

Multiple stressors in freshwater ecosystems: biocides and climate change

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Headlines

- Human-induced environmental stressors, such as chemical pollution and climate change, are having increasingly widespread and severe impacts on biodiversity and natural resources.
- These rarely occur in isolation, with ‘multiple stressors’ overlapping in time and space.
- Freshwaters, such as lakes, rivers and ponds, which provide us with food, recreation, flood defences and other valuable ecosystem services, are influenced by stressors from many sources (e.g. industrial chemical spills, diffuse agrochemical pollution and ultimately global warming), and are particularly vulnerable.
- The impact of multiple stressors is rarely simply the sum of their individual parts, with more complex interactions commonly occurring.
- Stressors can interact with each other directly (e.g. by altering one another’s severity), or indirectly through altered species sensitivity, or via changes in species feeding relationships within the food web.
- A new predictive framework is required for anticipating the combined impacts of stressors, and especially for biocides (e.g. pesticides and antibiotics) and climate change, which are both growing global threats.
- A collaborative effort is needed to combine the knowledge of on-the-ground conservationists, academic researchers, and policymakers to create tools that can cope with the complexity of multiple stressors in natural systems.
- An ecological networks approach could be used to detect both the immediate impacts of stressors (through the microbial community) and to predict responses across the wider ecosystem.

Contents

Glossary.....	2
Multiple threats to freshwater ecosystems...2	
Coping with ‘ecological surprises’ and the combined effects of stressors	3
Biocides and collateral damage via indirect impacts on food webs	4
Biocides and climate change.....	5
Recommendations for the management of freshwater ecosystems	7
References	7
Acknowledgements	10
About the authors.....	10

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Glossary

Additive effects: The combined impact of two stressors is equal to the sum of their independent effects; **non-additive effects are more** (a **synergistic effect**) or less (an **antagonistic effect**) than the sum of their independent effects.

Biocide: Chemicals designed specifically to target and kill particular pathogen or pest species, including insecticides, fungicides, antibiotics and disinfectants.

Bacteria biosensor: A species (or group) of bacteria which responds rapidly to a given biocide (e.g. by changes in its abundance and/or molecular make-up) and can therefore be used to confirm or detect the presence of said biocide.

Ecological network: A representation of species interactions, in which pairs of species (i.e. nodes) are connected when they are interacting directly (by links).

Ecosystem services: The benefits provided by ecosystems that contribute to making human life both possible and worth living. Examples of ecosystem services include products such as food and water, regulation of floods, and recreation in natural areas.

Freshwater ecosystem: Lakes, ponds, rivers, streams, springs and wetlands.

Multiple stressors: Human-induced stressors that overlap in time or space, such as chemical pollutants and climate change.

Multiple threats to freshwater ecosystems

Human activity is putting increasing pressure on our natural environment and human-induced impacts are becoming both more widespread and severe¹. This new generation of environmental stressors include chemical pollution and climate change, and among the former, 'biocides' are of particular concern. This paper considers these from their functional mode of action on organisms, rather than the varying and narrower regulatory or legislative definitions that often separate Plant Protection Products (e.g. agrochemical insecticides) from those used more for veterinary, medicinal or hygiene purposes (e.g. antibacterial disinfectants, preservatives or pest control substances).

In this framework, biocides include insecticides, herbicides, antibiotics and fungicides. In addition to increasing exposure to this rapidly diversifying chemical cocktail, our planet's climate is changing, with annual mean temperature rising and extreme events (e.g. heatwaves, drought flooding, hurricanes) becoming

increasingly prevalent². As such, chemical and climatic stressors rarely, if ever, occur in isolation and they are manifested over many scales in both time and space^{3,4,5}.

Freshwater ecosystems are particularly vulnerable to environmental stressors, and even the largest rivers and lakes are still relatively small and fragmented within a predominantly terrestrial landscape^{6,7}. They are also biodiversity hotspots of vital importance for providing essential ecosystem services, including drinking water, food and flood prevention: multiple stressors therefore represent a clear and present danger to both natural systems and human societies^{5,8}.

The combined effects of stressors almost always differ from the sum of their parts, yet these are still poorly understood, with current knowledge mostly limited to a few model species and a tiny number of stressor combinations⁵. Most national and international monitoring and management strategies are still based on the perspective that one dominant stressor is influencing the ecosystem – usually nutrient enrichment (i.e. via fertilisers or sewage run-off) or acidification (i.e. via acid rain). New initiatives, such as the EU SOLUTIONS Project⁹, are now beginning to take a broader view and aim to identify risks of chemical mixtures under different economic development and climate change scenarios. In general, current strategies do not consider the relationships and feedbacks between stressors, and their influence on ecosystem properties, despite these being embedded in new statutory regulatory requirements^{10,11}.

With ongoing chemical and climate change, the old baselines formerly used to gauge ecosystem health are increasingly obsolete as 'novel ecosystems' we have not seen before – with new combinations of native and non-native species coexisting within new environmental contexts – continue to emerge. To cope with this new reality, we need better ways to measure ecological status and its response to these multiple new drivers, especially if we are to translate this into sound policies and effective mitigation strategies.

Policy and legislation in this area is becoming increasingly holistic in its vision and advocacy for an ecosystem-based approach, as pioneered, for instance, in the European Union (EU) Water Framework Directive (WFD) for implementing its Integrated River Basin Management Plans, and new analogues are emerging in many other parts of the world¹². However, research into multiple stressor impacts is still very rare, with only a few studies explicitly quantifying their combined impacts^{3,4,5,8,13}. In general, this research shows that non-additive effects (i.e. not simply the sum of their parts) are the norm, with two stressors frequently causing a combined impact which is **less than** the sum of their independent effects. This may seem surprising, given that it is often assumed that stressors will amplify one another's effects, and it also has important implications for management strategies¹⁰.

Coping with ‘ecological surprises’ and the combined effects of stressors

The independent effects of some anthropogenic stressors are now relatively well described, at least at the individual or population level for a few focal species (e.g. changes in abundance, local extinctions, range shifts in mussels and fish^{14,15}). Whole community and ecosystem-level responses are less well known¹⁶, but these can include loss of biodiversity and/or ecosystem services, such as fish production or carbon sequestration. The combined effects of multiple stressors on multiple species are even less well-characterised, despite this being the predominant scenario in the real world. Unfortunately, these cannot be predicted from simple single driver and/or response approaches, no matter how much time and effort we invest in characterising these relationships, because of unknown interactions between stressors¹¹. The combined effect of two stressors has three basic potential outcomes:

it is either (a) additive (equal to the sum of their independent effects) or non-additive (either (b) less or (c) more than the sum of their independent effects). In the first non-additive case, the two stressors may mitigate one another’s effects: a so-called ‘antagonistic interaction’ between stressors (Figure 1). In the second case, the stressors amplify one another’s effects: a ‘synergistic interaction’ (Figure 1). Traditional simple approaches, that only account for individual stressors, or an approach that is simply additive, cannot explain these synergistic or antagonistic interactions, so these results have often been termed ‘ecological surprises’. In reality, the opposite is true: perfect additivity should be rare in complex natural systems where many different variables can alter interactions (both between species and stressors), so such results are not surprising at all: they are exactly what we should expect to see in nature^{3,5,8,13,14,17}. As such, we need to move beyond simplistic additive null models (as shown in Figure 1) to predict the combined impacts of stressors, and instead take stressors’ modes of action into account¹⁷.

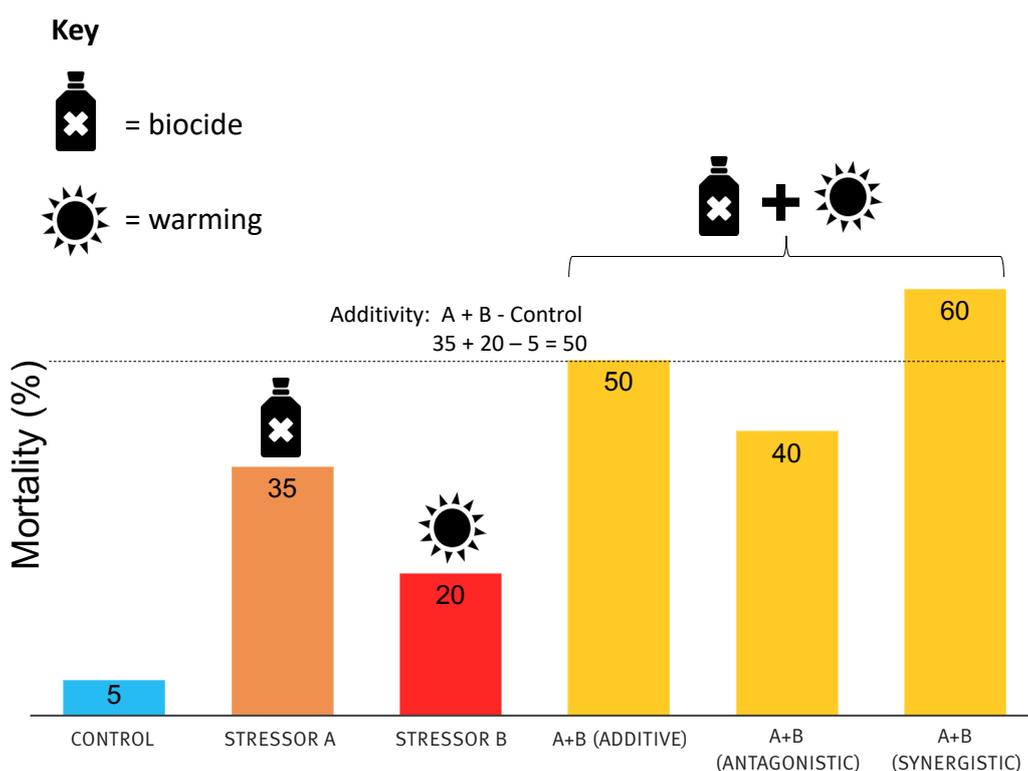


Figure 1: Conceptual approach to determining interaction types from experimental response data (e.g. % mortality or diversity loss) in multiple stressor studies. Figure 1 shows the percentage of a population that suffer mortality with no stressors (control), stressor A (biocide), stressor B (warming), and with both stressors (A+B). When both stressors occur, their combined impact can be classified as additive, antagonistic or synergistic. This is calculated by comparing the response under both stressors with the additive sum of their individual effects relative to the control (see Piggott et al. 2016¹³ for further examples).

Evidence suggests that non-additive effects can be caused by:

1. One stressor altering the sensitivity of a species to other stressors. For instance, a pollution event reduces the health of a native fish (Figure 2, label a), making it more susceptible to an extreme warming event (Figure 2, label b)^{18,19}.
2. The presence of one stressor changing the intensity of another through physical or chemical alteration of the second stressors severity or prevalence (Figure 2, label d²⁰). For instance, warming can alter the potency of pollutants via several mechanisms (e.g. the rate it enters the organism and is metabolised or its breakdown rate), either amplifying or mitigating the pollutant's negative effects²¹.
3. Altered feeding relationships among species and cascading food web effects (Figure 2, label e). For instance, if herbivorous snails lose their food resource to a herbicide spill, their populations may crash, even if the chemical is not directly toxic to the snails themselves²².
4. Positive or negative co-tolerance of species sensitivity to each stressor (i.e. if a species is tolerant to both warming and pollution, this is positive co-tolerance, whereas negative co-tolerance would occur if a species is sensitive to one stressor, but not the other). For example, insects are sensitive to insecticides and plants are sensitive to herbicides, so the presence of both stressors will have an amplified effect on diversity (as opposed to both stressors being insecticides, which would not directly impact the plants²³).

How individual organisms respond to multiple stressors will ultimately shape effects at the higher organisational levels of populations, assemblages, food webs or entire ecosystems (Figure 2, label e), and non-additive effects can therefore manifest at any level. Stressors may affect any part of a food chain, from plants to top predators (Figure 2, label e) and all of these interactions between stressors and organisms can alter food web structure and, by extension, ecosystem services (Figure 2, label f).

Biocides and collateral damage via indirect impacts on food webs

Biocides are intended to target one particular taxonomic group of pests or pathogens: insecticides will harm insects; fungicides target fungi; antibiotics and disinfectants kill bacteria. Unfortunately, they can also affect other unrelated groups via both direct toxic effects (e.g. heavy-metal based fungicides are also often poisonous to fish) and also indirectly via the food web. Consequently, their intended targets in one context can lead to widespread indirect collateral damage: insecticides are toxic not just to pest species in crops, but also to the diverse insect fauna that forms the core of freshwater food webs, such as mayflies and dragonflies, and this has indirect consequences for the species that feed on them in turn²².

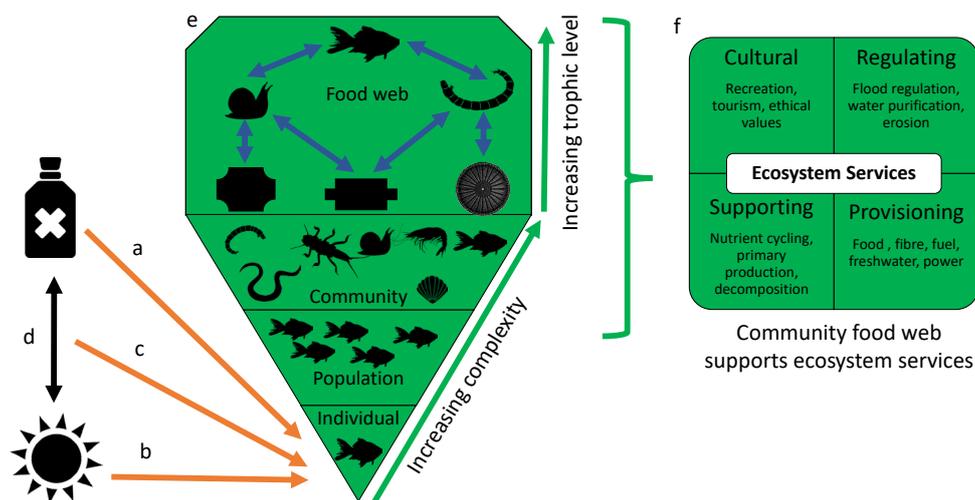


Figure 2: A representation of freshwater food webs and how the ecosystem services they deliver can be affected by multiple stressors (in this example a biocide and warming). Figure 2 depicts how stressors have independent (label a and b) and combined effects (label c), from individuals-to-ecosystems. Their combined effects can be altered by direct interactions between the stressors (label d) or interactions between species (label e). The effect of multiple stressors can cascade across levels of organisation and through trophic interactions (label e) to alter whole food webs and ecosystem services (label f).

This collateral damage can therefore ripple through the food web, far beyond the obvious anticipated impacts on the original target species (see Figure 2). For instance, if an insecticide spill kills a predatory fish's prey, the predator's population may crash even though the insecticide is not directly toxic to fishes. Since all freshwater food webs are built upon two major routes of energy input which then flow through multiple food chains in the network, we need to consider how stressors affect these trophic pathways from resources to top predators. In freshwater ecosystems, these two major trophic pathways flow (1) from algae and plants to herbivores ('green pathways'), and (2) from leaf-litter to microbial decomposers and invertebrate detritivores ('brown pathways'). Biocides that affect these pathways can alter the wider food web as they are integrated by intermediate consumers (e.g. insects) and apex predators (e.g. predatory fish, see Figure 3). Note this is distinct from biomagnification – whereby toxins concentrate in consumers' tissues as they move up the food chain²⁴ – instead, we refer to the indirect effects that propagate through the food web, which may persist even long after the toxin itself has vanished from the system.

Since many biocides eventually end up in our fresh waters, especially in the lowland catchments and floodplains of the world's major rivers which contain a high proportion of the world's most populated areas, they could create potentially toxic cocktails that can become increasingly potent downstream. Land-use can also influence the chemical mix: for instance, one might expect to find more medical antibiotics in an urban context and more agricultural pesticides in a rural region. At present, our capacity to quantify or predict the combined effects of these mixtures is still limited – particularly when trying to predict the full spectrum of ecological responses in the context of climate change. When and how biocide mixtures affect ecosystems will depend on their direct and indirect effects in isolation, how these change in mixtures and with additional climatic stressors, and the structure of the food web.

Biocides and climate change

Understanding how biocide impacts will be modulated by climate change is the crucial next step for managing and conserving future freshwater ecosystems. It is particularly important to understand how temperature change will affect concentrations of toxins in time and space, as well as their breakdown rates. Temperature provides a framework for understanding and predicting responses to stressors because it is a 'master variable' that sets the pace of life – especially for the 'cold-blooded' ectotherms (e.g. invertebrates and fishes) that dominate fresh waters but whose metabolism is determined largely by environmental temperature²⁵).

Biocides can act in pulses, such as an acute pesticide spill, and/or chronic pollution such as persistent antibiotic pollution from household wastewaters. Across this spectrum of scales in time and space, chemical stressor impacts are modulated by water temperature and volume, as this drives the metabolism of all members of the food web and therefore of the ecosystem as a whole.

Warming associated with climate change can increase the toxicity of some chemicals whilst also accelerating their degradation and shortening their environmental half-life²¹. In addition to incremental long-term warming trends, pulsed extreme weather events, such as heatwaves, floods and droughts are also predicted to become more prevalent under climate change². These will shape the delivery and activity of biocides, by influencing their dilution via precipitation and hydrology, and metabolic activity may be pushed beyond its normal envelope for many organisms, amplifying the overall stressor load. As the threat of water scarcity increases globally²⁶, the shrinking subset that is 'clean' will become increasingly valuable for supporting biodiversity and ecosystem services, whilst being exposed to increasing human exploitation.

We now know that many properties of freshwater food webs are sensitive to both biocides and different components of climate change (e.g. Box 1). We also see how food webs can be linked relatively directly to ecosystem services via associated processes and key species (as shown in Figure 2): for instance, fisheries production is supported by energy fluxes from the lower trophic levels. At the base of the food web, other key ecosystem services – such as water purification and carbon sequestration – are driven by microbial activity. Taking a food web approach is therefore a logical necessity for understanding how multiple stressors alter the delivery of ecosystem services, and also how services are linked to one another in our rapidly changing world^{11,27}.

The River Kennet case study (Box 1²²) highlights an initial "proof-of-concept" step towards developing approaches to detect and predict multiple stressor impacts in the future. Even in this seemingly complex food web, each species is no more than two feeding links removed from any other, enabling very rapid responses to (and recovery from) perturbations (Figure 3). By characterising the ecological network and associated functional metrics, it is possible to anticipate a far wider range of potential responses. Importantly, with such approaches it is not the identity of the species that matters per se, rather it is their functional role, i.e. the configuration of nodes and links in the system and how energy moves among them. This conceptual understanding can be transferred across ecosystems through both space and time: for example, the properties of food webs in English streams can be compared with their counterparts in Australia, even though they may have no species in common.

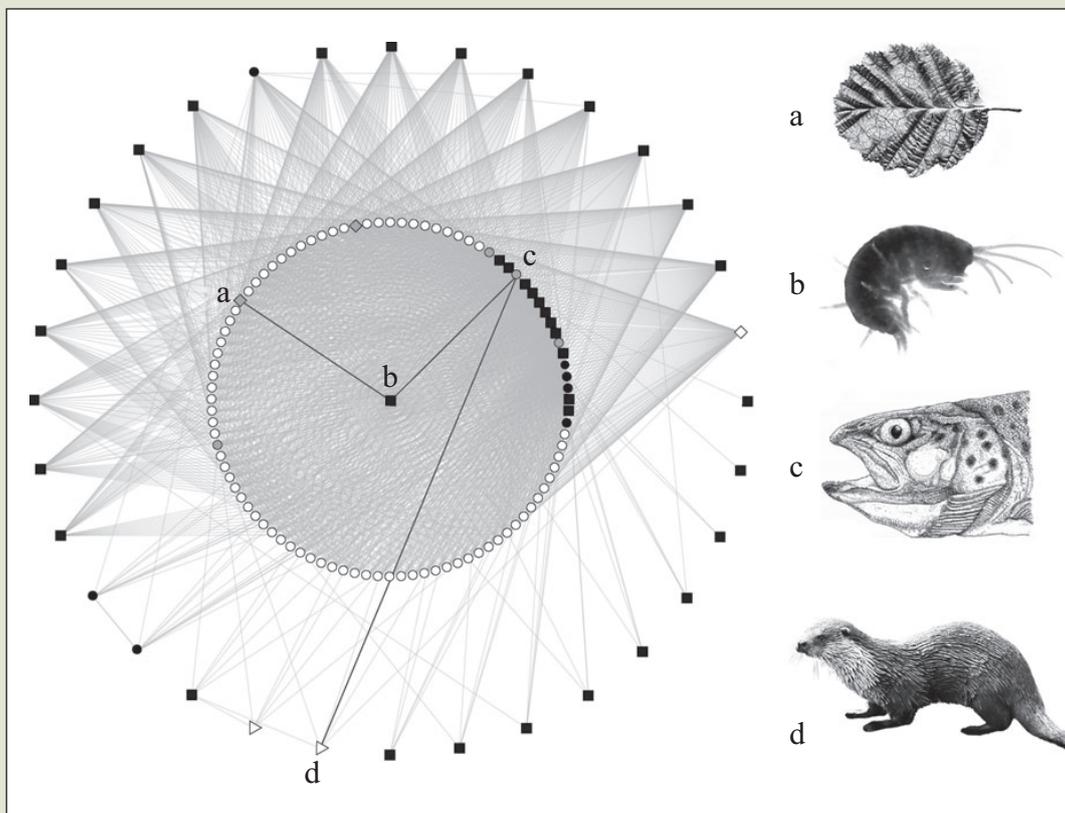
Box 1: River Kennet case study: gene-to-ecosystem impacts of a catastrophic pesticide spill

In 2013 an insecticide spill in the River Kennet, the largest tributary of the River Thames, triggered multiple direct and indirect impacts that rippled through the food web²². Molecular data revealed elevated abundances of genes associated with microbes that (1) exploit the chemical as a food resource (i.e. a *direct positive* response), and (2) process the dead invertebrates killed by the spill (i.e. an *indirect positive* response;²²).

Many algae in the impacted sites bloomed and were an order-of-magnitude bigger compared with unaffected sites, due to the loss of invertebrates that would otherwise eat them.

In contrast, a crash in the numbers of freshwater shrimp (*Gammarus pulex*) slowed the processing of leaf-litter at impacted sites. This meant the insecticide had opposing indirect effects on the 'green' (herbivorous) versus the 'brown' (detritivorous) pathways in the food web²².

Such indirect cascading effects are common in nature, but only become apparent if regulators and scientists use more holistic approaches to monitor and predict stressor impacts across multiple trophic levels. The major challenge now is to understand how multiple stressors operate within these complex natural systems.



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Figure 3: An example food web from the River Kennet where nodes represent algal primary producers (white circles), detrital resources (grey diamonds), invertebrates (black squares), fishes (grey circles) or mammals (white triangles), and lines represent trophic links. We have highlighted one food chain, from a major basal resource, leaf litter, to the top predator, Eurasian otter, via the shrimp *Gammarus pulex* and brown trout. The two concentric circles of nodes represent the shortest food-web distances to or from the shrimp – those in the inner circle are a single link away and those in the outer circle are separated by two links in the shortest path.

Recommendations for the management of freshwater ecosystems

A robust understanding of the interactive effects of biocides and climate change is needed to ensure that the actions taken actually improve the overall state of ecosystems. Policymakers should use a whole-ecosystem or network approach to predict the impacts of multiple stressors and to prioritise interventions based on their likely effectiveness¹¹.

Emerging monitoring techniques can assist by providing new ways to manage freshwater ecosystems better: these novel indicators of ecosystem state and the next generation of biomonitoring tools will need to be sensitive to the multiple stressors operating within a given catchment¹¹. For instance, bacterial assemblages can be used as rapid ‘biosensors’ that a new biocide has entered the ecosystem since their abundance and/or molecular make-up responds almost immediately when they are exposed to a pesticide. As such, bacteria are the ‘first responders’ at the base of the food web, whereas the more integrated response of invertebrates and fish populations will take longer to be detected.

A network-based approach can be used to detect both initial impacts (via bacterial biosensors) and to predict wider food web and ecosystem consequences. Ultimately, this should be able to predict how natural ecosystems will respond under future scenarios, and because such approaches focus on the functional attributes of species (what they do) rather than their taxonomy (who they are), which varies hugely from place to place, it could be applied universally¹⁶. This is especially important where the changing climate is already reshaping natural ecosystems away from their longstanding ‘normal’ baseline conditions: continuing to ignore the complexity of the real world as an inconvenient truth is no longer an option, if we are to monitor, model and manage natural ecosystems effectively¹¹.

The development of such tools will require new interdisciplinary research, knowledge integration and collaboration, capable of dealing with the ecological complexity of freshwater ecosystems: many of these component pieces already exist in a nascent form, but we now need to forge them into a more unified approach to meet the demands of managing the twenty-first century’s threatened freshwaters. Policymakers and conservationists at the forefront of environmental protection need to be brought into these developments so that policy frameworks and action on the ground reflects the latest scientific understanding, and vice-versa.

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Acknowledgements

We would like to thank the Grantham Institute – Climate Change and the Environment and particularly Alyssa Gilbert and Simon Levey for their help throughout the development of the paper. We would also like to thank the reviewers for their helpful insights: Dr Stephen Thackeray (Centre for Ecology and Hydrology) and Dr Pippa Curtis-Jackson (Environment Agency). The authors are funded by the NERC programme Emerging Risks of Chemicals in the Environment (NE/S000348/1).

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