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Editorial

Ecological Monitoring and Assessment of Freshwater Ecosystems: New Trends and Future Challenges

Konstantinos Stefanidis [1,2,*] and Eva Papastergiadou [1,*]

1 Department of Biology, School of Natural Sciences, University of Patras, University Campus Rio, 26500 Patras, Greece
2 Hellenic Centre for Marine Research (HCMR), Institute of Marine Biological Resources and Inland Waters, 46.7 km of Athens—Sounio Ave., 19013 Attiki, Greece
* Correspondence: kstefani@upatras.gr (K.S.); evapap@upatras.gr (E.P.); Tel.: +30-2291076439 (K.S.); +30-261096925 (E.P.)

1. Introduction

Freshwater ecosystems, particularly rivers and lakes, are under severe pressure due to increasing anthropogenic activities, such as water extraction, flow regulation, pollution, and habitat fragmentation [1–3]. Local, regional, and global drivers of environmental change (e.g., land cover transformation, pollution, the introduction of invasive species, and climate change) are responsible for the loss of many freshwater biota and ecosystem functions all around the world [4,5]. The global biodiversity crisis is more acute in freshwater ecosystems than in any other ecosystem. The current rate of wetland loss is three times that of forest loss and almost 27% of freshwater species are threatened with extinction [5]. Furthermore, human-driven changes greatly impact the delivery of ecosystem services, affecting the well-being of humans. Hence, introducing new conservation management and restoration measures is mandatory to improve biodiversity, ecological quality, and the supply of clean water and other ecosystem services to humans. Over recent decades, extensive national and international regulations have been adopted to protect water resources. In Europe, the biological monitoring, assessment methods, and classification systems in use have been greatly improved by the EU Water Framework Directive (WFD 2000/60) through monitoring programs based on species composition and abundance [6,7]. The goal of the Water Framework Directive is to restore or maintain the ecological state of the freshwater systems that are present across all of the EU member states. Thus, the WFD provides very detailed guidelines for the implementation of ecological monitoring and the assessment of all European inland and coastal waters [6,7].

Ecological monitoring is essential for understanding an ecosystem's functions and dynamics. The collection of biological and environmental data has greatly improved our capacity to understand the impact of many anthropogenic activities on biotic communities and the overall health of an ecosystem. Traditionally, ecological monitoring is based on extensive field surveys to acquire information about the diversity and composition of species' communities and their relationships with the environment. Significant advancements in monitoring have created novel methods and have delivered new tools that further increase the efficiency of data collection and reduce the associated costs. The use of wireless sensor network arrays (e.g., camera traps, acoustic sensors) promotes real-time monitoring with high-frequency measurements over large spatial scales [8]. The analysis of satellite images is another source of high-quality biotic and abiotic data with wide applications in ecological studies including freshwater ecosystems [9]. Recently, function-based assessments (species traits) and molecular methods (eDNA-based bioassessment) have been proposed to complement or even replace current monitoring methods [10–14].

This Editorial refers to the Special Issue "Ecological Monitoring and Assessment of Freshwater Ecosystems: New Trends and Future Challenges". The wide scope of the Special

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Issue aims to highlight new research findings and significant advances concerning all aspects of bioassessment and the processes that occur within aquatic ecosystems. Emphasis was placed on contributions that explored the dynamics and functioning of freshwater ecosystems, developed new methods for monitoring and assessing ecological quality, studies that included the use of biotic metrics or indices, environmental DNA methods, experimental studies, and those that promoted the use of modelling approaches.

Twelve articles were finally accepted for publication from a total of twenty submissions that were considered for the SI. The twelve selected studies cover four continents of the world (Europe, North America, Africa, and Asia) and present results within the scope of the SI that expand our current knowledge on various topics, including the use of freshwater biota as indicators of environmental change, the application of models for predicting biological parameters, and the use of eDNA methods for monitoring invasive species (Table 1).

Table 1. Analysis of the contributions published in this Special Issue.

No of Contribution	Focus on Biotic Component/Ecosystem Process	Type of Research	Spatial Scale/Geographical Coverage
1	Chlorophyll-a concentration/eutrophication	Modelling	Ecosystem/Greece
2	Aquatic macrophtes/responses to salinization and increased irradiance	Experiment/Ecophysiology	Ecosystem/Habitat/Greece
3	Riverine habitats	Assessment method/index development	Catchment/China
4	Fish/predator effects	Experiment	South Africa
5	Phosphorus and nutrient losses at catchment scale	Modelling	Multiple catchments/Europe
6	Zebra and quagga mussels/eDNA assessment	eDNA assessment	Ecosystem/the USA
7	Aquatic macrophytes/ecological quality index	Assessment method/index development	Multiple ecosystems/Greece
8	Meiofauna/responses to salinization	Field study	Catchment/Vietnam
9	Cyanobacteria/study on akinetes	Biochemical/molecular study	Organism/South Korea
10	Water quality and biological quality indices	Field study	Catchment/South Korea
11	Macroinvertebrates and fish/responses to pollution	Field study	Multiple ecosystems/South Africa
12	Freshwater sponges/responses to pollution	Field study	Multiple ecosystems/Serbia

2. Main Messages of the Special Issue and the Book

The current Special Issue and the contributions within it discuss many themes related to aquatic diversity that focus on the monitoring of ecological quality, experimental studies, modelling, and the decline in species and also contribute to conservation and preventing future loss of freshwater biodiversity. The first article, by Hadjizolomou et al., examines the potential to optimize artificial neural networks (ANN) for predicting the chlorophyll-a concentration in lakes with limited field data. The authors found that the ANN's performance is greater when the leave-one-out (LOO) cross validation method is used and increases with the k-fold number. They also investigated the effects of the input parameters on the prediction of Chl-a concentration by conducting a sensitivity analysis, and they found that changes in conductivity and water temperature caused a higher % of changes in the predicted outcome. Based on their results, the authors concluded that ANN models can be a useful tool for predicting chlorophyll-a, and potentially other lake variables, even when data scarcity is an issue.

The next contribution was written by Malea et al., who experimentally investigated the physiological responses of the submerged plant *Stuckenia pectinata* to high levels of salinity and irradiance. Their results highlighted the plant's significant photo-acclimation potential, which could be used to regulate the number and size of its reaction centres and photosynthetic electron transport chain through the dissipation of excess energy into heat. They also found that the interaction between salinity and irradiance had a significant effect on the plant's Chl-a, b contents, which may indicate its potential ability to acclimatize by adjusting the Chl a, b contents. However, they did not report significant impacts on the relative growth rate, which could mean that the plant may become acclimatized by reallocating resources to compensate for growth. Thus, the authors conclude that the regulation of photosynthetic pigment content and photosystem II performance comprised the plant's primary growth strategy within the high salinity and irradiance conditions that are likely to occur due to eutrophication and future climatic change.

The third article, by Yu et al., deals with the use of satellite imagery for assessing the habitat quality of mountain streams in the Chishui basin in China. This study employs a series of metrics based on water environmental status, river morphology, riparian zone, and human disturbance, combined with stream order, elevation, slope, and sinuosity, to classify habitats into types. The habitat assessment was conducted with the use of the habitat quality index (CHQI) and the results indicated that the headwaters of three rivers (Tongmin, Tongzi, and Xishui) have been impacted by anthropogenic activity. The authors conclude that their habitat assessment method can be used for further biomonitoring in other mountain river systems as well.

The fourth contribution, by Munyai et al., comes from South Africa and investigates the effect of two predator fish, *Oreochromis mossambicus* and *Enteromius paludinosus*, on Chironomidae prey. The study uses a comparative functional response approach to assess the interactions of the two predator species when they are feeding on a readily consumed prey within multiple predator scenarios. The findings of this article reported that each species displayed a significant Type II FR, characterized by high feeding rates at low prey densities. *Oreochromis mossambicus* had a steeper (initial slope, i.e., higher attack rate) and higher (asymptote of curve, i.e., shorter handling time and higher maximum feeding rate) FR, whereas *E. paludinosus* exhibited lower-magnitude FRs (i.e., lower attack rate, longer handling time, and lower feeding rate). In multiple predator scenarios, the feeding rates were predicted well by using those of single predators, both in conspecific and interspecific pairs, and thus the authors did not find evidence for antagonistic or synergistic multiple predator effects (MPEs). The results from this study, although experimental, improve our current knowledge about how trophic interactions among conspecific or interspecific fish species in Austral tropical wetlands might influence their aquatic prey species.

The article by Wade et al. is the fifth contribution to the SI and examines the relative importance of the climate and land use drivers in nutrient loss in nine study catchments in Europe and a neighbouring country (Turkey). Catchment-scale biophysical models were applied within a common framework to quantify the integrated effects of projected changes in the climate, land use (including wastewater inputs), N deposition, and water use on the quantity and quality of river and lake water in the mid-21st century. The proposed land use changes were derived from catchment stakeholder workshops, and the assessment quantified changes in mean annual N and P concentrations and loads. The main finding of this study was that, at most of the sites, the projected effects of climate change alone on nutrient concentrations and loads were small, whilst land use changes had a larger effect and were of sufficient magnitude that, overall, a move to more environmentally focused farming achieved a reduction in N and P concentrations and loads despite the projected effects climate change. However, at Beyşehir lake in Turkey, increased temperatures and lower precipitation reduced water flows considerably, making climate change, rather than more intensive nutrient usage, the greatest threat to the freshwater ecosystem. Individual site responses did, however, vary, and were dependent on the balance of diffuse and point source inputs. The simulated changes in the chlorophyll-a content in the lake were not generally proportional to changes in nutrient loading. Thus, further work is required to accurately simulate the extremes in flow and water quality and determine how reductions in freshwater N and P translate into an aquatic ecosystem response.

The next article by Marshall et al. presents and evaluates new eDNA assays that target the extended repeat sections of zebra and quagga mussels. These assays lower the limit of detection of genomic DNA by 100-fold for zebra mussels and 10-fold for quagga mussels. Additionally, these newly developed assays facilitated longer durations of detection during degradation mesocosm experiments and a greater sensitivity for the detection of eDNA in water samples collected across western Lake Erie compared to standard assays that target mitochondrial genes. Finally, this study illustrates how important it is to understand the complete genomic structure of an organism in order to improve eDNA analyses.

The seventh contribution, by Stefanidis et al., presents a methodological approach for the implementation of a WFD-compliant macrophyte index in the riverine systems of Greece and the results from the pilot application of the index. The study analyses the methodological framework for defining the stressor gradients and the least disturbed sites along with the reference conditions that are required for the derivation of the ecological quality classes. It also includes the classification of the river reaches into five quality classes that were derived from the application of the Macrophyte Biological Index IBMR for Greek rivers ($IBMR_{GR}$). The main findings showed that hydromorphological modifications were the main environmental stressors and that they were strongly with the correlated $IBMR_{GR}$, while physicochemical stressors were of lesser importance. In addition, the ecological assessment showed that almost 60% of the sites failed the WFD target of a "Good" ecological quality class, which agrees with classification assessments that were based on other BQEs for Greece and many other Mediterranean countries. Overall, this work provides the first assessment of the ecological classification of Greek rivers using the BQE of aquatic macrophytes, which has significant implications for ecological monitoring and decision making within the framework of the implementation of the WFD.

Nguyen et al. contributed to the SI with an article that assesses the impact of salinity variations on riverine ecosystems with a particular focus on the responses of meiofauna to salinization along the Van Uc River continuum in Vietnam. The main findings of this study were that the meiofaunal richness indices were higher in the estuary and slightly decreased upriver. Nematoda was the most dominant taxon at the salty stations, while Rotifera was more abundant at the less salty ones. The results from a multiple variate analysis indicated a strong interplay among salinity, nutrients, and pore water conductivity, which shaped the meiofaunal distribution. The inclusion of pore water salinity, nutrients, and meiofaunal community structure indicated that there was a greater extent of the saline ecosystem in the estuary than previously thought, posing a greater risk to freshwater salinization. Hence, the contribution by Nguyen et al. highlights the potential role of meiofauna as a bioindicator but also calls for a reformation of salinity assessment for better freshwater conservation and management.

The next contribution, by Kim et al., analyses the akinete structure, as well as akinete-specific proteins and their amino acid sequences, of the cyanobacterium *Dolichospermum circinale*. The akinetes were produced from vegetative cells isolated from the North Han River, Korea, while the akinete protein was obtained using electrophoresis, and its antibody-binding reaction potential (ig-score) was quantified. The authors found that the homology of the *D. circinale* akinete-specific protein was very low (9.8%) compared to that of *Anabaena variabilis*, indicating that its composition was substantially different, even among phylogenetically close taxa. Overall, this article represents the first known report on the *D. circinale* akinete protein and its amino acid sequence and offers insights into their practical application for detecting akinetes in freshwater systems.

The tenth article is by Lee et al., also from South Korea, and investigates the effects of land use on stream water quality and biological conditions in sub-watersheds and micro-watersheds across the Han River. By employing random forest models, the authors found that water quality and biological indicators were significantly affected by forest areas at both scales, and the sub-watershed models performed better than the micro-watershed models. The effects on the water quality and biological indicators were similar regardless of the scales, although the relationship between land use and stream conditions was slightly more sensitive in the micro-watersheds than in the sub-watersheds. In addition, their results showed that urban and agricultural areas showed a lower proportion of water quality and biological condition variability in the micro-watersheds than in the sub-watersheds, while the forests showed the opposite results. Hence, the authors concluded that the spatial scale matters when developing effective watershed management strategies to maintain stream ecosystems.

The next study, by Munyai et al., examined the relationships between environmental parameters and biotic communities, with an emphasis on the effects of water and sediment quality parameters on the macroinvertebrate and fish communities in three subtropical reservoirs in South Africa. The results of their redundancy analysis and two-way ANOVA showed that there were significant differences among the reservoirs, and they identified four water variables (water temperature, oxidation–reduction potential, pH and conductivity) and one sediment metal (Mg) as the most important parameters driving the structure of the fish community. In addition, high concentrations of metals in the sediment of one of the three reservoirs (Nandoni) suggested that anthropogenic activities have significantly influenced the sediment quality, with severe implications for the ecological conditions. Finally, the authors propose the need to adopt measures that improve the conservation of these ecosystems.

The final contribution, by Andjus et al., assess the frequency of spicule malformations in freshwater sponges in relation to selected environmental parameters of the streams and the presence of river pollutants. The authors conducted a morphological analysis using light and scanning electron microscopy, and recorded the number of anomalies (spicules with bulbous enlargements that are sharply bent, bifurcated, scissor- and cross-like, and t-shaped). The results reported single- and double-bent spicules as the main types of anomalies. An important finding was that the authors found statistically significant differences in the concentrations of ammonia, orthophosphates, sodium, chloride, manganese, and lead between the sites with the lowest and the highest numbers of anomalies. Thus, this study indicates that several pollutants could be responsible for the occurrence of spicule anomalies.

This Special Issue is devoted to articles that present new and original research on topics focused on the ecological monitoring and assessment of freshwater ecosystems. More than half of the articles examine the relationships between environment and freshwater biota at various scales, from the organism level to the community and ecosystem levels (Table 1). These studies focused on various biotic groups, including aquatic macrophytes (contributions No 2 and 7), freshwater sponges (contribution No 12), fish (contribution No 4 and 11), and invertebrates (contributions No 8 and 11). In addition, some articles explored new methods and research techniques for ecological monitoring, such as contribution No 6 which assesses novel eDNA assays for zebra and quagga mussels, contribution No 9 which deals with the use of akinete structure of cyanobacteria as indicator of environmental change, and contribution No 1 which focuses on using ANN models to predict Chl-a concentration in shallow lakes. The spatial scale of the studies varied from the local (ecosystem scale) to the regional (catchment scale) and transregional (multiple catchments) scales. Six of the articles include studies conducted at the local scale (one ecosystem or catchment), three of the articles concern studies at a wider scale (multiple ecosystems or catchments), and two of the articles were based on experimental research. Finally, one study involved the biophysical modelling of nine catchments from Europe and Turkey to investigate the effects of climate change and land use transformation on nutrient losses under different future scenarios.

3. Conclusions

Freshwater ecosystems cover less than 1% of the Earth's surface and yet are among the most diverse and threatened systems in the world [5]. The importance of freshwater ecosystems in maintaining biodiversity and ecosystem services is becoming increasingly clear, but, at the same time, freshwater ecosystems are highly vulnerable to human impacts such as climate change and land use changes. The collection of articles in this SI show that regional (e.g., land cover transformation, pollution, hydromorphological alterations, and invasive species) and global (e.g., climate change) environmental changes are responsible for the loss of many aquatic biota and ecosystem functions worldwide. Furthermore, there is a growing number of studies, and EU policies such as the WFD 2000/60 and the habitats directive, that highlight the need to improve current monitoring schemes and undertake conservation and restoration actions within inland waters.

In this context, future research will prioritize gaining a better understanding of how these changes can affect species, communities, functions, and ecosystem services by employing new methods and tools.

In the current SI, several topics were discussed which provide significant insights into the monitoring, conservation, and management of freshwater ecosystems. Catchment models, machine learning, eDNA assessments, and remote sensing are all tools that are gaining ground in ecological monitoring (see contributions 1,3,5,6). In addition, ecological studies based on field monitoring have begun to focus on less explored biotic groups, such as sponges and meiofauna, and ecophysiological responses, producing promising results with further implications for the ecological assessment of freshwater ecosystems (contributions 2,4,8,9,12). Finally, the development and testing of new ecological indices remain top priorities for freshwater ecologists (contributions 3 and 7) but also for managers who are called in to assess the impact of anthropogenic stressors on freshwater ecosystems.

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