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de Vries, J.; Kraak, M.H.S.; Verdonschot, R.C.M.; Verdonschot, P.F.M.

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Quantifying cumulative stress acting on macroinvertebrate assemblages in lowland streams



Jip de Vries^{a,*}, Michiel H.S. Kraak^a, Ralf C.M. Verdonschot^b, Piet F.M. Verdonschot^{a,b}

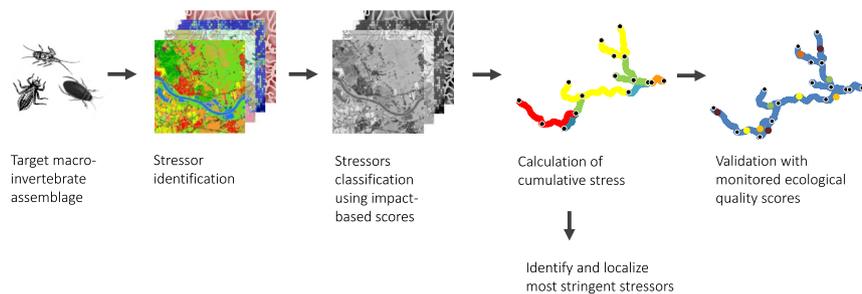
^a Freshwater and Marine Ecology, Institute for Biodiversity and Ecosystem Dynamics, University of Amsterdam, P.O. Box 94248, 1090 GE, Amsterdam, the Netherlands

^b Wageningen Environmental Research, Wageningen UR, P.O. Box 47, 6700 AA, Wageningen, the Netherlands

HIGHLIGHTS

- A method to quantify cumulative stress acting on macroinvertebrates is presented.
- Stressor intensity was categorized based on impact on macroinvertebrate assemblages.
- Stressor specific contributions to overall stress explain species distributions.
- This method may provide a better focus for management resources.

GRAPHICAL ABSTRACT



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ABSTRACT

Macroinvertebrates in lowland streams are exposed to multiple stressors from the surrounding environment. Yet, quantifying how these multiple stressors impact macroinvertebrate assemblages is challenging. The aim of this study was to develop a novel method to quantify the cumulative stress acting on macroinvertebrate assemblages in lowland streams. To this purpose, we considered 22 stressors from different stressor categories such as hydrological, morphological and chemical stressors, acting over multiple spatial scales ranging from instream to the catchment scale. Stressor intensity was categorized into classes based on impact on the macroinvertebrate assemblages. The main stream was divided into segments, after which for each stream segment, the cumulative stressor contribution from headwater catchments, from the riparian zone and from upstream was calculated. To validate the cumulative stress quantification method, the lowland stream Tengelroyse Beek in the Netherlands was used as a case study. For this stream it was shown that independently derived ecological quality scores based on macroinvertebrate samples collected at multiple sites along the stream decreased with increasing calculated cumulative stress scores, supporting the design of the cumulative stress quantification method. Based on the contribution of each specific stressor to the cumulative stress scores, the reasons for the absence and presence of macroinvertebrate species may be elucidated. Hence, the cumulative stress quantification method may help to identify and localize the most stringent stressors limiting macroinvertebrate assemblages, and can thereby provide a better focus for management resources.

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1. Introduction

The Water Framework Directive (WFD) aims to achieve a good ecological status of aquatic ecosystems by protecting ecosystem structure

* Corresponding author.

E-mail address: j.de.vries@uva.nl (J. de Vries).

and functioning. Assessing ecosystem structure is generally based on community metrics, such as the macroinvertebrate community composition, as they integrate water quality parameters over longer timescales (De Pauw et al., 2006; Metcalfe, 1989). Water-type specific target species lists serve as reference situations for quality assessments and as goals for the restoration of degraded water bodies (De Pauw et al., 2006). Yet, in many cases the actual causes of the presence and absence of specific target species remain virtually unknown, and subsequently, little is known about the factors that drive successful river restoration (Pander and Geist, 2013).

So far, research to understand the presence and absence of species strongly focused on local environmental factors and single stressors (Tockner et al., 2010). Although these studies have increased the available knowledge of the relationships between single species and single stressors, they do not reflect the actual situation in multistressed ecosystems, where stressors hardly ever act in isolation (Ormerod et al., 2010). For example, in lowland stream ecosystems, sewage pollution may raise the instream nutrient load, decrease oxygen concentrations and add toxic compounds (Camargo and Alonso, 2006). Focus on single stressors can thus lead to erroneous conclusions on the reasons for the absence of target macroinvertebrate species, which may lead to misguided and failed restoration efforts (Bond and Lake, 2003; Feld et al., 2011). Moreover, alleviating local instream stressors may sort little effect if the causes of instream stress act on a larger scale, in the stream valley or in the upstream parts of the catchment (Death and Collier, 2010; Leps et al., 2015). Hence, there is a strong need to quantify the joint pressure of the multiple stressors acting instream as well as on

the catchment scale, acting on macroinvertebrate assemblages in lowland streams.

Recently, an increasing number of studies investigated the effects of two or a few stressors on stream assemblages at the mesocosm and field scales (Davis et al., 2018; Elbrecht et al., 2016; Waite et al., 2019). However, until now, analysing the joint impact of multiple stressors in a spatially explicit assessment context has been restricted to the risk assessment of the impact of human activities on ecosystems on global and regional scales (Clark et al., 2016; Halpern et al., 2008; Landis et al., 2017). These approaches had in common that stressor intensities and habitat vulnerabilities are quantified to calculate a cumulative impact score. Thereby, these studies have shown that analysing the joint impact of multiple stressors can be used to gain insight into the reasons for the observed low ecological water quality. However, these previous approaches did not consider all relevant stressors, lacked empirical justification of stressor weights and therefore largely relied on expert judgements. Moreover, these approaches focused on regional scales, having application areas other than lowland streams, such as coastal areas and large rivers (Clark et al., 2016; Halpern et al., 2008; Landis et al., 2017). The influence of hydrological connectivity on the distribution of stress within stream networks was therefore not included. Hence, no fully suitable stressor quantification method is available that considers the set of most important potential stressors, spanning multiple spatial and temporal scales, acting on macroinvertebrate assemblages in lowland streams. The aim of this study was to develop a novel method to quantify the cumulative stress acting on macroinvertebrate assemblages in lowland streams. To this purpose, we 1) identified

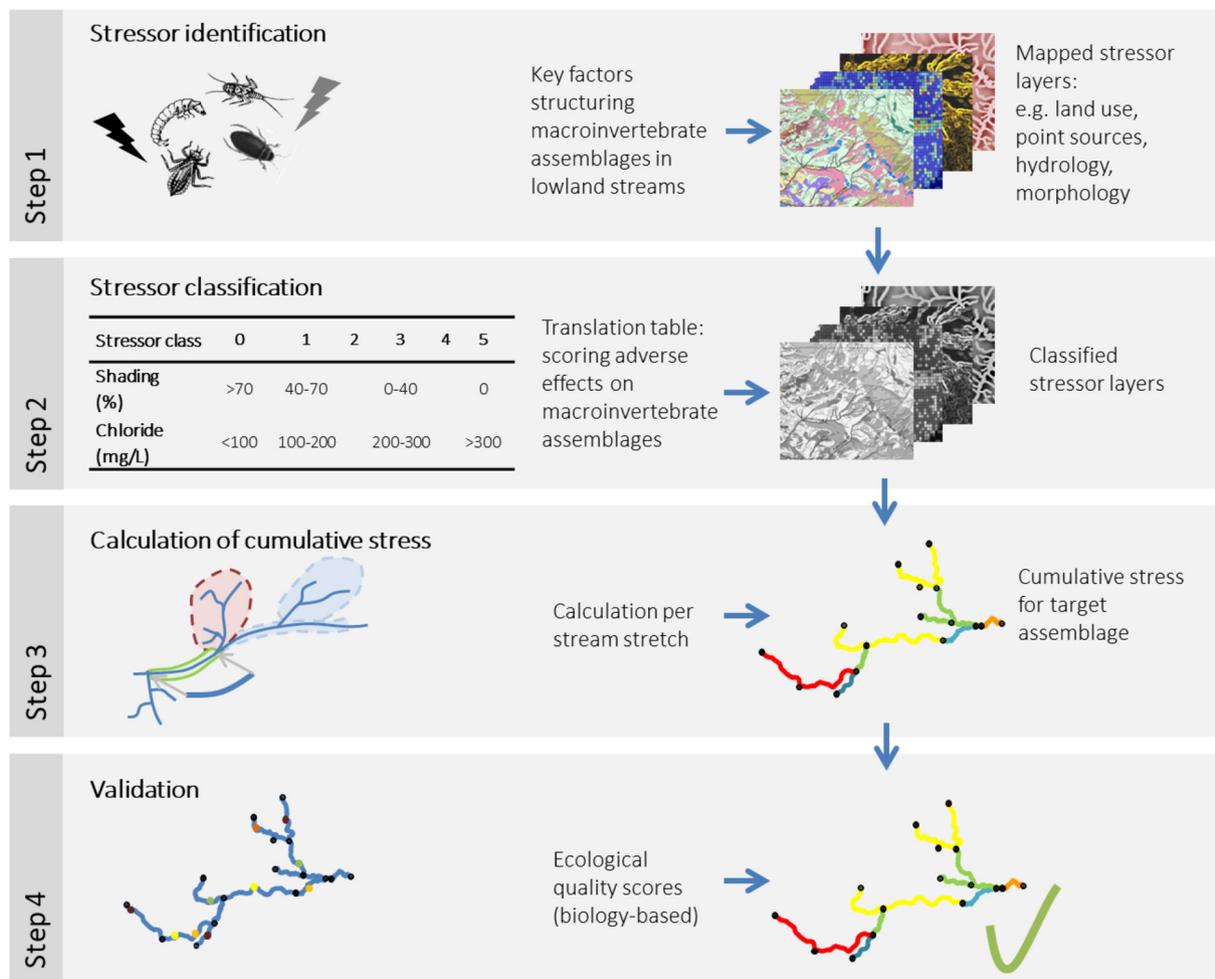


Fig. 1. Overview of the cumulative stress quantification method: 1) stressor identification for macroinvertebrate assemblages, 2) stressor classification based on the impact on macroinvertebrate assemblages, 3) calculation of cumulative stress per stream segment, 4) validation with independent ecological quality scores.

stressors acting on multiple scales relevant for macroinvertebrate assemblages in lowland streams, 2) quantified the adverse effects of the identified stressors, 3) calculated the cumulative stress, and 4) validated the cumulative stress-scores, using independent macroinvertebrate-based quality scores.

By considering the combined effects of stressors acting instream as well as on the catchment scale, and by quantifying their joint ecological impacts on the macroinvertebrate assemblages, this method can be used to identify the most important (combinations of) stressors that limit the distribution of macroinvertebrate assemblages in a specific area. In turn, this allows prioritizing of the actions needed to restore macroinvertebrate diversity in lowland streams.

The design of the cumulative stress quantification method is described in the next section, going through each of the four main steps. To validate the method, this is followed by the application of the cumulative stress quantification method in the catchment of the lowland stream Tungelroyse Beek in the Netherlands. To this purpose, the four main steps are applied for this specific case study.

2. Design of the cumulative stress quantification method

The cumulative stress quantification method consisted of four steps (Fig. 1): 1) stressor identification, based on key environmental factors structuring macroinvertebrate assemblages in lowland streams, 2) stressor classification according to the impact of these stressors on macroinvertebrate assemblages using tables that translate the impacts into stressor classes, 3) calculation of cumulative stress-scores for predefined stream segments, 4) validation of the resulting stress-scores using independent, macroinvertebrate-based quality scores.

2.1. Stressor identification

Stressors should be identified for each specific macroinvertebrate assemblage, having specific environmental preferences and sensitivities. We considered assemblages rather than communities, as the latter are not easily defined as a static entity along an environmental gradient (Gleason, 1939; Nijboer, 2006), whereas assemblages are considered to be the set of species occurring at a certain site at a certain moment

(Nijboer, 2006). According to these considerations, in the Netherlands, target macroinvertebrate species assemblages formulated for water quality assessments are indeed water type and region specific and are derived from reference or best-available sites (Van der Molen et al., 2016).

The selection of stressors was based on the key environmental factors that directly determine the presence of organisms at a specific site (Frissell et al., 1986; Feld and Hering, 2007; Verdonschot et al., 2000a, 2000b; Verberk et al., 2012). The key environmental factors were chosen to represent the following categories: system conditions (environmental factors acting at high spatial and temporal scales, related to climate and geology), chemistry, stream morphology, stream hydrology, biology and stream maintenance operations (e.g. mowing of water vegetation) (Verdonschot et al., 1998) (Tables 1 and A1).

Drivers of the selected stressors may act on scales varying from instream to the entire catchment. Instream habitat shapes assemblages at a local scale, for example by hydromorphological parameters such as substrate composition and flow variability. On the other hand, local chemical water quality and discharge are factors determined by processes at the catchment scale (Leps et al., 2015). For example, there is a strong relationship between the nutrient concentrations in surface waters and the surrounding land use (Boyer et al., 2002; De Wit, 1999). For these larger-scale environmental factors, catchment-wide data coverage is required. This way, variability between single point measurements caused by processes acting locally can be averaged out.

2.2. Stressor classification

Stressor classification was carried out to make stressors that act on different spatial scales and that are expressed in different units mutually comparable and to allow summation of their impact. The stressors were classified using a table translating the intensity of a stressor to an impact on a macroinvertebrate assemblage (as in Clark et al., 2016), by categorizing the stressor intensities into impact-based classes (Fig. 1, Table A1). These classes represented the increasing adverse effects on the macroinvertebrate assemblage, ranging from 0 (not causing stress) to 5 (high stress). For each of the categories (system conditions, chemistry, stream morphology, stream hydrology, biology and stream

Table 1

Selected stressors allocated to stressor categories and spatial compartments, with the type of classification into stressor classes (qualitative or quantitative boundaries). See Table A1 for complete table and references.

Spatial compartment	Category	Stressor	Qualitative (L)/quantitative (N)	References	
Riparian zone and headwater catchments	Chemistry - diffuse sources	Nutrients	L	1, 2, 3	
		Silt	L	4	
		Toxic substances	L	5, 6	
		Nutrients	N	7	
		Silt	L	8, estimation	
		Toxic substances	N	9	
	Chemistry - Point sources - sewer overflow	Nutrients	N	10, 11, 12	
		Silt	L	8	
		Toxic substances	N	9	
		Chloride	N	13	
		Shading	N	14	
		Streambed drying	N	15	
Main stream	System conditions	Peak flows	N	16	
		Seepage	L	14	
		Weir presence, stagnation	L	17	
		Natural longitudinal profile	N	17	
		Natural lateral profile	N	17	
		Bank form	L	17	
	Stream morphology	Bank fixation	L	17	
		Connectivity	N	18	
		Mowing of aquatic vegetation	N	14, 17, estimation	
		Invasive species	L	19	
		Stream maintenance operations			
		Biology			

maintenance operations, Table 1), the reasoning behind the stressor selection and the establishment of the classification is argued below.

For some environmental factors no information was available on assemblage-specific impacts. In these cases, the impact on macroinvertebrates in general was scored, based on impact studies or estimations (Table 1).

2.2.1. Chemistry - diffuse sources

In agricultural areas, diffuse inputs are often a major nutrient source for surface waters (Drewry et al., 2006). Other important ecological stressors related to land use are siltation (dos Reis Oliveira et al., 2018) and the input of toxicants associated with run-off (Reichenberger et al., 2007). Land use-related stressors were chosen over the incidental instream chemical measurements available from monitoring networks, because the latter only give a temporal and spatial snapshot of the chemical status, whereas land use temporally and spatially integrates stress levels and is a strong predictor of nutrient loading (Allan, 2004; De Wit, 1999).

For each land use type, the nutrient, silt and toxicant input into surface waters was deduced from empirical studies (Table 1). Based on these values, each land use type was appointed a stressor class according to the general impact on macroinvertebrates (Table A2).

2.2.2. Chemistry - point sources

Water treatment plants, sewer overflows and industrial discharges are point sources that contribute to high stress levels when they contain nutrients, silt, chloride and toxic substances. For each point source, the nutrient content and the amount of silt and toxic substances were scored as stressors. For nutrients, the impact classes were based on volume, load or frequency of sewer overflows per year as a proxy of nutrient content (Elbersen-van der Straten and Van den Bomen, 2002). For point sources other than sewer overflows, the classes were based on the amount of nutrients in the inflowing water, where the class boundaries were taken from eutrophication studies of Leentvaar (1979), Vollenweider (1968) and Wegl (1983). The stressor classes for toxic substances were appointed by comparing toxicant concentrations to predicted no effect-concentrations for specific species within macroinvertebrate assemblages (ECHA, n.d.). Class boundaries for the adverse impact of chloride were taken from a classification system for brackish water (Venice system, 1958).

2.2.3. System conditions

Climate is one of the most important system conditions, driving temperature and light patterns. Canopy shading is a local factor that acts positively on the ecological status of streams, among others by buffering water temperature by decreasing light irradiation, increasing organic matter inputs and providing habitat structure (Feld et al., 2018, 2011). This factor can become a stressor when absent or only scarcely present considering shaded lowland streams as reference conditions. Shading of the stream was accordingly classified into five stressor classes based on the proportion of shaded area. These classes were based on a macroinvertebrate environmental preference dataset linked to a previously constructed and independent macroinvertebrate-based typology of the region (Dutch stream typology; Verdonschot et al., 2000a).

2.2.4. Stream hydrology

Hydrological factors that potentially act as a stressor within the main stream are low and high flow impacts on organisms. These affect macroinvertebrate assemblages through interference with feeding and respiration, and may cause mortality resulting from desiccation during low flows and drift during high flows (de Brouwer et al., 2017; Rolls et al., 2012). Potential hydrological stressors with indirect effects are loss of habitat through siltation or bed scouring and a lack of seepage on locations where this would have occurred under reference conditions (Hart and Finelli, 1999). Flow requirements were derived for macroinvertebrates in general (Driver, 1977; Verdonschot and van den Hoorn,

2010). Stressors used for this category can be seen in detail in Tables 1 and A1.

2.2.5. Stream morphology

As morphological stressors acting within the main stream, longitudinal and lateral stream profile, bank form and the presence of bank fixation were identified. For example, natural banks provide necessary structures for macroinvertebrate assemblages, like habitat in the form of hollow banks and tree roots, and habitat elements like wet-dry gradients and associated vegetation (Flecker and Allan, 1984; Garcia et al., 2012; Tolkamp, 1980). High habitat diversity may mitigate the effects of multiple stressors such as low flow and fine sedimentation (Graeber et al., 2017). These morphological structures can act as stressors when they are in a non-natural state or even absent. Ranges for the stressor classes were derived from a morphological quality assessment index (Rinaldi et al., 2013). In addition, a decreased connectivity within the stream network may act as a stressor (van Puijenbroek et al., 2019). The stress classes representing decreased connectivity were expressed as the number of dams and weirs per stream segment (Tables 1 and A1).

2.2.6. Stream maintenance operations

The impact of mowing of the instream and bank vegetation and other forms of stream maintenance operations by water managers on macroinvertebrate assemblages, including channel dredging, was based on the maintenance frequency (Tables 1 and A1). Because little is known about the relationship between the intensity of vegetation management and stream macroinvertebrate assemblages, the level of stress posed on the system was directly related to the maintenance frequency (Beltman, 1987).

2.2.7. Biology

Biological factors that may act as a stressor are the presence of non-indigenous species including crayfish, fish and plant species, considered as pests. When these play a dominant role, they may alter aquatic ecosystem structure and functioning (Crowl et al., 2008). As the effects on the macroinvertebrate assemblages were not known in detail, only the presence of invasive species was scored as a stressor (Tables 1 and A1).

2.3. Calculation of cumulative stress

2.3.1. Spatial compartments

To calculate the cumulative stressor scores, the main streams were divided into series of stream segments. These segments were delimited

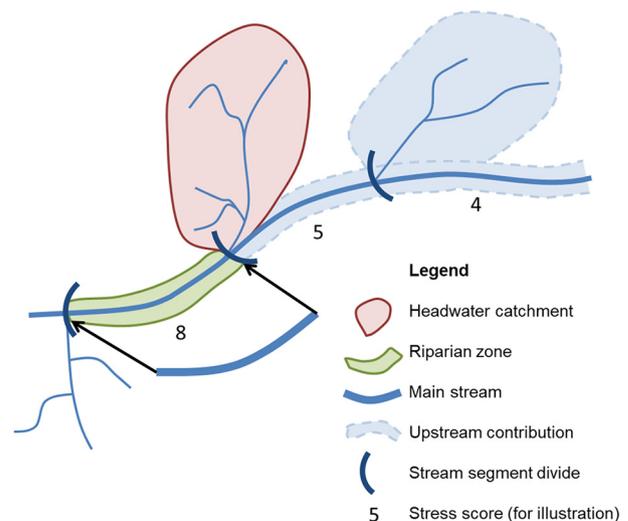


Fig. 2. Schematic overview of the spatial compartments defined in the cumulative stress quantification method for the main stream.

Legend

- Monitoring points morphology and biology
- Point sources - sewer overflows
- ▲ Segment divide
- Main stream and stream valley area
- Secondary streams
- Catchment area

0 5 Kilometers

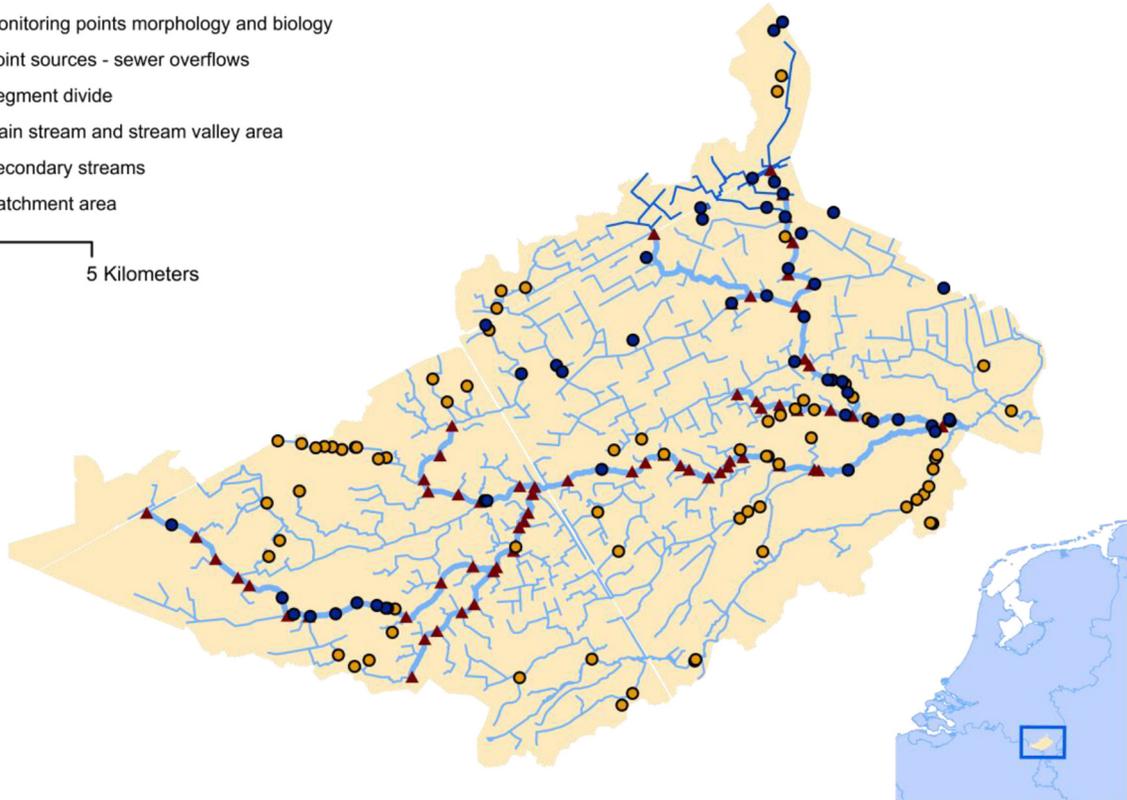


Fig. 3. Overview of the Tugelroyse Beek catchment, available monitoring data points and segment delimitation. Flow direction is to the right.

by the inflow points of two subsequent headwater catchments. As a consequence, the stream segments varied in length.

Three spatial compartments around each stream segment were defined: the main stream, the headwater catchments entering the main stream, and the riparian zone consisting of the area adjacent to the stream segment (Fig. 2).

2.3.2. Stress contribution from headwater catchments

The stressor scores for chemical stressors from diffuse and point sources originating from the headwater catchments were averaged over each headwater catchment. The stressor contribution to the main stream was corrected for relative discharge of the headwater catchment at the point of entry compared to the discharge of the main stream according to Eq. 1:

$$S_h = \text{mean}(C_h) * \frac{Q_h}{Q_m}, \tag{1}$$

where S_h = total stress-score for headwater catchment, C_h = stressor-class per stressor within headwater catchment, Q_h = discharge of headwater catchment, Q_m = discharge of the main stream at point of entry.

2.3.3. Stress contribution from riparian zones

The riparian zone was delineated as a zone of 350 m on both sides of the main stream (Allan, 2004; Morley and Karr, 2002). In this area, stress originates mainly from land use and point sources. To correct for the variable size of stream segments and therefore of the riparian zones, the contribution was multiplied by the relative surface area of the riparian zone compared to the total area of the riparian zones in the catchment according to Eq. (2).

$$S_r = \text{mean}(C_r) * \frac{A_r}{A_t}, \tag{2}$$

where S_r = total stress-score for riparian zone, C_r = stressor-class per stressor within riparian zone, A_r = surface area of riparian zone, A_t = total surface area of riparian zones in catchment.

2.3.4. Stress contribution in the main stream

Scores of stressors acting in the main stream were averaged per segment and corrected for the relative length of that specific segment. Stream gradient was included as an additional factor to include the

Table 2
Overview of available data for stressors in the Tugelroyse Beek catchment.

Category	Stressor	Data availability and quality
Chemistry	Diffuse sources: nutrients, silt, toxic substances	Entire area, 25 m grid cell size
	Point sources: nutrients, silt, toxic substances	Full coverage of catchment, partly only presence, partly also volume, load and frequency known. Estimation made for missing values.
System conditions	Shading	47 locations along main streams
Stream hydrology	Streambed drying	Main streams, from simulation
	Peak flows	Main streams, from simulation
	Seepage	Main streams
Stream morphology	Weir presence, stagnation	Main streams
	Natural longitudinal and lateral profile	47 locations along main streams
	Bank form	47 locations along main streams
	Bank fixation	47 locations along main streams
	Connectivity	Main streams
Stream maintenance operations	Mowing	Main streams
	Biology	Invasive species

mitigating effect of a high mean flow velocity on the total stress (Withers and Jarvie, 2008) according to Eq. (3):

$$S_s = \left(\left(\text{mean}(C_s) * \frac{l_s}{l_t} \right) / G, \right) \quad (3)$$

where S_s = total stress-score for main stream, C_s = stressor-class per stressor acting in main stream, l_s = length segment, l_t = total stream length, G = stream gradient class (1–3, with 3 for a high gradient segment, see Table A3 for exact class boundaries).

2.3.5. Cumulative stress quantification

Stressor scores were appointed to each of the three spatial compartments around each stream segment. The total stress for a given segment was calculated by adding the total cumulative stress of the previous segment, the headwater catchments and the riparian zone to that of the main stream. To account for retention of stressors when transported downstream, such as nutrient absorption (Birgand et al., 2007; Triska et al., 2019), stressor contributions from headwater catchments and upstream segments were halved according to Eq. (4):

$$S_{i+1} = 0.5 * S_i + S_s + S_r + 0.5 * S_h, \quad (4)$$

where S =Cumulative stress-score with subscript i for segment i , s for stress acting in main stream, r for stress from the riparian zone, h for stress from the headwater catchments.

The stressor contribution at confluences of stems of the main stream was weighed by the relative discharge of the individual stems. As opposed to the stress score per individual stressor, which ranges from 0 to 5, the cumulative stress score has a minimum of 0 and a maximum depending on the size of the catchment.

For illustrative purposes, a stepwise approach was followed to document the cumulative stress score calculation. First, stress scores were calculated per stressor category and spatial compartment only. Later, stress addition from upstream segments, retention of stressors in upstream segments and dilution at stream confluences was included to show cumulative stress scores per segment.

2.4. Validation of the cumulative stress quantification method

The resulting cumulative stress per stream segment was compared to independent, macroinvertebrate-based quality scores derived from biological samples collected at multiple sites within the catchment. To test whether there was a relationship between the calculated cumulative stress and monitored ecological quality scores, a linear regression was performed in R (R Core Team, 2018).

3. Application of the cumulative stress quantification method

To test the applicability of the cumulative stress quantification method, the cumulative stress per stream segment was calculated for a test catchment, the Tungalroyse Beek in the Netherlands.

3.1. Study area

The Tungalroyse Beek catchment is located in the south of the Netherlands (51° 14' 42" N, 5° 53' 10" E). The major stream within the catchment is the stream Tungalroyse Beek, which is mainly groundwater fed, with a mean discharge of 1.0 m³/s (drainage area 157 km²). In the past, the stream was positioned within extensive bogs and marshlands, but over the years the landscape and flow regimes have been changed to fit the demands of agriculture, resulting in a strongly modified and over most of its length channelized stream. The main land use in the catchment is agriculture, covering 64% of the surface area, followed by 20% of natural and 16% urban land use (Fig. A1).

According to the presently developed cumulative stress quantification method, the main streams within the catchment were divided into segments, which were delimited by the inflow of the headwater catchments (Fig. 3).

3.2. Stressor identification

3.2.1. Data collection

The data required to quantify the selected stressors was collected within regular monitoring programs of the local water authority (Waterboard Limburg). To quantify the diffuse sources, land use data was taken from the LGN7 map, providing the land use types in 2012 (Hazeu et al., 2014). Data on hydrological stressors was not available for the entire catchment. Therefore, the hydrological stressor layers were derived from a steady-state hydrological simulation (Simgro, based on discharge data from 1996 to 2011), giving the yearly-averaged flow velocity and the number of high and low flow events. Morphological parameters were recorded for 47 locations (each location representing a stretch of 50–100 m) along the main streams. For each segment, the monitoring point for which most data was available was chosen as representative for the entire segment. The five most recent entries, covering a variable period of about 5 years, were selected to calculate the stress-scores.

For some stressors, no or insufficient information was available (Table 2). This was the case for the stressors from the stressor group point sources within the category chemistry. For these stressors, the stress acting on the macroinvertebrate assemblages was estimated based on the known volume of similar point sources in the catchment to be able to include the set of most important stressors.

3.3. Stressor classification

The stressor classification was carried out as described above in the general method description (Section 2). This included using assemblage-specific impact classes where this was possible. If assemblage-specific impact classes could not be made, stressors were classified based on the general impact on macroinvertebrates as referenced in Table A1.

3.4. Stepwise approach towards the cumulative stress calculation

A two-step approach was followed to illustrate the cumulative stress score calculation. First, stress scores were calculated per stressor category (chemistry, system conditions, stream hydrology/stream morphology, and biology/stream maintenance operations) and spatial compartment (main stream, headwater catchments and riparian zone). Hence, in this first stressor score calculation, stress addition from upstream segments, retention of stressors in upstream segments and dilution at stream confluences were not included yet.

In the second step of the calculation, these category- and spatial compartment-specific scores were used to calculate the cumulative stress scores per segment according to the cumulative stress quantification method outlined above (Section 2.3).

3.5. Validation of the cumulative stress calculation

Macroinvertebrate-based ecological quality scores (EQR) of the period 2008–2018 at 28 locations within the main streams within the catchment were used for validation of the cumulative stress calculation. These quality scores were based on abundance and diversity data obtained from the local water authority and calculated using the Dutch standard assessment method (Van der Molen et al., 2016). The calculated scores were log-transformed before analysis to improve normality.

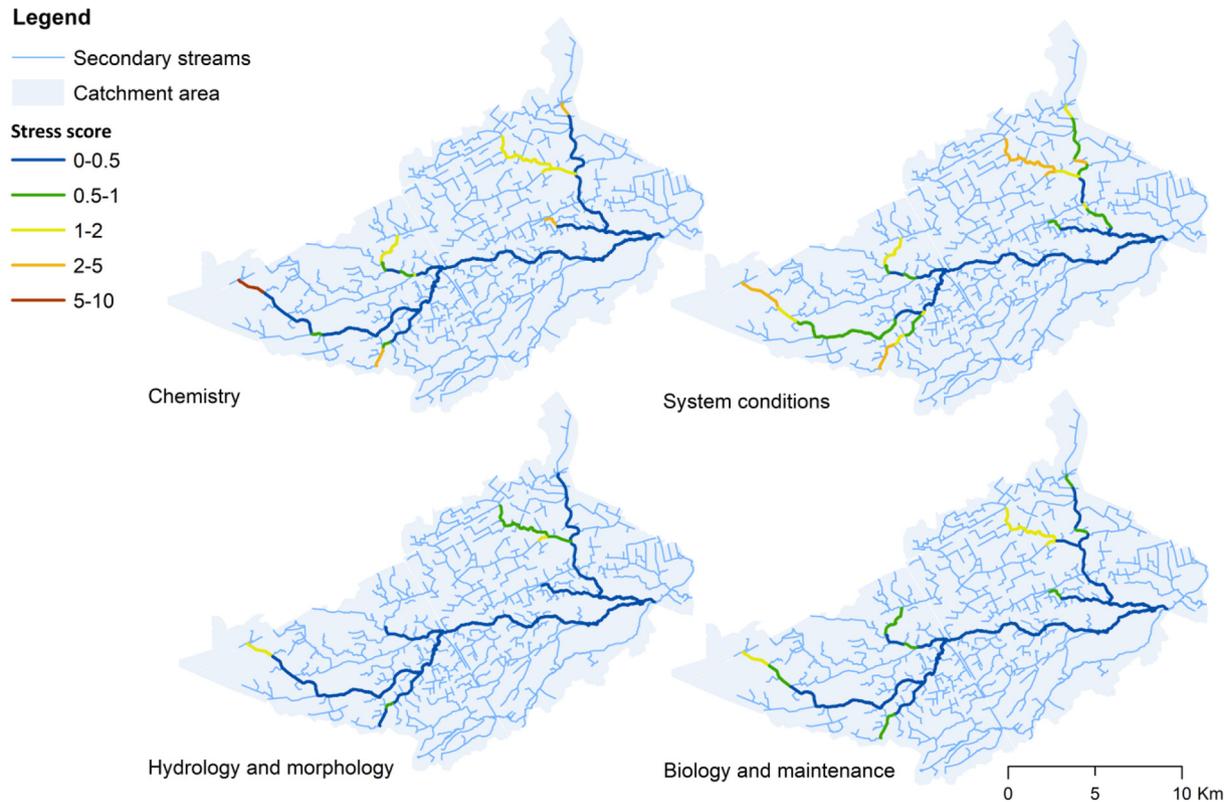


Fig. 4. Calculated stress scores for the separate stressor categories chemistry, system conditions, stream hydrology/stream morphology, and biology/stream maintenance operations. Scores range from blue (low stress) to red (high stress). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

3.6. Results of the application of the cumulative stress quantification method

The stress scores calculated were higher in the upstream segments, for the categories chemistry and system conditions, whereas most downstream parts of the main streams showed the lowest stress scores (Fig. 4). Especially for the categories hydrology/stream morphology and biology/stream maintenance, calculated stress scores were low in the entire catchment. For the stressor lack of shading in the category system conditions, locally high stress scores were calculated.

Next, cumulative stress scores were calculated per defined spatial compartment (headwaters, riparian zone and the main stream), by summing the contribution of all stressors, showing considerable variation over the entire catchment area (Fig. 5). Stress scores for the headwater catchments were generally moderate. Locally, high scores were calculated at the far north and northwest parts of the catchment, based on land use and point sources. For the riparian zone compartment, scores ranged from very low stress levels to a cumulative stress score of 2 on a range of 0 to 10. Higher stress scores were observed for the main stream, with stress scores locally falling in a high class, indicating severe levels of stress.

The final calculated cumulative stress per stream segment, combined from the compartment-specific cumulative stress scores as described in the cumulative stress quantification method above (Section 2.3), decreased in downstream direction (Fig. 6): The highest stress scores were found in the headwaters of the main streams, whereas the lowest stress scores were found near the stream mouth, where the stream drains into the river Meuse. The monitored ecological quality scores showed a variable pattern, with generally lower quality scores upstream and higher quality scores downstream.

Comparing the spatial distribution of the ecological quality scores and the calculated cumulative stress resulted in a significant negative relationship between the EQR and the logarithmized calculated

cumulative stress (Fig. 7; $R^2 = 0.57, P < 0.001$), although there was considerable scatter. This negative relationship demonstrates that the cumulative stress quantification method indeed reflected the impact of environmental and anthropogenic stress on the macroinvertebrate assemblage reasonably well.

4. Discussion

4.1. Case study: spatial stressor patterns and identification of bottlenecks for improvement of water quality in the stream Tungalroyse Beek

The cumulative stress quantification method was used to gain insight into the spatial distribution of stressors impacting macroinvertebrate assemblages in the lowland stream Tungalroyse Beek. A rather atypical pattern was observed where the highest calculated cumulative stress and lowest ecological quality scores were found in the headwater segments, whereas downstream, the highest ecological quality scores and lowest cumulative stress scores were recorded.

Concerning the spatial compartment of the headwater catchments, a clear contributor to the high stress scores could be identified locally. On the catchment scale, in the calculated cumulative stress scores, this local contribution to a high stress score remained evident. In addition, it was shown how multiple stressors combine when taking the spatial connectivity into account, including stressor addition from upstream segments, retention of stress in upstream segments and dilution at stream confluences. These processes are relevant, because the influence of stressors may extend to downstream segments, influencing ecological water quality not only locally. The cumulative stressor calculation therefore gives a more realistic insight into stressor patterns than when considering local stressors only.

In the present case study, the high cumulative stress scores in the headwater segments were caused by the presence of a zinc smelter in the upper north western branch, which discharges treated industrial

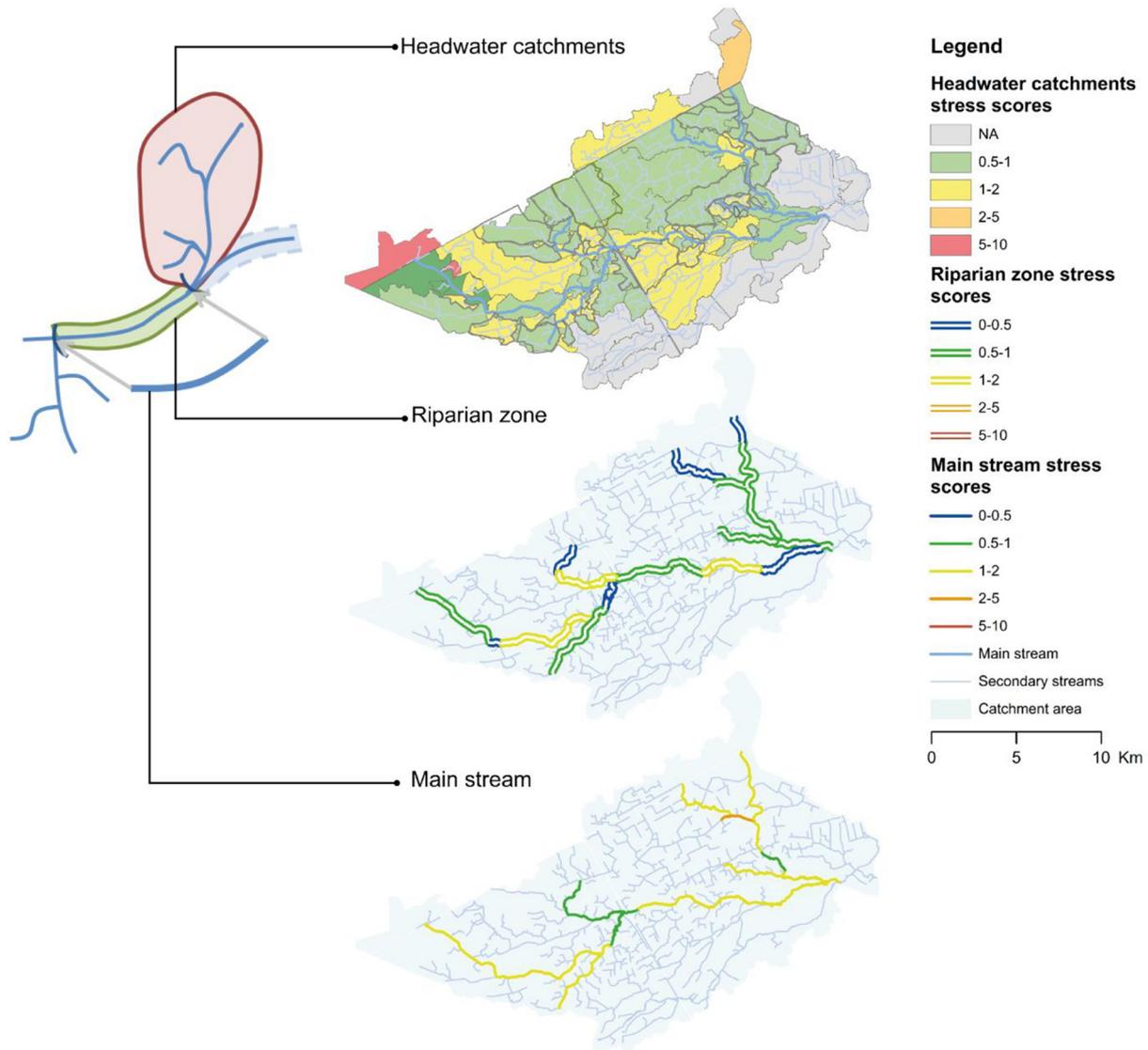


Fig. 5. Calculated stress-scores for the separate spatial compartments headwaters, riparian zone and main stream. Scores range from blue (low stress) to red (high stress). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

waste water into the stream. This might be a bottleneck for reaching a higher ecological water quality in the upper stream segment. In the downstream segments, a higher flow velocity and shading of the streambed compensated the stress originating from the upstream segments, while also toxicological and nutrient-related stress from point and diffuse sources was diluted, resulting in lower cumulative stress scores. With the newly developed cumulative stress quantification method, it was thus possible to identify the stressor with the highest contribution within the segments with high cumulative stress. This way, the environmental factor which is most limiting a good ecological water quality could be identified, showing that the method has a diagnostic potential.

The findings in our case study did not reflect patterns in water quality that are often described for lowland streams and rivers, where increasing inputs from agricultural and urban areas impact water quality more and more in downstream direction (e.g. Glińska-Lewczuk et al. 2016; Usseglio-Polatera & Beisel 2002). In the present case-study we observed the opposite. We identified that local processes were of importance determining this pattern, such as the presence of point sources upstream and a diluting effect with shaded conditions downstream. These compensating effects of shading have been described before for streams (Ghermandi et al., 2009). Even though a

pattern of increasing water quality in downstream direction is not typical of lowland streams, the stress pattern agrees with monitored quality scores (Fig. 7), which supports the approach followed in the cumulative stress quantification method.

More scatter is present in the left-hand side of Fig. 7. This is what one would expect, as in situations with high ecological quality, the potential stressors that may have been missed in the calculation will have a stronger influence on the calculated cumulative stress, whereas potentially missed stressors might change the calculated stress less in situations which have already a low ecological quality.

4.2. Cumulative stress quantification method

Quantifying the multiple stressors acting on macroinvertebrate assemblages is fundamental to understanding the reasons for their presence and absence (Jackson et al., 2016; Ormerod et al., 2010). A number of challenges arose when developing the cumulative stress quantification method, like dealing with the spatial scale of action of the different stressors and quantifying the impacts on specific macroinvertebrate assemblages.

Previous efforts have provided insight into the reasons for low ecological water quality by analysing the joint impact of multiple stressors.

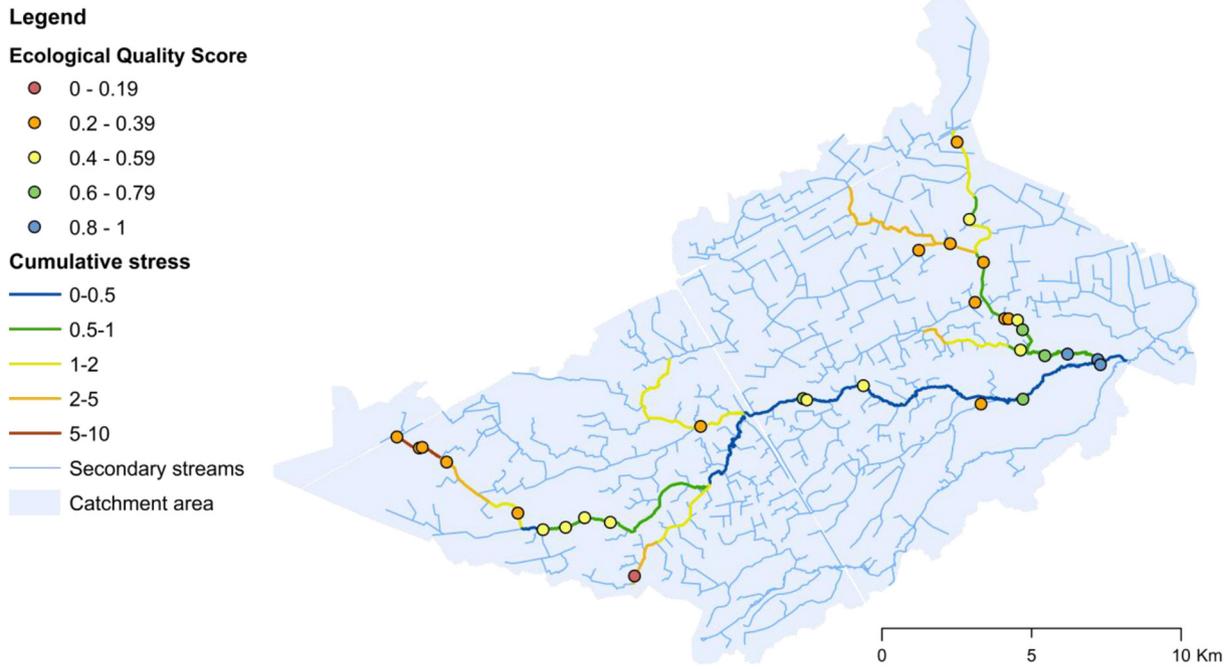


Fig. 6. Cumulative stress-scores (calculated) and Ecological Quality Scores (monitored) used for validation. Scores range from blue (high quality, low stress) to red (low quality, high stress). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

Landis et al. (2017) calculated the risk from multiple stressors grouped in chemical, ecological and exposure stress, but their assessment did not represent the complete set of relevant environmental stressors. Other stressor quantification methods focussed on ecosystems on larger spatial scales, with different hydrological connectivity, and therefore did not make a distinction in spatially connected compartments (Halpern et al., 2009). In addition, stressor weights were based on expert judgement instead of empirical data (Clark et al., 2016; Selkoe et al., 2009).

Adding to these previous methods, we aimed for considering the set of most important stressors acting on multiple spatial and temporal scales, acting on macroinvertebrate assemblages in lowland streams. Therefore, in the current study, this gap was addressed in the design of the cumulative stress quantification method. To this purpose, a study area is subdivided into short stream segments of about 500–1500 m, with each three spatially connected compartments for stressor quantification, in which all relevant stressors for

macroinvertebrate assemblages are included that act on these different spatial scales. This spatial distinction of stressors is especially useful if the study region has a high spatial connectivity through the stream network, and local stressors may pose pressure on downstream areas. Also, the stressor weights in our method are mostly based on empirical evidence from literature.

It is still under debate at which scale stressors on a local macroinvertebrate assemblage are best reflected. The importance of landscape structures and processes acting at regional scales on ecological water quality has been emphasized (Lake et al., 2007; Poff, 1997; Roth et al., 1996; Stoll et al., 2016), but monitoring of water quality and the formulation of measures is often still based on local measurements (De Pauw et al., 2006; Goethals and de Pauw, 2001). However, such local monitoring data have the disadvantage that they only provide a snapshot of the ecological status in time and space, whereas the nutrient concentrations in the water may be variable due to temporal dynamics and discontinuously discharging point sources (Withers and Jarvie, 2008). Also, measurements are taken in the water column, which for example may hold lower nutrient concentrations compared to the sediment (Birgand et al., 2007), where benthic macroinvertebrates live. Therefore, relationships between local water quality measurements and regional water quality assessments may not be clear (Smith et al., 2010). In this study, the choice was made to represent nutrient-related stress regionally by using stress scores based on land-use type, as this was thought to average out local variability and to integrate chemical stress over space and time. The calculated stress scores are corroborated by the monitored ecological quality scores, which supports our choice for using land use and point sources to represent nutrient-related stress.

Furthermore, quantifying the impact per stressor is a challenging task, as knowledge concerning stressor-response relationships is limited for specific organism groups. Previous impact assessments dealt with this issue by quantifying the impact on an ecosystems-scale (Clark et al., 2016; Halpern et al., 2009), or by excluding certain stressors with known significant impacts, but for which data was unavailable (Halpern et al., 2008; Selkoe et al., 2009), making such impact assessments incomplete. To deal with data gaps and to still be able to include

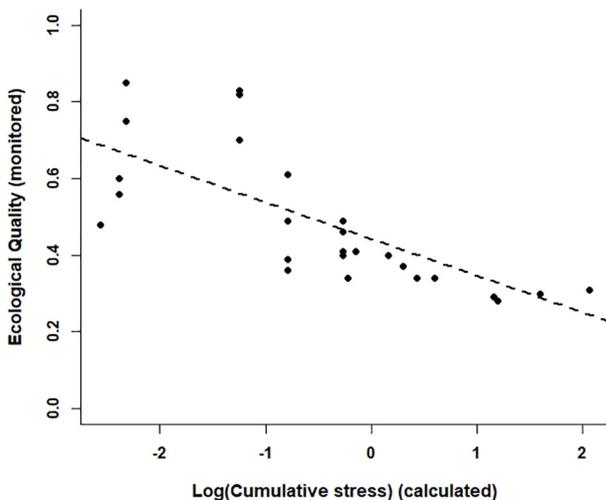


Fig. 7. Relationship between monitored ecological quality scores and calculated cumulative stress scores ($R^2 = 0.57$, $P < 0.001$, $a = 0.44$, $b = -0.096$).

relevant stressors for which the exact size or distribution was unknown, in the current application, proxy data, interpolating from existing data or estimations were used. In addition, in the design of the present cumulative stress quantification method, we aimed at classifying the impact on a macroinvertebrate assemblage. In the future, our cumulative stress quantification method could be further refined considering the species-specific stressor-response relationships for classifying the impact on macroinvertebrates and by improving the quantification for impact parameters that are now still based on estimations.

There is a number of assumptions that underlie cumulative impact assessments in general (Halpern and Fujita, 2013) of which some also apply to the cumulative stress quantification method presented here. First, stressor layers were assumed to be of equal importance. Second, a consistent ecosystem response based on a combination of stressors was assumed for the entire catchment. Also, an additive model was used, which possibly results in over- or underestimating the cumulative stress scores. The cumulative stress scores may change when synergistic or antagonistic interactions between stressors are included in the calculation (Piggott et al., 2015; Schäfer and Piggott, 2018). A possible synergistic interaction is the combined effect of elevated nutrient concentrations and hydromorphological stress (Lemm and Feld, 2017), a possible antagonistic interaction is the combined lower stress levels for *Daphnia* exposed to a toxicant at low temperatures (Folt et al., 1999).

Our results represent the currently best estimate of the cumulative stress acting on macroinvertebrates in lowland streams. Although the calculated scores were partly based on estimations of the assemblage-specific impact, their combination in the cumulative stress calculation still gave insight on the most stringent stressors limiting presence of macroinvertebrate assemblages. The cumulative stress quantification method may be further developed by expanding the species-specific impact standardisation, implementing calculation schemes for different water types, incorporating stressor interactions and using more detailed data. Thereby, the method can be further validated by application to multiple catchments.

4.3. Use of the cumulative stress quantification method to define measures for lowland stream restoration

The case study showed that it was possible to identify those stressors that contributed most to the cumulative stress per stream segment, supporting the design of the cumulative stress quantification method. This opens the possibility to use our approach to simulate the effect of management scenarios by setting hypothetical stress scores from theoretically implemented measures, for the water type and for the specific macroinvertebrate assemblage for which the method was parametrised. This way, the cumulative stress quantification method can show if selected measures indeed alleviate multiple stressors, as opposed to measures targeting single stressors. For example, strips of riparian forest along streams increase, among others, shading, provide organic matter, decrease the need for instream vegetation management and provide more variation in bank structure. The cumulative stress quantification method may be used to quantify the effects of such measures on the stressors acting on macroinvertebrate assemblages in lowland streams. This would be especially useful in view of targeting restoration efforts of water managers.

5. Conclusions

Our approach provides a method for quantifying the cumulative stress acting on macroinvertebrate assemblages in lowland streams. Although applied to a single catchment and a single group of organisms, it is shown that the presently developed method increases the understanding of the reasons for the absence and presence of macroinvertebrate assemblages, especially by identifying the contribution of individual stressors to the total stress at a given location. Applying our

newly developed cumulative stress quantification method, management resources can be better prioritized to restore macroinvertebrate diversity in deteriorated lowland streams.

Author contributions

PV, RV and JV designed the methodology and acquired and analysed the data. JV wrote the main manuscript text. All authors were involved in the study design, contributed to the writing of the manuscript and gave final approval for publication.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.scitotenv.2019.133630>.

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