'native benthic species (% individuals of density)' (stream types D03/K02,  $r^2 = 0.64$ , the strongest observed correlation).

#### General degradation

The general degradation gradient included parameters describing eutrophication/organic pollution, catchment land use and hydromorphological stress. Diatom metrics had lower correlation coefficients when responses were compared between the general degradation gradient and the nutrient enrichment gradient (Table 9; Appendix; e.g. maximum  $r^2 = 0.14$  versus maximum  $r^2 = 0.5$  for general degradation and nutrient enrichment, respectively, in mountain streams and maximum  $r^2 = 0.34$  versus maximum  $r^2 = 0.67$  in the lowland stream group). This finding implies that the relatively strong response of diatoms to eutrophication was weakened when the hydromorphological parameters were included in the gradient analysis. The 'Halobienindex' correlated most strongly to general degradation gradients (stream type S05/S06;  $r^2 = 0.69$ ). At least one macrophyte

**Table 9** Correlation analyses of metrics and general degradation. See Table 5 for details.

	No sites	Diatoms			Macrophytes			Invertebrates			Fish		
		max $r^2$	75 perc	share sg	max $r^2$	75 perc	share sg	max $r^2$	75 perc	share sg	max $r^2$	75 perc	share sg
All mountain streams	87	0.12	0.06	• 22.2	• 0.25	• 0.21	● 69.0	• 0.29	0.14	● 62.5	0.12	0.07	17.9
All lowland streams	98	• 0.34	• 0.29	● 88.9	• 0.21	0.17	● 82.8	• 0.25	0.16	● 56.3	• 0.29	● 0.25	● 64.3
Lowland streams U15 U23	25	0.18	0.18	• 22.2	• 0.28	• 0.26	6.9	• 0.25	• 0.22	9.4	• 0.26	● 0.26	14.3
Lowland streams D03 K02	21	• 0.21	• 0.21	3.7	● 0.42	• 0.39	● 48.3	● 0.52	● 0.49	• 25.0	● 0.53	• 0.38	● 42.9
Lowland streams S05 S06	27	● 0.69	● 0.52	● 74.1	● 0.57	• 0.59	• 20.7	● 0.44	● 0.43	15.6	0.16	● 0.16	3.6
Lowland streams O02	25	● 0.54	• 0.34	● 66.7	• 0.32	• 0.27	• 31.0	● 0.56	● 0.49	● 53.1	• 0.38	0.30	• 35.8

metric correlated to the general degradation gradient with  $r^2 > 0.2$  in all stream types. The metrics 'MTR' and 'Macrophyte Biological Index for Rivers (IBMR)' correlated most strongly (stream type S05/S06;  $r^2 = 0.57$ ).

Benthic macroinvertebrates responded strongly to the general degradation gradient (at least one metric with  $r^2 \geq 0.25$ ) as several metrics were correlated to both eutrophication/organic pollution and hydromorphological stress. In general, the same metrics that were found to be best correlated with hydromorphological degradation were also best suited to assess general degradation. In the mountain streams the share of xenosaprobic (=ultraoligotrophic) taxa ( $r^2 = 0.29$  in all mountain streams) and in the lowland streams the German Fauna Indices, the EPT-taxa and the share of Plecoptera ['EPT-Taxa' (%)] in stream type O02:  $r^2 = 0.56$ ; 'Plecoptera (%)' in stream type D03/K02:  $r^2 = 0.52$ ; 'Plecoptera (%)' in stream type S05/S06:  $r^2 = 0.44$ ) were most strongly correlated with the general degradation gradient.

Fish metrics were less strongly correlated to general degradation gradients in mountain than in lowland streams. Strongly correlating fish metrics were 'density of eurytopic species ( $n \text{ ha}^{-1}$ )' (stream type D03/K02,  $r^2 = 0.53$ ) and 'native benthic species (% individuals of density)' (stream type O02,  $r^2 = 0.38$ ).

#### Redundancy analysis

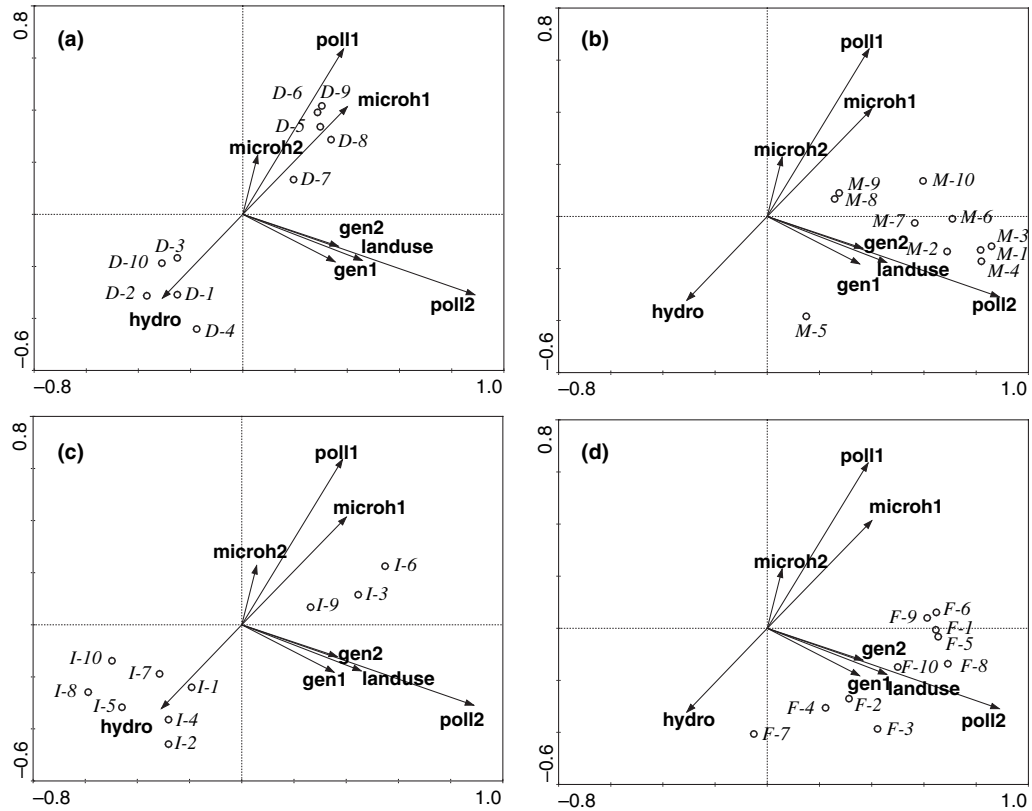
*Mountain streams.* All diatom metrics responded similarly to the stress gradients (Fig. 2). The two different metric groups in Fig. 2a show that some metrics are positively, whereas others are negatively correlated to the eutrophication/organic pollution gradient (poll1) and the hydromorphological gradient (hydro), depending on whether metric results decrease or increase with stress. All macrophyte metrics responded similarly but their response was different compared with diatom metrics. Macrophyte metrics were strongly correlated to the land-use gradient and to the second PC eutrophication/organic pollution (poll2; mainly determined by ammonium concentration) gradient, although the relation to the main eutrophication/organic pollution gradient [poll1; mainly determined by total phosphorus concentration and biological oxygen demand in 5 days (BOD5)] was weaker. Invertebrate metrics covered a comparatively wide range of stress gradients, being correlated to

both the hydromorphological gradient (which was negatively correlated to the eutrophication/organic pollution gradient poll1) and to the land use gradient and the second PC eutrophication/organic pollution (poll2). Fish metrics gave mainly the same information as the macrophytes metrics and were best correlated with the second PC eutrophication/organic pollution (poll2) and general degradation gradients.

*Lowland streams.* As in the mountain streams, all diatom metrics responded similarly and were mainly correlated to the eutrophication/organic pollution gradient (poll; Fig. 3a). The macrophyte metrics formed a tight cluster to the left in the ordination and were seemingly negatively correlated with the general degradation gradients (gen1 and gen2) and the eutrophication/organic pollution gradient (poll). Only two metrics ('MTR' and 'IBMR') showed some relation to the eutrophication/organic pollution gradient. Similar to the mountain streams, invertebrate metrics in lowland streams were mainly correlated with the hydromorphological and eutrophication/organic pollution gradients. Fish metrics were dispersed across the ordination and individual metrics were correlated to all types of stress.

#### Discussion

In many cases organism groups and metrics responded predominantly to specific stressors, although this specificity was often obscured by different stressors acting at the same site, particularly in the RDA analysis. By excluding sites strongly affected by a second stressor, we were better able to isolate indicator response along 'single' stress gradients. Nonetheless, all organism groups responded most strongly to the eutrophication/organic pollution gradient. The diatom metrics tested here often detected eutrophication effects better than metrics calculated using the other three organism groups. This finding supports our conjecture that both nutrient enrichment (eutrophication) and organic inputs are correlated in our data set, but that the major changes are because of nutrient addition and not organic pollution and the subsequent decrease in oxygen concentration. Of the four organism groups/metrics tested here, diatoms were best suited for assessing the effects of nutrient enrichment in lowland streams, while both diatoms and macroinverte-



**Fig. 2** Redundancy analysis of metrics and environmental parameters (mountain streams). All four subfigures are related to the same analysis but only those metrics calculated with a certain organism group are displayed in each subfigure. Gradients: gen1 = 1st PCA axis general degradation; gen2 = 2nd PCA axis general degradation; hydro = 1st PCA axis hydromorphological degradation (reach scale); land use = land use index; microh1 = 1st PCA axis hydromorphological degradation (microhabitat scale); microh2 = 2nd PCA axis hydromorphological degradation (microhabitat scale); poll1 = 1st PCA axis eutrophication/organic pollution; poll2 = 2nd PCA axis eutrophication/organic pollution (compare Table 10). (a) diatom metrics; (b) macrophytes metrics; (c) invertebrate metrics; (d) fish metrics.

No	+/-	Abbreviation (see Table 4)
D-1	-	IPS
D-2	-	EPI-D
D-3	-	ROTT
D-4	-	CEE
D-5	+	TDI DVWK
D-6	+	TDI Rott
D-7	+	TDI Seen Hofmann
D-8	+	Saprobienindex Rott
D-9	+	Halobienindex
D-10	-	PHYLIP DI_Typ4
M-1	+ or -	n_sp_subm
M-2	+ or -	n_sp_floating
M-3	+ or -	n_sp_amphi
M-4	+ or -	n_sp_terr
M-5	+ or -	cover_moss_liv
M-6	+ or -	cover_sp_subm
M-7	+ or -	cover_sp_amphi
M-8	-	MTR
M-9	-	IBMR
M-10	+	Ellenberg_N*

No	+/-	Abbreviation (see Table 4)
I-1	-	ASPT
I-2	-	EPT-Taxa (%)
I-3	+	gatherers
I-4	-	GFID01
I-5	-	GFID05
I-6	+	MAS_IC
I-7	-	metarhithral
I-8	-	Plecoptera (%)
I-9	+	SI(ZM)
I-10	-	xeno
F-1	+	n_sp_Tol
F-2	-	n_ha_Hab_wc
F-3	-	n_sp_Hab_rh
F-4	-	n_ha_Hab_rh
F-5	+	n_sp_Hab_eury
F-6	+	n_ha_Hab_eury
F-7	-	perc_nha_Re_lith
F-8	-	n_sp_Lon_ll
F-9	+	n_ha_Fe_omni
F-10	-	n_sp_Mi_potad



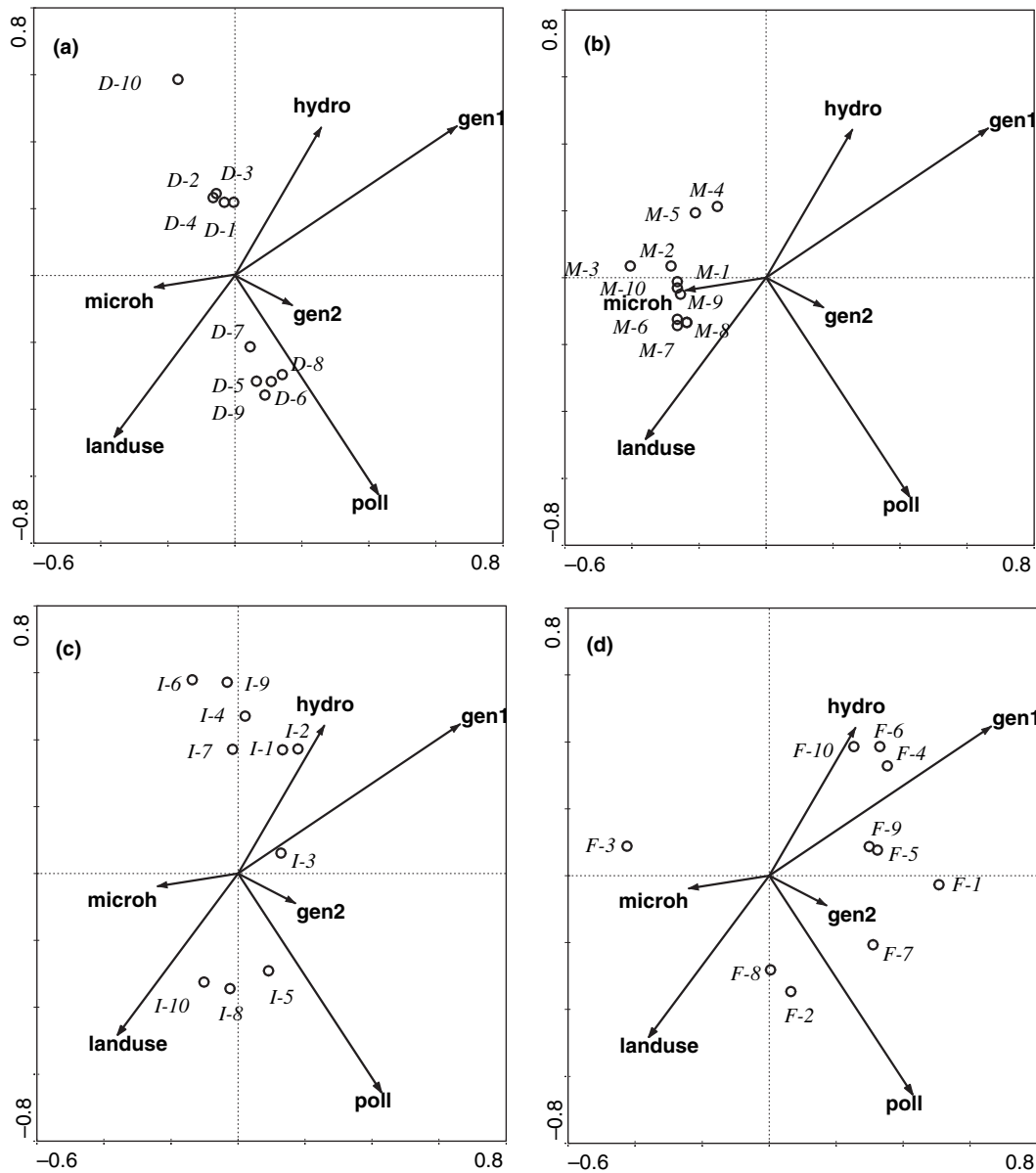


Fig. 3 RDA of metrics and environmental parameters (lowland streams). All four subfigures are related to the same analysis but only those metrics calculated with a certain organism group are displayed in each subfigure. (a) diatom metrics; (b) macrophytes metrics; (c) invertebrate metrics; (d) fish metrics. Gradients: see Fig. 2 and Table 10 for figure legend.

No	+/-	Abbreviation (see Table 4)	No	+/-	Abbreviation (see Table 4)
D-1	-	SHE	I-1	-	ASPT
D-2	-	EPI-D	I-2	-	DSFI
D-3	-	ROTT	I-3	-	epirhithral
D-4	-	IDG	I-4	-	EPT-Taxa (%)
D-5	+	TDI DVWK	I-5	+	gatherers
D-6	+	TDI Rott	I-6	-	GFID01
D-7	+	Diatomeenindex Schweiz	I-7	-	GFID05
D-8	+	Saprobienindex Rott	I-8	+	pelal
D-9	+	Halobienindex	I-9	-	Plecoptera (%)
D-10	?	PHYLIP DI_Typ4	I-10	+	SI(ZM)
M-1	+ or -	n_sp_floating	F-1	-	n_ha_Hab_b

Fig. 3 Caption (Continued)

No	+/-	Abbreviation (see Table 4)	No	+/-	Abbreviation (see Table 4)
M-2	+ or -	n_sp_amphi	F-2	+	n_ha_Hab_eury
M-3	+ or -	cover_sp_amphi	F-3	+	n_ha_Re_phyt
M-4	-	MTR	F-4	-	n_sp_Fe_insev
M-5	-	IBMR	F-5	-	n_sp_Hab_b
M-6	-	Species number (S)	F-6	-	n_sp_Intol
M-7	-	Genus number (G)	F-7	-	n_sp_Mi_long
M-8	-	Family number (F)	F-8	+	n_sp_Tol
M-9	-	Species number (S*)	F-9	-	perc_nha_Hab_b
M-10	-	Species number (S**)	F-10	-	perc_sp_Intol

brates seem to perform equally well in mountain streams. While diatoms and macrophytes are likely to respond to eutrophication, macroinvertebrates and fish are presumably most strongly affected by oxygen depletion following organic pollution. These findings were not unexpected, as several studies have both conceptually and empirically showed the utility of benthic algae (e.g. Kelly *et al.*, 1995; Rott *et al.*, 1997, 1999; Coring, 1999) and invertebrates (e.g. Zelinka & Marvan, 1961; Armitage *et al.*, 1983) in monitoring the effects of eutrophication in streams.

In contrast to nutrient enrichment effects, the land use stressor potentially affected the community through numerous cause-effect relationships acting singly or in concert. For example, sedimentation and hydromorphological alteration, diffuse pollution as well as direct inputs of toxic substances (metals, pesticides) may all be related to land use. Our results showed that the organism groups responded less strongly to land use gradients than to nutrient enrichment. We found that diatoms responded most strongly to the land use gradient, followed by macroinvertebrates and macrophytes and fish, which may

be a reflection of life histories. Probably, different pathways are responsible for land use effects on the individual organism groups. For example, fish metrics that showed the strongest response were often related to direct or indirect effects on habitat quality [e.g. 'number of lithophilic species' and 'density of limnophilic species ( $n \text{ ha}^{-1}$ )']. Conversely, the response of benthic invertebrate metrics reflected changes in certain organism groups (Plecoptera), which might reflect water quality, while both benthic diatom and macrophyte metrics were seemingly related to the effects of nutrient addition.

Responses to hydromorphological degradation on the reach scale revealed clear differences among the organism groups studied here. Macroinvertebrates responded most strongly to this gradient, followed by fishes and macrophytes, while diatoms did not respond to hydromorphological changes. At the level of individual stream types the response to hydromorphological degradation was often much stronger. The finding that benthic macroinvertebrates respond to changes in hydromorphology (reach scale) supports a number of earlier studies (e.g. Buffagni *et al.*, 2004;

**Table 10** Eigenvalues of the PCA axes 1–4 stressor gradients for lowland and mountain streams. Bold: axes used in the analysis (for these axes the short codes used in Figs 2 & 3 are given). The eigenvalues are also dependent on the size of the data sets.

Group	Stressor	Number of sites	Axis 1	Axis 2	Axis 3	Axis 4
Mountains	General degradation	76	<b>0.2577 (gen1)</b>	<b>0.0408 (gen2)</b>	0.0142	0.0090
Mountains	Eutrophication/organic pollution	66	<b>0.3336 (poll1)</b>	<b>0.2638 (poll2)</b>	0.1533	0.0966
Mountains	Hydromorphology (reach scale)	74	<b>0.4954 (hydro)</b>	0.2033	0.1009	0.0694
Mountains	Land use	75	<b>no PCA gradient used; land use index (landuse)</b>			
Mountains	Hydromorphology (microhabitat scale)	71	<b>0.2836 (microh1)</b>	<b>0.1819 (microh2)</b>	0.1467	0.1044
Lowlands	General degradation	98	<b>0.2869 (gen1)</b>	<b>0.2413 (gen2)</b>	0.1622	0.0692
Lowlands	Eutrophication/organic pollution	87	<b>0.5273 (poll)</b>	0.3595	0.0558	0.0323
Lowlands	Hydromorphology (reach scale)	79	<b>0.5341 (hydro)</b>	0.1788	0.1165	0.0534
Lowlands	Land use	86	<b>no PCA gradient used; land use index (landuse)</b>			
Lowlands	Hydromorphology (microhabitat scale)	75	<b>0.4654 (microh)</b>	0.1274	0.1060	0.0810

Hering *et al.*, 2004b; Lorenz *et al.*, 2004a), and is likely to be a result of both quantitative and qualitative changes in meso- and microhabitat composition. For example, in more site-intensive studies a strong correlation of macrophyte response to hydromorphological gradients has been observed (Baatrup-Pedersen & Riis, 1999; Bernez *et al.*, 2004; Schaumburg *et al.*, 2004), although several aspects of relationship between aquatic plants and stream structural quality are still under discussion (Passauer *et al.*, 2002). Particularly for the small mountain streams such correlations may be obscured by shading (Vermaat & Debruyne, 2003). Excluding the heavily shaded sites from the dataset could reveal much stronger correlations of macrophytes and hydromorphological features. Several studies reported a strong response of fish to hydromorphological changes on the reach scale (Gorman & Karr, 1978; Bain *et al.*, 1988); the comparatively weak response in our dataset may be because of (i) typological differences of the investigated streams, which obscure the relationships and/or (ii) how the hydromorphological stress gradient was defined: meso-scale variables might have resulted in much stronger relationships.

Constrained ordination of organism group/metrics and stress gradients largely supported the findings that were revealed with correlation. However, there were important differences between these two analyses: (i) sites affected by a second stressor were not excluded in the RDA, and thus, e.g. hydromorphology (reach scale) and eutrophication gradients were correlated in the RDA; (ii) only roughly one-third of the metrics used in correlation were used in RDA, and thus some metrics responding to specific stressors were excluded. As shown with correlation, diatom metrics responded strongly to eutrophication/organic pollution gradients in both the mountain and the lowland stream groups. The finding that diatom metrics were also correlated to hydromorphology (reach scale) gradients is probably because of these two gradients being correlated, because sites affected by both stressors have not been removed. Ordination showed that invertebrate metrics covered a wider range of stress types than diatom metrics. Nonetheless, as with diatoms, invertebrate metrics were mainly correlated to the combined eutrophication/hydromorphology gradients in both the mountain and lowland stream groups. This correlation implies that both benthic diatoms and macroinvertebrates

responded to similar gradients, and hence may provide redundant information. This interpretation, however, was not supported from the correlation results and is most likely to be an effect of the inclusion of sites that are affected by more than one stressor. Macrophyte and diatom metrics were also shown to respond differently, indicating that these two organism groups were supplying independent information. The different response of macrophytes and diatoms may be partly explained by the poorer correlation of macrophyte metrics to the main stressor gradients (i.e. nutrient enrichment). In contrast, in mountain streams fish and macrophyte metrics seemed to be correlated, which might be an artefact of the dataset, namely both groups were poorly represented (low taxonomic richness) in mountain streams. In lowland streams, fish metrics covered an even larger range than invertebrate metrics, despite relatively species-poor communities. These findings indicate that diatom/macroinvertebrate metrics are mainly related to the dominant eutrophication/hydromorphology gradient in the data set, while fish and macrophytes were responding more to general types of degradation.

In designing biomonitoring programmes, consideration should be given to the stream type being addressed, the types of stressors potentially affecting the integrity of the stream ecosystem and the time frame of the study (including knowledge of interannual variability and potential lag-phase responses of degradation and recovery; e.g. Stevenson *et al.*, 2004). By combining conceptual models (expert opinion) and empirical data, more cost-effective monitoring programmes incorporating knowledge of how different organism groups react to different human-generated stressors, can be designed (e.g. USEPA, 2000). For example, as the response of the four organism groups addressed in this paper are partly correlated (i.e. redundant) it is not necessary to monitor all groups simultaneously. From our study, the following more general recommendations can be derived for European bioassessment of streams. In small Central European mountain streams, benthic diatoms and macroinvertebrates are the most diverse organism groups and best reflect the main stress gradients. Fish assemblages are usually species-poor and, therefore, assemblage-based metrics tested here may have a limited capacity to detect stressors. Species-based metrics, e.g. trout biomass, proportion of juveniles,

might show stronger responses to stressors and should be tested in future work. Further, macrophytes are often patchily distributed and, thus, less suitable for monitoring purposes. In medium-sized lowland streams in Central and Northern Europe all four of the organism groups are, in principle, suitable for monitoring. The selection of indicator(s) depends on the stressor-type being assessed and the monitoring type. For the effects of land use and eutrophication all organism groups (fish, benthic macroinvertebrates, benthic diatoms and macrophytes) are, in principal, well suited.

Although the effects of eutrophication (nutrient enrichment) and organic pollution (e.g. increased BOD) are of different origin, they are often correlated and, thus, similar indicators can be used in most cases to detect both types of stressors. All four organism groups studied here (fish, benthic macroinvertebrates, benthic diatoms and macrophytes) respond to eutrophication/organic pollution and are, in principle, suitable as indicators. However, the rates and trajectories of change may vary among the organism groups. For example, benthic diatoms often show a stronger response (high sensitivity) and low error (high precision) compared with the other three organism groups (Johnson *et al.*, 2006a). Hence, benthic diatoms may be best suited for situations in which only eutrophication/organic pollution is assessed. If the focus of the study is on nutrient enrichment, benthic diatoms and/or macrophytes should be considered, as nutrient enrichment may be the main factor directly affecting both groups. If the focus of the study is on organic pollution, benthic macroinvertebrates and/or fish should be considered, as these groups are more directly affected by oxygen condition. Land-use affects stream communities by altering, for example, nutrients (eutrophication), habitat quality (sedimentation) and toxicity (e.g. pesticides), and these effects are most strongly reflected by fish, benthic invertebrates and benthic diatoms. With the exception of diatoms, all organism groups (fish, benthic macroinvertebrates and macrophytes) respond to hydromorphological degradation and the selection of the most appropriate organism group depends on stream type. In lowland streams and in medium-sized to large rivers all three groups can be considered. The relatively species-poor fish and macrophyte assemblages of small streams may limit the use of these two organism groups and hence benthic invertebrates should be considered for

monitoring the effects of hydromorphological degradation at the reach scale (see also Johnson *et al.*, 2006b). For meso-scale hydromorphological variables fish are usually considered as the most appropriate option (Bain *et al.*, 1988), while for effects at smaller (habitat) spatial scales benthic invertebrates should be considered, based on our results. If multiple stressors are assessed then benthic invertebrates and/or macrophytes should be considered. Use of macroinvertebrates in such conditions is recommended by Dolédec *et al.* (1999) and Statzner *et al.* (2001), as functional composition approaches enable the discrimination of different disturbance types because of well known species traits of benthic invertebrates.

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### References

- Armitage P.D., Moss D., Wright J.F. & Furse M.T. (1983) The performance of a new biological water quality score system based on macroinvertebrates over a wide range of unpolluted running-water sites. *Water Research*, **17**, 333–347.
- Baatrup-Pedersen A. & Riis T. (1999) Macrophyte diversity and composition in relation to substratum characteristics in regulated and unregulated Danish streams. *Freshwater Biology*, **42**, 375–385.
- Bain M.B., Finn J.T. & Booke H.E. (1988) Streamflow regulation and fish community structure. *Ecology*, **69**, 382–392.
- Barbour M.T., Gerritsen J., Snyder B.D. & Stribling J.B. (1998) *Rapid Bioassessment Protocols for Use in Streams and Wadeable Rivers: Periphyton, Benthic Macroinvertebrates and Fish* (2nd edn). EPA/841/B/98-010 U.S. Environmental Protection Agency Office of Water, Washington D.C.

- Battarbee R.W., Flower R.J., Juggins S., Patrick S.T. & Stevenson A.C. (1997) The relationship between diatoms and surface water quality in the Hoylandet area of Nord-Trøndelag, Norway. *Hydrobiologia*, **348**, 69–80.
- Belpaire C., Smolders R., Auweele I.V., Ercken D., Breine J., Van Thuyne G. & Ollevier F. (2000) An Index of Biotic Integrity characterizing fish populations and ecological quality of Flandrian water bodies. *Hydrobiologia*, **434**, 17–33.
- Bernez I., Daniel H., Haury J. & Ferreira M.T. (2004) Combined effects of environmental factors and regulation on macrophyte vegetation along three rivers in Western France. *River Research and Applications*, **20**, 43–59.
- Böhmer J., Rawer-Jost C., Zenker A., Meier C., Feld C.K., Biss R. & Hering D. (2004) Assessing streams in Germany with benthic invertebrates: development of a multimetric invertebrate based assessment system. *Limnologica*, **34**, 416–432.
- ter Braak C.J.F. & Smilauer P. (2002) *CANOCO Reference Manual and CanoDraw for Windows User's Guide Version 4.5*. Biometris – Plant Research International, Wageningen and České Budějovice.
- Buffagni A. (1997) Mayfly community composition and the biological quality of streams. In: *Ephemeroptera & Plecoptera: Biology-Ecology-Systematics* (Eds P. Landolt & M. Sartori), pp. 235–246. MTL, Fribourg.
- Buffagni A. (1999) Pregio naturalistico, qualità ecologica e integrità della comunità degli Efemeroteri. Un indice per la classificazione dei fiumi italiani. *Acqua & Aria*, **8**, 99–107.
- Buffagni A., Erba S., Cazzola M. & Kemp J.L. (2004) The AQEM multimetric system for the southern Italian Apennines: assessing the impact of water quality and habitat degradation on pool macroinvertebrates in Mediterranean rivers. *Hydrobiologia*, **516**, 313–329.
- CEMAGREF (1982) *Etude des méthodes biologiques d'appréciation quantitative de la qualité des eaux*. Rapport Cemagref Q.E. Lyon-A.F. Bassin Rhône-Méditerranée-Corse, pp. 1–218.
- CEN (2003) *Water Quality – Sampling of Fish with Electricity*. EN 14011, European Committee for Standardization, Brussels.
- Coring E. (1993) *Zum Indikationswert bentischer Diatomeengesellschaften in basenarmen Fließgewässern*. Verlag Shaker, Aachen.
- Coring E. (1999) Situation and developments of algal (diatom)-based techniques for monitoring rivers in Germany. In: *Use of Algae for Monitoring Rivers III* (Eds J. Prygiel, B.A. Whitton & J. Bukowska), pp. 122–127. Agence de l'Eau Artois-Picardie, Douai.
- Council of the European Communities (2000) *Establishing a Framework for Community Action in the field of Water Policy*. Directive 2000/60/EC.
- van Dam H. (1997) Partial recovery of moorland pools from acidification: indications by chemistry and diatoms. *Netherlands Journal of Aquatic Ecology*, **30**, 203–218.
- van Dam H., Mertens A. & Sinkeldam J. (1994) A coded checklist and ecological indicator values of freshwater diatoms from the Netherlands. *Netherlands Journal of Aquatic Ecology*, **28**, 117–133.
- Dawson F. H. (2002) *Guidance for the Field Assessment of Macrophytes of Rivers within the STAR Project*. <http://www.eu-star.at/frameset.htm>.
- De Pauw N. & Hawkes H.A. (1993) Biological monitoring of river water quality. In: *Proceedings of the 'Freshwater Europe Symposium on River Water Quality Monitoring and Control'* (Eds W.J. Walley & S. Judd), pp. 87–111. Published by Aston University, Aston, U.K.
- Dell'Uomo A. (1996) Assessment of water quality of an Apennine river as a pilot study for diatom-based monitoring of Italian watercourses. In: *Use of Algae for Monitoring Rivers II* (Eds B.A. Whitton & E. Rott), pp. 65–72. Institut für Botanik, Universität Innsbruck, Innsbruck.
- Denys L. (1991a) A check-list of the diatoms in the holocene deposits of the Western Belgian coastal plain with a survey of their apparent ecological requirements. I. Introduction, ecological code and complete list. *Ministère des Affaires Economiques – Service Géologique de Belgique*.
- Denys L. (1991b) A check-list of the diatoms in the holocene deposits of the Western Belgian coastal plain with a survey of their apparent ecological requirements. II. Centrales. *Ministère des Affaires Economiques – Service Géologique de Belgique*.
- Descy J.P. (1979) A new approach to water quality estimation using diatoms. *Nova Hedwigia Beiheft*, **64**, 305–323.
- Descy J.P. & Coste M. (1990) *Utilisation des diatomées benthiques pour l'évaluation de la qualité des eaux courantes*. Rapport final, UNCED, Namur, Cemagref, Bordeaux.
- Dolédec S., Stutzner B. & M. Bournard (1999) Species traits for future biomonitoring across ecoregions: patterns along a human-impacted river. *Freshwater Biology*, **42**, 737–758.
- DVWK (Deutscher Verband für Wasserwirtschaft und Kulturbau e.V.) (1999) *Durchgehendes Trophiesystem auf der Grundlage der Trophieindikation mit Kieselalgen*. DVWK Materialien 6/1999, Bonn.
- Ellenberg H., Weber H.E., Düll R., Wirth V., Werner W. & Paulißen D. (1992) Zeigerwerte von Pflanzen in Mitteleuropa. *Scripta Geobotanica*, **18**, 1–257.

- Furse M.T., Hering D., Moog O. *et al.* (2006) The STAR project: context, objectives and approaches. *Hydrobiologia*, **566**, 3–29.
- Ghetti P.F. (1997) Manuale di applicazione Indice Biotico Estesio (I.B.E.). I macroinvertebrati nel controllo della qualità degli ambienti di acque correnti. Provincia Autonoma di Trento, Agenzia provinciale per la protezione dell'ambiente. Trento.
- Gorman O.T. & Karr J.R. (1978) Habitat structure and stream fish communities. *Ecology*, **59**, 507–515.
- Hall R.I., Leavitt P.R., Quinlan R., Dixit A.S. & Smol J.P. (1999) Effects of agriculture, urbanization, and climate on water quality in the northern Great Plains. *Limnology and Oceanography*, **44**, 739–756.
- Haury J., Peltre M.C., Tremolieres M. *et al.* (2002) A method involving macrophytes to assess water trophy and organic pollution: the Macrophyte Biological Index for Rivers (IBMR) - application to different types of rivers and pollutions. In: *Proceedings 11th EWRS Internat. Symp. Aquatic Weeds* (Eds A. Dutartre & M.H. Montel), pp. 247–250. Moliets Et Maa, France.
- Heiskanen A.S., van de Bund W., Cardoso A.C. & Noges P. (2004) Towards good ecological status of surface waters in Europe – interpretation and harmonisation of the concept. *Water Science and Technology*, **49**, 169–177.
- Hellawell J.M. (1986) *Biological Indicators of Freshwater Pollution and Environmental Management*. Elsevier Applied Science, London.
- Henrikson L. & Medin M. (1986) Biologisk bedömning av försurningspåverkan på Lelångens tillflöden och grundområden 1986 (Biological assessment of acidification on the Lelängen tributaries and shallow areas 1986). *Aquakelogerna, Report to the Local Countyboard of Älvsborg*.
- Hering D., Moog O., Sandin L. & Verdonschot P.F.M. (2004b) Overview and application of the AQEM assessment system. *Hydrobiologia*, **516**, 1–20.
- Hering D., Meier C., Rawer-Jost C., Feld C.K., Biss R., Zenker A., Sundermann A., Lohse S. & Böhmer J. (2004a) Assessing streams in Germany with benthic invertebrates: selection of candidate metrics. *Limnologia*, **34**, 398–415.
- Hofmann G. (1994) Aufwuchs-Diatomeen in Seen und ihre Eignung als Indikatoren der Trophie. *Bibliotheca Diatomologica*, **30**, 1–241.
- Hofmann G. (1999) *Trophiebewertung von Seen anhand von Aufwuchsdiatomeen*. In: *Biologische Gewässeruntersuchung* (Eds W. Von Tümpling & G. Friedrich), **2**, 319–333. Gustav Fischer Verlag, Jena.
- Holmes N.T.H., Newman J.R., Chadd S., Rouen K.J., Saint L. & Dawson F.H. (1999) *Mean Trophic Rank: a Users Manual*. R & D Technical Report No. E38, Environment Agency, Bristol, UK.
- Hürlimann J., Elber F. & Niederberger K. (1999) In: *Use of Algae for Monitoring Rivers: an Overview of the Current Situation and Recent Developments in Switzerland. Use of Algae for Monitoring Rivers III* (Eds J. Prygiel, B.A. Whitton & J. Bukowska), pp. 39–56. Agence de l'Eau Artois-Picardie, Douai.
- Illies J. (ed) (1978) *Limnofauna Europaea*. Gustav Fischer Verlag, Stuttgart.
- Jalas J. (1955) Hemorobe und hemerokore Pflanzenarten. Ein terminologischer Reformversuch. *Acta Societas Fauna Flora Fennici*, **72**, 1–15.
- Johnson R.K., Hering D., Furse M.T. & Clarke R. (2006a) Detection of ecological change using multiple organism groups: metrics and uncertainty. *Hydrobiologia*, **566**, 115–137.
- Johnson R.K., Hering D., Furse M.T. & Verdonschot P.F.M. (2006b) Indicators of ecological change: comparison of the early response of four organism groups to stress gradients. *Hydrobiologia*, **566**, 139–152.
- Karr J.R. (1981) Assessment of biotic integrity using fish communities. *Fisheries*, **6**, 21–27.
- Karr J.R. & Chu E.W. (1999) *Restoring Life in Running Waters: Better Biological Monitoring*. Island Press, Washington, DC.
- Kelly M.G., Penny C.J. & Whitton B.A. (1995) Comparative performance of benthic diatom indices to assess river water quality. *Hydrobiologia*, **302**, 179–188.
- Knoben R.A. Roos E.C. & van Oirschot M.C.M. (1995) *Biological Assessment Methods for Watercourses*. UN/ECE Task Force on Monitoring & Assessment. Volume 3. RIZA report no. 95.066. Lelystad.
- Kolkwitz R. & Marson M. (1909) Ökologie der tierischen Saprobien. *Internationale Revue der gesamten Hydrobiologie*, **2**, 126–152.
- Lancaster J., Real M., Juggins S., Monteith D.T., Flower R.J. & Beaumont W.R.C. (1996) Monitoring temporal changes in the biology of acid waters. *Freshwater Biology*, **36**, 179–201.
- Leclercq L. & Maquet B. (1987) Two new chemical and diatomic indicators of the quality of running water. Application to the Samson and its tributaries (basin of the Belgian Meuse). Comparison with other chemical indicators, biocenose and diatomic indicators. *Royal Belgian Institute of Natural Sciences, Work document* **38**, 1–113.
- Lenat D.R. & Crawford J.K. (1994) Effects of land use on water quality and aquatic biota of three North Carolina Piedmont streams. *Hydrobiologia*, **294**, 185–199.
- Lenoir A. & Coste M. (1996) Development of practical diatomic index of overall water quality applicable to the French National Water Board Network. In: *Use of Algae for Monitoring Rivers II* (Eds B.A. Whitton &

- E. Rott), pp. 29–43. Institut für Botanik, Universität Innsbruck, Innsbruck.
- Lorenz A., Hering D., Feld C.K. & Rolauffs P. (2004a) A new method for assessing the impact of morphological degradation on the benthic invertebrate fauna for streams in Germany. *Hydrobiologia*, **516**, 107–127.
- Lorenz A., Kirchner L. & Hering D. (2004b) 'Electronic subsampling' of macrobenthic samples: how many individuals are needed for a valid assessment result? *Hydrobiologia*, **516**, 299–312.
- Malmqvist B. & Rundle S. (2002) Threats to the running water ecosystems of the world. *Environmental Conservation*, **29**, 134–153.
- Margalef R. (1984) The Science and praxis of complexity. In: *Ecosystems: Diversity and Connectivity as Measurable Components of Their Complication* (Ed. E.A. Aida), pp. 228–244. United Nations University, Tokyo.
- Mazor R.D., Reynoldson T.B. Rosenberg D.M. and Resh V.H. (2006) Effects of biotic assemblage, classification, and assessment method on bioassessment performance. *Canadian Journal of Fisheries and Aquatic Sciences*, **63**, 394–411.
- McNaughton S.J. (1967) Relationships among functional properties of Californian grasslands. *Nature*, **216**, 168–169.
- Northcote T.G. (1998) Migratory behaviour of fish and its significance to movement through riverine fish passage facilities. In: *Fish Migration and Fish Bypasses*. *Fishing News Book* (Eds M. Jungwirth, S. Schmutz & S. Weiss), pp. 3–18. University Press, Cambridge.
- Oberdorff T., Pont D., Hugueny B. & Chessel D. (2001) A probabilistic model characterizing fish assemblages of French rivers: a framework for environmental assessment. *Freshwater Biology*, **46**, 399–415.
- Ormerod S.J., Rundle S.D., Wilkinson S.M., Daly G.P., Dale K.M. & Juttner I (1994) Altitudinal trends in the diatoms, bryophytes, macroinvertebrates and fish of a Nepalese river system. *Freshwater Biology*, **32**, 309–322.
- Passauer B., Meilinger P. Melzer A. & Schneider S. (2002) Does the structural quality of running waters affect the occurrence of macrophytes? *Acta Hydrochimica et Hydrobiologica*, **30**, 197–206.
- Pielou E.C. (1966) The measurement of diversity in different types of biological collections. *Journal Theoretical Biology*, **13**, 131–144.
- Podraza P., Schuhmacher H. & Sommerhäuser M. (2000) Composition of macroinvertebrate feeding groups as a bioindicator in running waters. *Verhandlungen der internationalen Vereinigung für theoretische und angewandte Limnologie*, **27**, 3066–3069.
- Pont D., Hugueny B., Beier U., Goffaux D., Melcher A., Noble R., Rogers C., Roset N. & Schmutz S. (2006) Assessing river biotic condition at the continental scale: a European approach using functional metrics and fish assemblages. *Journal of Applied Ecology*, **43**, 70–80.
- Prygiel J., Carpentier P., Almeida S. *et al.* (2002) Determination of the biological diatom index (IBD NF T 90–354): results of an intercomparison exercise. *Journal of Applied Phycology*, **14**, 27–39.
- Rosenberg D.M. & Resh V.H. (1993) Introduction to freshwater biomonitoring and benthic macroinvertebrates. In: *Freshwater Biomonitoring and Benthic Macroinvertebrates* (Eds D.M. Rosenberg & V.H. Resh), pp. 1–9. Chapman and Hall, New York.
- Rott E., Hofmann G., Pall K., Pfister P. & Pipp E. (1997) *Indikationslisten für Aufwuchsalgen. Teil 1: Saprobielle Indikation*. Bundesministerium für Land- und Forstwirtschaft, Wien, 73 pp.
- Rott E., Pfister P., van Dam H., Pipp E., Pall K., Binder N. & Ortler K. (1999) *Indikationslisten für Aufwuchsalgen. Teil 2: Trophieindikation sowie geochemische Präferenz, taxonomische und toxikologische Anmerkungen*. Bundesministerium für Land- und Forstwirtschaft, Wien, 248 pp.
- Rumeau A. & Coste M. (1988) Initiation à la systématique des diatomées d'eau douce pour l'utilisation pratique d'un indice diatomique générique. *Bulletin Francais de la Peche et de la Pisciculture*, **309**, 1–69.
- Sandin L., Dahl J. & Johnson R.K. (2004) Assessing acid stress in Swedish boreal and alpine streams using benthic macroinvertebrates. *Hydrobiologia*, **516**, 129–148.
- Sawyer J.A., Stewart P.M., Mullen M.M., Simon T.P. & Bennett H.H. (2004) Influence of habitat, water quality, and land use on macro-invertebrate and fish assemblages of a southeastern coastal plain watershed, USA. *Aquatic Ecosystem Health & Management*, **7**, 85–99.
- Schaumburg J., Schranz C., Foerster J., Gutowski A., Hofmann G., Meilinger P., Schneider S. & Schmedtje U. (2004) Ecological classification of macrophytes and phytobenthos for rivers in Germany according to the Water Framework Directive. *Limnologica*, **34**, 283–301.
- Schneider S., Krumpholz T. & Melzer A. (2000) Trophieindikation in Fließgewässern mit Hilfe des TIM (Trophie-Index Macrophyten) – Erprobung eines neu entwickelten Index im Inniger Bach. *Acta Hydrochimica Hydrobiologica*, **28**, 241–249.
- Schweder H. (1992) Neue Indices für die Bewertung des ökologischen Zustandes von Fließgewässern, abgeleitet aus der Makroinvertebraten-Ernährungstypologie. *Limnologie Aktuell*, **3**, 353–377.

- Seber G.A.F. & LeCren E.D. (1967) Estimating population parameters from catches large relative to the population. *Journal of Animal Ecology*, **36**, 631–643.
- Shannon C.E. & Weaver W. (1949) *The Mathematical Theory of Communication*. University of Illinois Press, Urbana.
- Simpson E.H. (1949) Measurement of Diversity. *Nature*, **163**, 688.
- Skriver J., Friberg N. & Kirkegaard J. (2001) Biological assessment of running waters in Denmark: introduction of the Danish Stream Fauna Index (DSFI). *Verhandlungen der internationalen Vereinigung für theoretische und angewandte Limnologie*, **27**, 1822–1830.
- Sladeczek V. (1986) Diatoms as indicators of organic pollution. *Acta Hydrochimica Hydrobiologica*, **14**, 555–566.
- Snyder C.D., Young J.A., Villella R. & Lemarié D.P. (2003) Influences of upland and riparian land use patterns on stream biotic integrity. *Landscape Ecology*, **18**, 647–664.
- Sonneman J.A., Walsh C.J., Breen P.F. & Sharpe A.K. (2001) Effects of urbanization on streams of the Melbourne region, Victoria, Australia. II. Benthic diatom communities. *Freshwater Biology*, **46**, 553–565.
- Stanner D. & Bordeau P. (Eds) (1995) *Europe's Environment. The Dobris Assessment*. European Environment Agency, Copenhagen.
- Statzner B., Bis B., & Usseglio-Polatera P. (2001) Perspectives for biomonitoring at large spatial scales: a unified measure for the functional composition of invertebrate communities in European running waters. *Basic and Applied Ecology*, **1**, 73–85.
- Steinberg C. & Schiefele S. (1988) Biological indication of trophy and pollution of running waters. *Zeitschrift für Wasser- und Abwasser-Forschung*, **21**, 227–234.
- Stevenson R.J., Bailey R.C., Harrass M.C. et al. (2004) Designing data collection for ecological assessments. In: *Ecological Assessment of Aquatic Resources: Linking Science to Decision Making* (Eds M.T. Barbour, S.B. Norton, H.R. Preston & K.W. Thornton), pp. 55–84. SETAC, Pensacola, Florida, USA.
- Svendsen L. & Rebsdorf A. (1994) Kvalitetssikring af Overvågningsdata. Retningslinier for Kvalitetssikring af Ferskvandskemiske Data i Vandmiljøplanenes Overvågningsprogram. (Quality Assurance of Monitoring Data). *Teknisk Anvisning fra DMU*, **7**.
- Townsend C.R., Hildrew A.G. & Francis J.E. (1983) Community structure in some southern English streams: the influence of physicochemical factors. *Freshwater Biology*, **13**, 521–544.
- Trempe H. & Kohler A. (1995) The usefulness of macrophyte monitoring-systems, exemplified on eutrophication and acidification of running waters. *Acta Botanica Gallica*, **142**, 541–550.
- Triest L., Parminder K., Heylen S. & de Pauw N. (2001) Comparative monitoring of diatoms, macroinvertebrates and macrophytes in the Woluwe River (Brussels, Belgium). *Aquatic Ecology*, **35**, 183–194.
- USEPA (2000) *Stressor identification guidance document*. U.S. Environmental Protection Agency, Washington, USA, EPA-822-B-00-025, 228 pp.
- Verdonschot P.F.M. (2006) Evaluation of the use of Water Framework Directive typology descriptors, reference sites, and spatial scale in macroinvertebrate stream typology. *Hydrobiologia*, **566**, 39–58.
- Verdonschot P.F.M. & R.C. Nijboer, 2004. Testing the European stream typology of the water Framework Directive for macroinvertebrates. *Hydrobiologia*, **175**, 35–54.
- Vermaat J.E. & Debruyne R.J. (2003) Factors limiting the distribution of submerged waterplants in the lowland river Vecht (The Netherlands). *Freshwater Biology*, **30**, 147–157.
- Walsh C.J., Sharpe A.K., Breen P.F. & Sonneman J.A. (2001) Effects of urbanization on streams of the Melbourne region, Victoria, Australia. I. Benthic macroinvertebrate communities. *Freshwater Biology*, **46**, 535–551.
- Watanabe T., Asai K., Houki A., Tanaka S. & Hizuka T. (1986) Saprophytic and eurytopic diatom taxa to organic water pollution and diatom assemblages index (DAIpo). *Diatom*, **2**, 23–73.
- Westlake D.F. (1975) Macrophytes. In: *River ecology* (Ed. Whitton B.A.), pp. 106–128. University of California Press, Berkeley, California.
- Zelinka M. & Marvan P. (1961) Zur Präzisierung der biologischen Klassifikation der Reinheit fließender Gewässer. *Archiv für Hydrobiologie*, **57**, 389–407.
- Ziemann H. (1999) Bestimmung des Halobienindex. In: *Biologische Gewässeruntersuchung* (Eds W. Tümping & G. Friedrich), *Methoden der Biologischen Gewässeruntersuchung*, **2**, 310–313.

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Appendix Spearman rank correlation ( $r^2$ ) of metrics and environmental gradients in the individual stream type (groups)

	All mountain streams					All lowland streams					Lowland streams U15/ U23					Lowland streams D03/ K02				
	e	l	h	m	g	e	l	h	m	g	e	l	h	m	g	e	l	h	m	g
Diatom metrics																				
D_IPS		0.21	0.06		0.06	0.42	0.21			0.33	0.29	0.41			0.39					
D_Slad	0.24	0.24				0.50	0.33			0.26	0.25	0.28			0.36	0.40				
D_Descy		0.06				0.23	0.22			0.28	0.26	0.18								
D_L&M	0.26	0.17				0.55	0.38			0.27	0.25	0.39			0.66	0.41				
D_She	0.16	0.15	0.05		0.06	0.44	0.29			0.36	0.26	0.35			0.46					
D_Wat	0.10	0.12			0.05					0.14		0.41				0.36				
D_TDI	0.10	0.16				0.51	0.19				0.12								0.25	
D_%PT	0.30	0.17		0.14		0.59	0.24			0.13	0.20	0.35			0.32				0.30	
D_EPI-D	0.40	0.27		0.07		0.59	0.20			0.27	0.25	0.25				0.75	0.20			
D_Rott(Om)	0.40	0.32		0.06		0.58	0.16			0.11	0.29	0.34								
D_IDG	0.27	0.21		0.07		0.45	0.25			0.24	0.20	0.40			0.36	0.31	0.31	0.27		
D_CEE	0.31	0.19		0.10		0.36	0.12			0.18	0.20	0.46			0.31	0.31	0.21			
D_IBD	0.33	0.21		0.08		0.35	0.21			0.24	0.24	0.42			0.31	0.18				
D_IDAP	0.21					0.09	0.21			0.06	0.05	0.35			0.52					
D_TDI_DVWK	0.40	0.33		0.11		0.62	0.32			0.28	0.33	0.73			0.42	0.17	0.46	0.22		
D_TDI_Rott_G	0.34	0.24		0.20		0.67	0.23			0.29	0.34	0.55			0.57	0.18	0.33	0.21		
D_TDI_lakes	0.48	0.19		0.09		0.41	0.05			0.14	0.21	0.51			0.44	0.17				
D_DI_Swi	0.15	0.12	0.09	0.11	0.12	0.44	0.27			0.26	0.32	0.38								
D_SI_Rott	0.50	0.35		0.14		0.60	0.16				0.25	0.48						0.20		
D_Halo	0.35	0.12				0.41	0.25			0.25	0.22					0.29	0.22		0.21	
D_Phylip_DI_1a	0.27	0.23		0.19		0.61	0.17			0.24	0.31	0.54			0.39					
D_Phylip_DI_4	0.33	0.37		0.09		0.60	0.20			0.19	0.30	0.53			0.76	0.18	0.32	0.20		
D_Phylip_DI_7b	0.07	0.13			0.06	0.14	0.09				0.07	0.36			0.35					
D_Phylip_DI_9	0.06	0.11			0.06	0.17	0.06				0.07	0.44				0.17				
D_Phylip_RLI	0.16			0.17		0.43	0.05				0.17									
Macrophyte metrics																				
M_no_mo_li	0.15						0.12			0.27	0.11	0.17					0.00			
M_no_subm		0.14	0.25		0.24									0.20					0.22	
M_no_anch		0.10	0.18		0.13	0.05		0.09	0.08	0.08					0.50					
M_no_float		0.21	0.10	0.20					0.27	0.09						0.77			0.25	
M_no_amph		0.36		0.08	0.12			0.09	0.11	0.20						0.74			0.35	
M_no_terr	0.45				0.09		0.15			0.31	0.08									
M_cov_mo_li		0.14	0.24							0.04	0.21	0.25					0.19	0.38		
M_cov_subm		0.14	0.24		0.25					0.04		0.19					0.19	0.38		
M_cov_anch		0.08	0.19		0.12			0.12	0.12	0.10		0.19				0.35				
M_cov_float	0.11	0.18						0.06	0.17	0.17										
M_cov_amph	0.08	0.15	0.11	0.19	0.12		0.04		0.22	0.07		0.16	0.43		0.71			0.21		
M_cov_terr		0.33			0.11			0.12	0.09	0.17			0.48	0.64		0.64			0.38	
M_MTR	0.27	0.24				0.22	0.12				0.05									
M_IBMR	0.26	0.24		0.16		0.30	0.15			0.09	0.06									
M_Ellenberg_N						0.12	0.25			0.07							0.21			
M_Hemeroby index						0.09	0.12					0.40		0.35	0.28	0.31				
M_sp_no		0.15		0.09	0.17		0.07	0.16	0.18							0.82			0.36	
M_ge_no		0.15		0.09	0.16		0.06	0.15	0.18							0.82			0.39	
M_fa_no		0.15		0.08	0.17		0.07	0.14	0.17							0.81			0.39	
M_Div(SW)			0.16		0.21		0.12	0.22	0.21				0.70	0.63		0.63	0.35	0.42		
M_dom					0.10		0.10		0.08											
M_Ellenberg_N*						0.16	0.20													
M_ty_sp_no		0.11	0.12		0.16		0.06	0.19	0.18							0.78		0.30	0.40	
M_ty_dom			0.11		0.11		0.08	0.06	0.07			0.16								
M_ty_evenn					0.21		0.08	0.23	0.14				0.62	0.30		0.30	0.40	0.31		
M_Ge_sp_no		0.18	0.15	0.12	0.20			0.23	0.11							0.80	0.30	0.36		

## Appendix (Continued)

	All mountain streams					All lowland streams					Lowland streams U15/ U23					Lowland streams D03/ K02				
	e	l	h	m	g	e	l	h	m	g	e	l	h	m	g	e	l	h	m	g
M_Div(Si)_Ge			0.11		0.22				0.23	0.10				0.63	0.55	0.31			0.33	
M_Ge_dom			0.14	0.09	0.12				0.14	0.04										
M_Ge_evenn		0.10	0.13		0.22		0.04		0.28	0.09				0.66	0.33	0.33			0.31	
Invertebrate metrics																				
I_Acid_Index	0.02		0.06								0.30									
I_ASPT	0.41	0.23		0.12		0.17	0.11	0.12		0.12	0.33				0.39	0.32			0.48	
I_Div(Marg)	0.21	0.15	0.11		0.10	0.05			0.06		0.31									
I_Div(SW)	0.20	0.10	0.15	0.09	0.09															
I_DSFI	0.28	0.23		0.09	0.10	0.08	0.12	0.15	0.09	0.25	0.18				0.31	0.32			0.51	
I_epirhithral	0.28	0.28	0.07	0.06	0.16	0.10	0.10		0.38					0.31			0.62			
I_EPT-taxa				0.08	0.08	0.09				0.05	0.37									
I_EPT-taxa (%)	0.44	0.31		0.07	0.09	0.06	0.05	0.26		0.11	0.23					0.45			0.34	
I_gatherers	0.30	0.17	0.14	0.10	0.11	0.18		0.08		0.04				0.20	0.39				0.49	
I_GFID01	0.41	0.27		0.20		0.08		0.15		0.11	0.26					0.36			0.29	
I_GFID02	0.26	0.17		0.08			0.08		0.31	0.05	0.16						0.39			
I_GFID03	0.18	0.16	0.09		0.09			0.05	0.08											
I_GFID04	0.18	0.21			0.14			0.16		0.16	0.26									
I_GFID05	0.41	0.27		0.20	0.09	0.08		0.15		0.11	0.20					0.36			0.29	
I_grazers	0.26	0.17		0.08			0.08		0.31	0.05							0.39			
I_IBE_Aqem	0.18	0.16	0.09		0.10			0.05	0.08		0.21		0.19							
I_Index_BR	0.18	0.21			0.17			0.16		0.16										
I_littoral	0.49	0.17		0.19				0.19		0.19	0.19				0.35					
I_MAS_IC	0.27	0.21	0.12	0.12	0.12		0.08				0.28									
I_metarhithral	0.33	0.20		0.33				0.15	0.08	0.19										
I_no_families	0.14		0.12		0.05	0.05	0.04				0.37									
I_Oligochaeta			0.13			0.10														
I_Oligochaeta (%)	0.19	0.16		0.16		0.10														
I_passive_filt	0.10	0.16		0.18		0.10	0.12		0.09				0.35							
I_pelal	0.25	0.25		0.23	0.07	0.16	0.05		0.17	0.11										
I_Plecoptera (%)	0.37	0.28	0.05	0.09	0.14	0.24	0.09				0.16				0.33	0.38			0.52	
I_RETI	0.24	0.19		0.14						0.06										
I_rheophile	0.29	0.22	0.08	0.08		0.09		0.14		0.09						0.38			0.26	
I_shredders			0.21		0.10	0.09			0.18								0.31			
I_SI(ZM)	0.24	0.23	0.10		0.22	0.12	0.10	0.15	0.13	0.19	0.19		0.35	0.25						
I_Trichoptera (%)	0.30	0.05	0.10	0.14		0.11	0.10				0.21									
I_xeno	0.07	0.10	0.23		0.28		0.06			0.06				0.18	0.37					
Fish metrics																				
F_n_sp_tol	0.10	0.08	0.12	0.10			0.13				0.20								0.26	
F_n_sp_intol			0.10					0.17	0.06	0.24	0.58				0.20	0.38			0.48	
F_perc_sp_intol	0.09	0.12					0.04	0.15	0.10	0.19	0.47								0.30	
F_n_ha_hab_wc	0.31	0.16		0.16						0.13										
F_n_sp_hab_b	0.12									0.04					0.42					
F_perc_sp_hab_b																				
F_n_ha_hab_b						0.09			0.14					0.69						
F_perc_nha_hab_b																		0.64		
F_n_sp_hab_rh	0.14		0.06		0.06	0.10		0.15		0.19		0.20				0.24			0.30	
F_n_ha_hab_rh	0.26	0.09		0.14				0.11		0.28	0.18			0.19	0.41				0.30	
F_perc_sp_hab_li		0.07					0.17		0.12	0.10	0.17				0.35	0.40				
F_n_ha_hab_li		0.07					0.18		0.12	0.10	0.17				0.35	0.40				
F_n_sp_hab_eury	0.10	0.08	0.12	0.10				0.11			0.22								0.22	
F_n_ha_hab_eury	0.09	0.08	0.12	0.11				0.09						0.26	0.19	0.42			0.53	
F_n_sp_re_lith	0.14							0.13		0.26	0.24	0.18							0.26	
F_perc_nha_re_lith		0.13	0.10	0.14			0.07	0.17	0.08	0.29	0.24			0.26		0.25			0.39	



## Appendix (Continued)

	Lowland streams S05/S06					Lowland streams O02				
	e	l	h	m	g	e	l	h	m	g
M_cov_subm										
M_cov_anch										0.27
M_cov_float		0.16	0.20						0.23	
M_cov_amph	0.32	0.42		0.25						0.32
M_cov_terr						0.27		0.30		0.21
M_MTR	0.73	0.58			0.57		0.38			
M_IBMR	0.72	0.56			0.57		0.22			
M_Ellenberg_N	0.62	0.46			0.51		0.26			
M_Hemeroby index	0.63	0.58			0.49		0.38			0.24
M_sp_no		0.20				0.25				0.20
M_ge_no		0.21								0.21
M_fa_no		0.20								
M_Div(SW)	0.18	0.32		0.24						
M_dom			0.18							
M_Ellenberg_N*	0.64	0.46			0.52					
M_ty_sp_no	0.10									
M_ty_dom	0.04									
M_ty_evenn	0.05			0.41						
M_Ge_sp_no	0.14	0.23								
M_Div(Si)_Ge	0.12	0.15		0.22						
M_Ge_dom	0.06			0.07						
M_Ge_evenn	0.11			0.36						
Invertebrate metrics										
I_Acid_Index						0.04	0.19			
I_ASPT	0.72	0.66			0.43		0.19		0.21	0.45
I_Div(Marg)		0.15								
I_Div(SW)										
I_DSFI	0.32	0.32			0.22	0.46		0.38	0.32	0.49
I_epirhithral										
I_EPT-taxa					0.00					
I_EPT-taxa (%)	0.45	0.34						0.49	0.28	0.56
I_gatherers	0.35	0.23			0.11					0.19
I_GFID01	0.18							0.51	0.25	0.52
I_GFID02				0.21						
I_GFID03				0.30			0.43			0.38
I_GFID04						0.31		0.26	0.31	0.26
I_GFID05	0.18							0.51	0.25	0.52
I_grazers				0.21						
I_IBE_Aqem				0.30			0.43			0.38
I_Index_BR						0.31		0.26	0.31	0.26
I_littoral						0.47		0.38	0.33	0.25
I_MAS_IC	0.28	0.30								
I_metarhithral			0.20			0.39		0.32	0.21	0.27
I_no_families										
I_Oligochaeta				0.19						
I_Oligochaeta (%)										
I_passive_filt								0.30		0.48
I_pelal							0.21	0.33	0.23	0.52
I_Plecoptera (%)	0.48	0.42			0.44		0.44			
I_RETI				0.17			0.20			
I_rheophile	0.16							0.27	0.24	0.40
I_shredders							0.23			
I_SI(ZM)			0.19				0.22	0.29		0.49
I_Trichoptera (%)							0.36			
I_xeno										0.38

## Appendix (Continued)

	Lowland streams S05/S06					Lowland streams O02				
	e	l	h	m	g	e	l	h	m	g
Fish metrics										
F_n_sp_tol	0.22	0.21				0.45				
F_n_sp_intol			0.18	0.29						0.21
F_perc_sp_intol			0.24							0.24
F_n_ha_hab_wc										
F_n_sp_hab_b						0.25		0.22		
F_perc_sp_hab_b			0.17		0.16					
F_n_ha_hab_b			0.16					0.46		
F_perc_nha_hab_b								0.32		0.38
F_n_sp_hab_rh	0.25	0.20		0.27			0.20			0.24
F_n_ha_hab_rh			0.20							
F_perc_sp_hab_li										
F_n_ha_hab_li										
F_n_sp_hab_eury						0.31		0.22		
F_n_ha_hab_eury								0.21		
F_n_sp_re_lith	0.41	0.40		0.24						0.20
F_perc_nha_re_lith	0.15	0.22								0.29
F_n_ha_re_pht	0.18					0.31		0.46	0.45	0.34
F_n_sp_lon_ll										
F_perc_sp_lon_ll										0.25
F_n_sp_lon_sl				0.26						
F_perc_sp_lon_sl		0.19		0.18						
F_n_ha_lon_sl		0.15	0.16					0.28		
F_n_ha_fe_pisc		0.15							0.22	
F_n_sp_fe_insev			0.16	0.24						0.27
F_n_ha_fe_insev			0.23					0.21		0.31
F_n_ha_fe_omni		0.18								
F_n_sp_mi_long										
F_n_sp_mi_potad										

All correlations are significant at  $P < 0.01$ .

Metric abbreviations: see Table 3.

e, eutrophication/organic pollution gradient; l, land use gradients; h, hydromorphology (reach scale) gradients; m, hydromorphology (microhabitat scale) gradients; g, general degradation gradients.