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# Identification and interaction of multiple stressors in central European lowland rivers



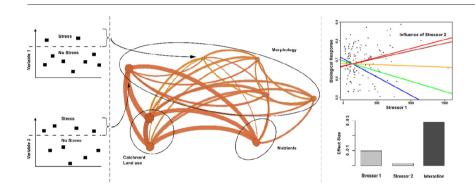
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#### HIGHLIGHTS

- Multiple stressors impact the ecological status of freshwaters, with often unknown interactions
- A common methodology to identify prominent multiple stressor combinations in survey data is lacking.
- We present an approach to identify cooccurring and interacting stressors and their effects on invertebrate responses.
- The approach is applicable across various kinds of ecosystem types and organism groups.
- The outcome helps ecosystem managers infer management measures, when stressors require concerted management efforts.

#### GRAPHICAL ABSTRACT



#### ARTICLE INFO

Article history:
Received 16 August 2016
Received in revised form 6 June 2017
Accepted 11 June 2017
Available online xxxx

Editor: D. Barcelo

Keywords:
Multiple stressors
Benthic macroinvertebrates
Species traits
Interactions
Social network analysis
Generalised linear models

#### ABSTRACT

Interactions of multiple stressors in lotic systems have received growing interest and have been analysed in a growing number of studies using experiment and survey data. In this study, we present a protocol to identify, display and analyse stressors of rivers and their interactions (additive, synergistic or antagonistic). We used a dataset of 125 samples of central European lowland rivers comprising hydromorphological, physico-chemical and land use stressor and pressure variables as well as benthic macroinvertebrate traits as biological response variables. To identify and visualise multiple stressor combinations jointly operating in the data set, we applied social network analysis. The main co-occurring stressor combination was fine sediment accumulation (hydromorphological stress) and enhanced phosphorus concentration (nutrient stress). Agricultural (cropland) and urban land use were identified as the main large scale environmental pressures. Stressor interactions were analysed using generalised linear regression modelling (GLM) including pairwise interaction terms. Altogether, 14 macroinvertebrate response variables were tested on six stressor combinations and revealed predominantly additive effects (80% of all significant models with absolute standardised effect sizes > 0.1). Significant antagonistic and synergistic interactions occurred in almost 20% of the models. Fine sediment stress was more influential and frequent than nutrient stress. The methodology presented here is standardisable and thus could help inform practitioners in aquatic ecosystem monitoring about prominent combinations of multiple stressors and their interactions. Yet, further understanding of the mechanisms behind the biological responses is required to be able to derive appropriate guidance for management. This applies to rather complex stressors and pressures, such as

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land use, for which more detailed data (e.g. nutrient concentrations, fine sediment entry, pesticide pollution) is often missing.

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#### 1. Introduction

Multiple stressors impact aquatic ecosystems, the biodiversity, and ecosystem services they provide (Hering et al., 2015). In Europe, there are regional differences in number and combinations of stressors, however, multiple stressors are evident at nearly half of the 120,000 water bodies of lakes, rivers, transitional, and coastal waters (EEA, 2012). Stressors of hydromorphological degradation (e.g. stagnation, fine sediment, habitat alteration, straightening) and eutrophication (enhanced nitrogen and phosphorus concentrations) are among the most prominent threats of aquatic ecology and eventually ecological status of Europe's freshwaters (Hering et al., 2015; Nõges et al., 2016). Other, often unmeasured or even unknown stressors may accrue (e.g. pesticides, salinisation, pharmaceuticals, micro-pollutants), with likewise unmeasured or unknown effects on the aquatic fauna and flora. More importantly, the simultaneous operation of two or more known stressors can trigger combined effects, i.e. stressor interactions (Folt et al., 1999). The presence of such interactions should be one reason for ineffective ecosystem management (e.g. Townsend et al., 2008) due to unpredictable effects on the biological communities (Piggott et al., 2012, 2015).

Disentangling multiple stressor effects is one of the growing research topics in riverine ecology at present (Ormerod et al., 2010; Hering et al., 2015). There is considerable evidence of multiple stressor interactions from freshwater ecosystems (Matthaei et al., 2010; Wagenhoff et al., 2011, 2012, 2013; Piggott et al., 2012), which suggest the operation of both synergistic (i.e. combined effect > sum of individual effects) and antagonistic (i.e. combined effect < sum of individual effects) effects on fish, benthic invertebrates, and benthic algae. Much of this evidence originates from experimental (flume) studies, that allow to control the number of stressors and their intensities. Experiments allow to elicit causal links of stressors and biological response indicators. As well, there is a growing number of studies using survey data from reach to catchment scale concerning about interactions between the focal stressors. The body of studies includes a reach-scale experiment replicated at the stream scale (Townsend et al., 2008), several stream surveys about agricultural stressors in New Zealand (Hofmann et al., 2016; Lange et al., 2016; Macher et al., 2016; Lange et al., 2014a, 2014b; Wagenhoff et al., 2011), a study on urban stressors in the Melbourne area using boosted regression trees on a large data set of rapid bioassessment sites (Walsh and Webb, 2016), and a European wide study investigating four stressor groups on fish assemblages at 3105 sites in 14 European countries (Schinegger et al., 2016). Under such uncontrolled survey conditions, including way more variables acting than measured, as opposed to experiments, disentangling multiple stressors effects is likely to be more challenging. Nevertheless, the tremendous asset of recent WFD monitoring data (EEA, 2012) provides an unprecedented opportunity of multiple stressor analysis at the pan-European scale.

Here, we present a multiple stressor analysis of river survey data taken during two EU-funded research projects (www.aqem.de: Hering et al., 2004 and www.eu-star.ac.at: Furse et al., 2006) between 2000 and 2002. We introduce social network analysis (SNA), a methodology that originally was developed to disentangle social relationships between human individuals (Wasserman and Faust, 1994). Ban et al. (2014) have applied SNA in an ecological context to identify the structure of multiple stressors of coral reefs based on a literature review. SNA can be used to reveal a structure of co-occurring multiple stressors in a dataset. It constitutes a tool to intuitively visualise stressors inter-

relationships by quantifying and plotting the presence of multiple stressors exceeding thresholds at similar samples of a dataset.

The aim of our study is to present a methodological framework to analyse multiple stressor effects on biological indicators based on data from field surveys. In addition to the cookbook recently provided by Feld et al. (2016), we initially put focus on the detection of pressures and stressors co-occurrence and the analysis of the most common combinations of stressors jointly operating in our data. We then quantify interactive stressor effects on selected benthic invertebrate indicators as suggested by previous studies. We compare the biological indicator's response to the most prominent stressor combination and close with some final remarks on the implications for freshwater ecosystem management and conservation.

#### 2. Material and methods

#### 2.1. Stressor data and biological response variables

We used data of 144 samples from lowland rivers in the Netherlands, Germany, Poland (Feld et al., 2013), and some additional samples from Sweden (Dahl et al., 2004). All samples were taken between 2000 and 2002 and comprise an extensive set of environmental variables including land use, hydromorphology, sediment cover, physicochemistry, and natural covariates (see Feld and Hering, 2007 for details on sampling methodology and data preparation). All sites are representing sand-bottom lowland rivers (Pottgiesser and Sommerhäuser, 2008). Here, we used a portion thereof representing potential pressure and stressor variables (Table A.1). Based on the DPSIR framework, our catchment scale land use variables are representing pressures and the reach scale variables are representing states, here called stressors.

To facilitate comparability, samples taken at sites with a catchment area < 8 km<sup>2</sup> were excluded, which resulted in 125 samples remaining for further analysis. Land use data for the whole upper catchment was derived from CORINE Land Cover (Statistisches Bundesamt, 1997), whereas all other stressor variables were recorded during macroinvertebrate sampling in the field or taken from maps (Feld, 2004; Hering et al., 2004). Information of sediment cover was sampled in the field with the AQEM macroinvertebrate sampling protocol (AQEM Consortium, 2002). One sample at a site consists of a 5% coverage of the whole sampling reach. If one sediment fraction was representative for 3 samples, there are 15% coverage at site and sample. Barbour et al. (1999) developed this method with respect to the sampling of major proportionally distributed microhabitats. We considered fine sediment fraction (<2 mm) to be a possible stressor, although it is ubiquitous in lowland rivers. However, high amounts of fine sediment can change the diversity of the microhabitat composition and can therefor cause stress to the benthic invertebrate communities.

Macroinvertebrates were sampled using multi-habitat sampling of 20 representative microhabitat units (unit size:  $25 \times 25$  cm), all of which were sampled along a stretch of 50–100 m using a  $25 \times 25$  cm square-frame handnet (mesh size:  $500\,\mu m$ ) (Hering et al., 2004). Sampling took place in three seasons, predominantly spring, but summer and autumn as well. Distributions of non-biological and biological variables were checked with grouped boxplots for each variable and season. Considering all seasons extended the gradient of the reach-scale non-biological variables and therefore boosted the variance of stressor data with benefits for the modelling process. After determination, the taxon lists (mainly at species level) have been used to calculate an

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