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The potential of chironomid larvae-based metrics in the bioassessment of non-wadeable rivers

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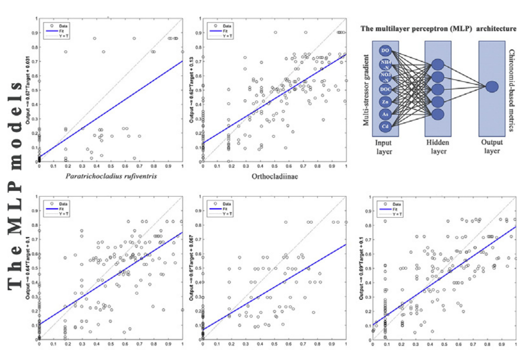
HIGHLIGHTS

- We tested chironomid community metrics for bioassessment of large rivers.
- The neural network revealed that community reacted to multi-stress gradient.
- The multilayer perceptron derived 5 models as reliable metrics for bioassessment.
- Metrics based on chironomids of large rivers can be used in multimetric approach.

GRAPHICAL ABSTRACT



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ABSTRACT

The chironomid community in non-wadeable lotic systems was tested as a source of information in the construction of biological metrics which could be used into the bioassessment protocols of large rivers. In order to achieve this, we simultaneously patterned the chironomid community structure and environmental factors along the catchment of the Danube and Sava River. The Self organizing map (SOM) recognized and visualized three different structural types of chironomid community for different environmental properties, described by means of 7 significant abiotic factors (a multi-stressor gradient). Indicator species analysis revealed that the chironomid taxa most responsible for structural changes significantly varied in their abundance and frequency along the established environmental gradients. Out of 40 biological metrics based on the chironomid community, the multilayer perceptron (MLP), an supervised type of artificial neural network, derived 5 models in which the abundance of *Paratrichocladius rufiventis*, Orthocladiinae, *Cricotopus* spp., *Cricotopus triannulatus* agg. and *Cricotopus/Orthocladius* ratio achieved a significant relationship (the r Pearson's linear correlation coefficient > 0.7) with the multi stressor environment. The sensitivity analysis "partial derivatives" (PaD) method showed that all 5 biological metrics within the multi-stressor gradient were mostly influenced by dissolved organic carbon (DOC). Despite short and monotonous environmental gradients and the absence of reference conditions, the chironomid community

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structure and biological metrics predictably changed along the multistress range, showing a great potential for the bioassessment of large rivers.

1. Introduction

When flowing through densely populated areas, non-wadeable rivers are exposed to multiple stressors including extreme environmental changes. This huge spectrum of environmental factors causes a net effect on the aquatic biota which is almost unknown to the scientific community (Jackson et al., 2016). Dams, channelization, timber harvesting, agriculture and urbanization cause severe changes to the major part of the catchment area of large rivers (Bere et al., 2016; Chiu et al., 2017; Dalu et al., 2017). In addition, the pollutants that come from all of these stressors are more rapidly diffused because of their high discharge, which is characteristic for large rivers (Allan and Castillo, 2007). The intricate interactions of anthropogenic activities complicate the effort of defining the cause-effect relationship between abiotic and biotic components and can undermine the interpretation of these relationships in the context of bioindication (Pan et al., 2012). Those extensive, long-lasting activities and resulting modifications of the environment make it almost impossible to find intact natural habitats (Birk et al., 2012). This presents a great obstacle for all bioassessment methods which are based on the reference condition approach. Another problem for the bioassessment approach in water management is the multinational catchments of big rivers, which often spread through different countries, with different national assessment protocols defined by their water management authorities. Furthermore, defining a biological indicator on a local scale (i.e. surveying the catchment within one country) can be problematic and result in error as it is necessary to measure and include a wide range of gradients for this process (Blocksom and Johnson, 2009).

For these reasons the development of the bioassessment protocols for large rivers has been substantially delayed in relation to those constructed for smaller lotic systems. Though there are well developed protocols for the ecological status assessment of wadeable streams and smaller rivers in Europe, which are in accordance with the Water Framework Directive (WFD) requirements, large rivers have been mostly neglected. There have only been a small number of studies testing how the biotic component behaves in the whole catchment area of big rivers (e.g. (Angradi et al., 2009; Birk et al., 2012; Blocksom and Johnson, 2009; Pan et al., 2012). The aforementioned researchers tried to harmonize different assessment approaches and to define a unique sampling technique and data analysis design for non-wadeable rivers (Birk et al., 2012).

A big breakthrough in the bioassessment research of aquatic ecosystems was the multimetric approach. It is realized through multimetric indices, which are commonly used in routine monitoring programs for freshwater and brackish water ecosystems in Europe (Hering et al., 2006) and the United States (Barbour et al., 1999; Davis and Simon, 1995; Hughes et al., 1998; Karr and Chu, 1998; Stoddard et al., 2008). This method simplifies complex biological data in the form of individual metrics, but a sufficient amount of information regarding the ecosystem health is kept. After the selection and calibration processes, the most suitable metrics, based on composition/abundance, richness/diversity, sensitivity/tolerance or functional group information are used to calculate the final score of the multimetric index, which can assess the overall condition of a site (Hering et al., 2006). In this approach, the biotic assemblages most commonly used are macroinvertebrates and fish. However, despite the fact that some groups of invertebrates, such as the Chironomidae family are often the most frequent, diverse and abundant taxonomic group in a water-body, consequently having an important role in the ecosystem's functioning, they have not yet been used for constructing metrics.

Within the AQEM (2002) and STAR (Standardisation of River Assessment Methods, www.eu-star.at) research projects >300 different metrics have been tested as descriptors of ecosystem health, however, none of these relies on chironomid community data. The main reason for this trend is the time-consuming identification process which can also be problematic or even impossible at the larval stage of some taxa groups (e.g. *Orthocladius*, *Cricotopus*, *Tanytarsus*). Also, the Chironomidae family shows a huge natural variability along spatial and temporal gradients, which usually masks variability caused by anthropogenic stressors (Milošević et al., 2013).

In this study, we attempted to test the potential of the chironomid community to provide acceptable information for the construction of metrics for non-wadeable rivers. To achieve this goal, we set the following tasks: (1) to pattern the chironomid community structure together with the environmental parameters along the catchment of the Danube and Sava River, (2) to test the relationship between the biotic (chironomids) and abiotic (natural and man-caused) variables in order to select the most influential suite of environmental parameters responsible for changes in the community structure, and finally, (3) to use the multistressor gradients to test the sensitivity of chironomid based metrics as well as their potential to predict the overall conditions of the sites tested.

2. Material and methods

2.1. Study area and sampling

The research included two big European rivers, the Danube and its largest tributary – the Sava River (Liška et al., 2015; Navarro-Ortega et al., 2015). With a length of 2860 km and a river basin covering an area of approximately 817,000 km^2, the Danube is the second largest European river. It flows into the Black Sea in Romania and Ukraine after passing through 10 countries from its headwater in the Black Forest in Germany. Many large and small cities have been built along the Danube's watercourse, thereby changing its hydromorphology, degrading the water quality and negatively influencing aquatic biota.

The Sava River has a 97,713 km^2 catchment area and plays a significant role in the whole Danube basin. It is 945 km long and flows through Slovenia, Croatia, Bosnia and Herzegovina and Serbia, where it flows into the Danube in Belgrade. There is great anthropogenic pressure along the whole Sava River basin in the form of hydroelectric power plants, industry, oil refineries, agriculture and untreated sewage, which influence the hydromorphology, water quality and biodiversity of the river (Milačić et al., 2015).

Macroinvertebrate sampling was conducted at 68 sites along the Danube between August and September 2013 (Fig. 1). In addition, a survey of the macroinvertebrate community along the Sava River was conducted in September 2015 at 15 sites, from Croatia to Serbia (Fig. 1). The section of the Sava River between Jasenovac and Belgrade (confluence to the Danube) is a typical large non-wadeable river. The upstream stretch could be considered as transitional, since it is somewhere between a typical large non-wadeable stream type and sub-alpine river (Lucić et al., 2015). Thus, only samples taken down-stream from Jasenovac were considered for this analysis (Fig. 1).

At each sampling site, prior to macroinvertebrate sampling, several physical and chemical parameters were estimated in order to define the water quality gradient: water temperature (T), conductivity (C), dissolved oxygen (DO), pH, the concentrations of ammonia nitrogen (NH4-H), nitrate nitrogen (NO3-N), orthophosphates (PO4-P), and dissolved organic carbon (DOC). In addition, to define the degree of heavy metal pollution, the concentrations of dissolved copper (Cu), zinc (Zn), nickel (Ni), lead (Pb), arsenic (As), chromium (Cr), mercury (Hg) and cadmium