BIODIVERSITY
Published: 10 October 2023
doi: 10.3389/frym.2023.1147094

MINIATURE STREAMS SHOW SCIENTISTS HOW MULTIPLE STRESSORS IMPACT OUR RIVERS

Daria Baikova[1], Lisa Boden[2], Aman Deep[2], Annemie Doliwa[3], Una Hadžiomerović[1], Florian Leese[4*], Iris Madge Pimentel[4*], Ntambwe A. Serge Mayombo[5], Jeremy Piggott[6], Sebastian Prati[3], Tom Lennard Stach[7], Jörn Starke[7] and Anna-Maria Vermiert[8]

[1] Department of Aquatic Microbiology, Faculty of Chemistry, University Duisburg-Essen, Essen, NRW, Germany
[2] Department of Biodiversity, Faculty of Biology, University Duisburg-Essen, Essen, NRW, Germany
[3] Department of Aquatic Ecology, Faculty of Biology, University of Duisburg-Essen, Essen, NRW, Germany
[4] Aquatic Ecosystem Research, Faculty of Biology, University Duisburg-Essen, Essen, NRW, Germany
[5] Department of Phycology, Faculty of Biology, University Duisburg-Essen, Essen, NRW, Germany
[6] Department of Zoology, School of Natural Science, Trinity College Dublin, The University of Dublin, Dublin, Ireland
[7] Environmental Metagenomics, Research Center One Health Ruhr of the University Alliance Ruhr, Faculty of Chemistry, University of Duisburg-Essen, Essen, NRW, Germany
[8] Department of Animal Ecology, Evolution and Biodiversity, Faculty of Biology and Biotechnology, Ruhr University Bochum, Bochum, NRW, Germany

YOUNG REVIEWERS:

KINABALU INTERNATIONAL SCHOOL
AGES: 10–11

YALE PATHWAYS/ OPEN LABS
AGES: 11–14

Rivers are the lifelines of our planet. We depend on them for drinking water and food production, and they are home to many plants and animals. Unfortunately, rivers are under pressure from stressors like increasing temperatures, pollution, and habitat destruction. We often know how a single stressor impacts a river, but when more than one stressor is present at the same time, the consequences are often unpredictable. To protect our rivers now and in the future, we must understand what happens in the rivers when multiple stressors

are present at the same time. However, it is difficult to understand what multiple stressors are doing to aquatic life by just observing a river or doing a laboratory experiment. In this article, you will learn how experimental miniature streams can help us to investigate the consequences of multiple stressors on rivers.

RIVERS UNDER STRESS

All of us feel under pressure sometimes and are affected by stress in one way or another. Just like humans, the natural environment can be under pressure from stress, such as high temperatures, pollution, and **habitat destruction** (Figure 1). Often, the environment is affected by multiple **stressors**—more than one kind of stress at the same time. Delicate **ecosystems** like rivers are very sensitive to multiple stressors, and the plants and animals living in them can be severely affected. Rivers are essential for us, too, because we depend on them for drinking water and food production. Yet, they contain <1% of the surface fresh water on our planet [1], making it even more important to protect what little river water is available.

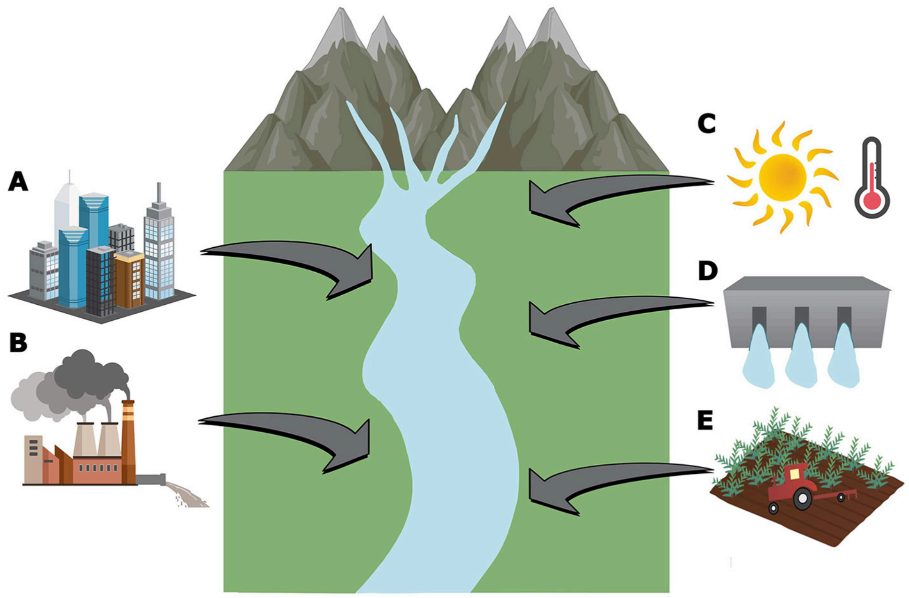

Figure 1

How can we protect our rivers if we do not know how multiple stressors affect them? Fortunately, scientists can create experimental miniature streams to safely test the effects of multiple stressors on a river, without actually harming the river or its inhabitants.

HABITAT DESTRUCTION

Damaging or reduction of the living space of an organism or groups of organisms, for example by building a dam in a river.

STRESSORS

Any environmental change that might affect (mostly negatively) organisms or entire ecosystems. Examples include fertilizers and pesticides that enter the water, or increased water temperatures and lower water levels in rivers.

ECOSYSTEMS

All the organisms in a defined environment (a lake or forest) that live and interact with each other.

Figure 1

Multiple stressors that commonly affect rivers. **(A, B)** Pollution by wastewater from cities and industries. **(C)** High water temperatures due to climate change and the removal of trees next to the river. **(D)** Habitat destruction caused, for example, by dams built to generate electricity. Dams form barriers that organisms cannot easily pass through. **(E)** Pollution by farming, as fertilizers and pesticides used during food production end up in rivers. Created with BioRender.com. Image credits: Sun **(C)**: Jane Thomas, dam **(D)**: Kate Moore, farming **(E)**: Tracey Saxby, https://ian.umces.edu/media-library.

WHEN $1 + 1 = 3$

When multiple stressors put pressure on rivers, the effects on the **aquatic organisms** can sometimes be different from what we would expect [2]. Imagine it is a particularly warm and dry summer, and a river is affected by both a low water level, due to poor rainfall, and a high water temperature. There are many animals living in this river, and some are more sensitive to these stressors than others. Mayfly **larvae** are sensitive to both stressors. A low water level alone would kill one out of five mayfly larvae, and high temperature alone would also kill one out of five larvae.

So how many mayfly larvae die when the two stressors are present at the same time?

We would expect that $1 + 1 = 2$, meaning that we could just add up the number of mayfly larvae dying from each stressor. This would mean that two mayfly larvae out of five are lost in the multiple-stressor scenario. However, we actually see that three out of five larvae die, which gives us a $1 + 1 = 3$ situation (Figure 2). How can this be?

AQUATIC ORGANISMS

All living creatures, for example, plants and animals, living in environments made up of water, like rivers, lakes, and the ocean.

LARVAE

Young insects are called larvae. After hatching from the egg, aquatic insects look very different from their adult flying forms. During this stage, the insects feed and grow.

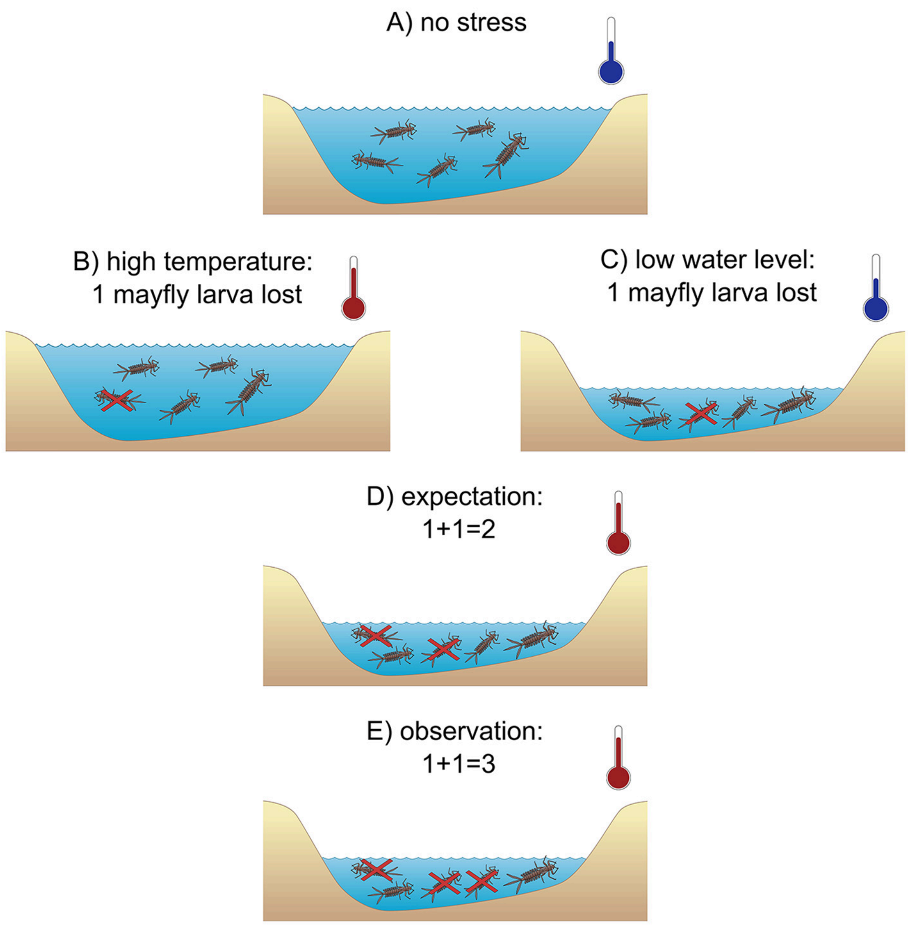

Figure 2

Figure 2

When $1 + 1 = 3$. **(A)** Five mayfly larvae live in an unstressed river. **(B)** During a hot summer, the heat stress kills one of the five mayflies. **(C)** During a dry summer, low water levels kill one of the mayflies. **(D)** We would expect two mayfly larvae to be lost if *both* heat stress and low water levels affect the river. **(E)** Instead, three mayfly larvae are lost if both stressors affect the river. This is a $1 + 1 = 3$ scenario. Image credits: River cross section and thermometer: Tracey Saxby, mayfly larva: Dieter Tracey, https://ian.umces.edu/media-library.

To answer this question, we must first understand how low water levels and high temperature put pressure on the river. Rivers are rich in oxygen because, when the water is flowing, it mixes with the air. Oxygen is essential for mayfly larvae and other aquatic animals. But if the speed of the water movement is reduced, less water and air are mixed together, which can decrease the amount of oxygen in the water. This can happen when the water level in the river is low because the water flows more slowly. An increase in water temperature can also reduce the amount of oxygen in the river, because oxygen does not dissolve well in warm water. But this still does not explain why we lose more mayfly larvae than expected when both stressors are present. So, what are we missing?

We must remember that rivers are complex ecosystems in which many kinds of organisms interact with each other. Stressors can change the way organisms interact. In our example, several kinds of algae live in the river, along with the mayfly larvae. Some algae *prefer* warm, slow water, and they multiply quickly under these conditions—using large quantities of oxygen as they do so. These algae reduce the already limited amount of oxygen in the water, which kills even more mayfly larvae than low water levels or high temperature alone.

Keep in mind that multiple stressors can interact in various ways. For example, we might also have a $1 + 1 = 1$ situation, in which the harmful effect of multiple stressors on the river is less than expected. We could even have a situation in which $1 + 1 = 0$, in which nothing changes because the two stressors cancel each other. But the most dangerous situation for a river is the $1 + 1 = 3$ scenario.

HOW CAN WE STUDY MULTIPLE STRESSORS?

To better understand the effects of multiple stressors, we can directly observe rivers by looking at changes in water quality and the organisms living there. For example, we could see what happens to a river after a hot summer with little rain. But every river is different, so it is impossible to know if the changes we see are caused only by low water levels and high temperatures, or whether they are caused by other stressors we did not consider. This could result in false conclusions and actions taken to protect the river that might not be effective.

An alternative to observing rivers themselves is to use aquariums or other types of containers in laboratory experiments, in which we can have everything under our control. The advantage of these experiments is that we can safely test various stressor combinations without harming the river. We can also have many aquariums with the same conditions, which are called **replicates**. If we add a stressor and observe the same change in all the aquarium replicates, we can be confident that the stressor we used is causing the change that we see.

REPLICATES

Copies of identical conditions within a scientific experiment. Only if the results are consistent among the replicates, we can be certain that they are not produced by chance.

The main disadvantage of using laboratory aquarium experiments is that they are over simplified compared to a real river. This is because fewer types of organisms are present in the experiment, and conditions like light and temperature do not change the same way they do in nature. To study multiple stressors, we need a realistic experiment in a natural setting, which means outside. We also need replicates so that we can be certain that our measurements are correct. At the same time, we do not want to harm the river itself. So, what can we do?

EXSTREAM: STUDYING MINIATURE STREAMS

We can use water from a river to create artificial, miniature streams in an outdoor experiment (Figure 3). By pumping water from the river into small enclosures, we can simulate the natural conditions of the river. To create the right habitats for various river organisms, we add sand, gravel, and stones taken from the river, to make them feel comfortable. We also add fallen leaves that can be used as food by organisms such as the mayfly larvae in our example. By pumping water from the river, organisms that live in the river are carried into these experimental miniature streams. Like in a real river, they can decide to stay, or they can leave if the conditions are not right. For example, if mayfly larvae feel stressed, they can move out of the miniature stream through a hole in the enclosure, by way of the water current.

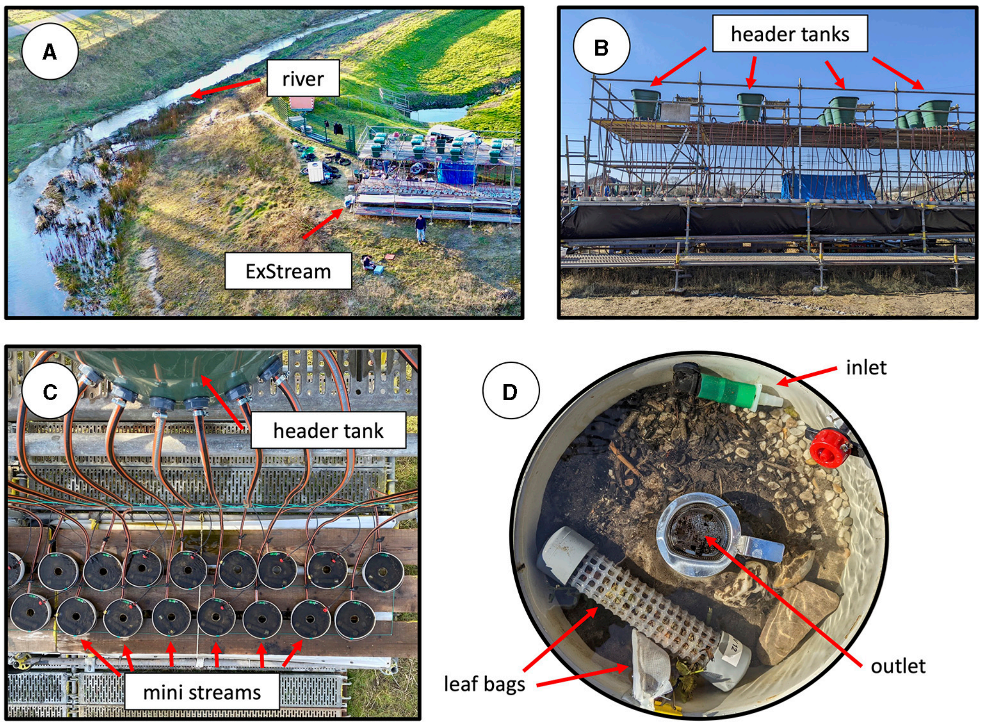

Figure 3

The experimental system ExStream. **(A)** Top view of ExStream. The system is always constructed next to a river. Water is taken from the river and pumped through 64 miniature streams. **(B)** Side view. The river water first enters the header tanks on top of a scaffold. **(C)** Top view from the header tank. Water flows downwards through pipes into the miniature streams. **(D)** Close-up of a miniature stream. The mini streams are filled with sediment from the river and fallen leaves as a food source for the animals. ©Photos **(A, C, D)** taken by Philipp M. Rehsen.

We call this system of miniature streams ExStream [3], and it can be manipulated like aquariums in a laboratory. For example, we can add warm water to simulate high water temperature, or we can control the speed of the water by changing the size of the inlet opening of the miniature streams.

If we manipulate only some of the miniature streams and let the others remain in natural conditions, we can compare them. The differences between manipulated and natural miniature streams tell us about the effects of the stressor. As we have many miniature stream replicates, we can see if the effect of the stressor is the same in all stressed miniature streams. By using stressors alone or in various combinations, we can identify the individual and combined effects of stressors on aquatic organisms like algae, insects, crustaceans, parasites, bacteria, and fungi. This helps us to understand what we can do to protect our rivers.

CONCLUSIONS

In nature, things are much more complicated than in our mayfly example. We only looked at two stressors and two organisms, mayfly larvae and algae. But many more organisms live in rivers, and more than two stressors can be present at the same time. The many river organisms can each be affected by different stressors, which can have multiple effects on the ways the organisms interact with each other. This complexity is what makes the consequences of multiple stressors so unpredictable, and why it is important to try to understand their effect on rivers. From the above example, we can learn that it is essential for slow-flowing river sections to avoid high water temperatures, for example by planting trees on the river's banks—this offers shade to cool down the water.

Miniature stream experiments allow us to identify and understand stressful situations when $1 + 1 = 3$. This is crucial given how delicate and complex rivers are and how important they are for us humans and for nature. Only by understanding how stressors interact can we effectively protect our rivers now and in the future.

ACKNOWLEDGMENTS

The idea for this contribution was developed within the framework of the Integrated Research Training Group (IRTG) of the Collaborative Research Centre RESIST (CRC 1439). In particular, we thank the IRTG management team, Vanessa Wirzberger, Verena Brauer, Bernd Sures, and Michael Eisinger for their support. The study was funded by the Deutsche Forschungsgemeinschaft (DFG, German Research Foundation) - CRC 1439/1 - project number: 426547801.

REFERENCES

1. Gleick, P. H. 1996. "Water resources," in *Encyclopedia of Climate and Weather*, ed S. H. Schneider (New York, NY: Oxford University Press). p. 817–23.
2. Piggott, J. J., Townsend, C. R., and Matthaei, C. D. 2015. Reconceptualizing synergism and antagonism among multiple stressors. *Ecol. Evol.* 5:1538–47. doi: 10.1002/ece3.1465
3. Piggott, J. J., Townsend, C. R., and Matthaei, C. D. 2015. Climate warming and agricultural stressors interact to determine stream macroinvertebrate community dynamics. *Glob. Change Biol.* 21:1887–906. doi: 10.1111/gcb.12861

SUBMITTED: 18 January 2023; **ACCEPTED:** 15 September 2023;
PUBLISHED ONLINE: 10 October 2023.

EDITOR: David Hiller, Yale University, United States

SCIENCE MENTORS: Milena Salgado-Lynn and Lam Vo

CITATION: Baikova D, Boden L, Deep A, Doliwa A, Hadžiomerović U, Leese F, Madge Pimentel I, Mayombo NAS, Piggott J, Prati S, Stach TL, Starke J and Vermiert A-M (2023) Miniature Streams Show Scientists How Multiple Stressors Impact Our Rivers. Front. Young Minds 11:1147094. doi: 10.3389/frym.2023.1147094

CONFLICT OF INTEREST: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

YOUNG REVIEWERS

KINABALU INTERNATIONAL SCHOOL, AGES: 10–11
We call our group KIS: Kids in Science. We work hard as a team to bring you articles about science for kids. We endeavor to review the text and make it easier for kids to understand so that you, too, can love science like we do!

YALE PATHWAYS/OPEN LABS, AGES: 11–14

The Pathways Reviewers have interests in many of the sciences: including biology (animal and marine), chemistry, and computer science! They are in grades 6–8. Anikaith loves computers, math, dogs and food! C'yonni dances and sings. Josie likes to swim and hang out with friends. Marvelous and Melinda enjoy learning about the brain. Miguel loves computer and video games. Quinn enjoys being outdoors playing soccer, skateboarding, and snowboarding.

AUTHORS

DARIA BAIKOVA

I am an ecologist and my current research investigates a microworld of bacteria in rivers. Invisible to the naked eye, bacteria perform an incredibly important role by decomposing dead organisms, allowing nutrients to move from rivers to the oceans. To understand how this is influenced by stressors, I study the ability of bacteria to transform organic matter and I examine their diversity through genetic analysis. This research will help us to reveal the effect of human-created pollution and climate change on bacteria and raise the awareness of their role in freshwater ecosystems.

LISA BODEN

I am a microbiologist currently working with protists living in freshwater habitats. My research focuses on analyzing the effects of stressors on the feeding behavior of protists grazing on bacteria. Such microbial predator-prey interactions are essential building blocks of aquatic food webs. In my research, I am combining genetic data with more traditional microscopic methods to analyze how feeding behavior of protists is affected by multiple stressors. With my research I want to contribute to a better understanding of freshwater habitats.

AMAN DEEP

I am a bioinformatician currently working with the fungal and bacterial community in freshwater habitats. My research focuses on coarse particulate organic matter decomposition by these microbes and the effects of multiple stressors like salt and temperature. To investigate this, I also use the ExStream system, from which biological samples are further used for genetic and computational analysis. From this analysis, I target communities and pathways that are involved in decomposition. My work contributes to a better understating of the roles of bacterial and microbial communities in the decomposition of organic matter in freshwater water habitats.

ANNEMIE DOLIWA

I am a biologist fascinated by the hidden world of parasites. Parasites play several important roles in ecosystems, can provide valuable information about their environments, and serve as living indicators of environmental stressors. My Ph.D. project focuses on parasites in freshwater habitats. I am investigating how stressors such as salt and temperature affect their species richness and infection rates. I use genetic and morphological methods to analyze parasite community responses to multiple stressors, both in field studies and in experimental systems such as ExStream. My goal is to identify parasites that can be used to assess stream degradation and restoration.

UNA HADŽIOMEROVIC
I am a freshwater ecologist with a focus on microbiology. Since microbes play an important role in functioning of a river ecosystem, my Ph.D. project is looking into one of the functions bacteria provide—recycling nutrients and carbon from dead organisms. To investigate how this is affected by salt and temperature, I use experimental manipulations (like the Ex-Stream system) as well as field monitoring. The impact of stressors on the bacterial community is detected with chemical and genetic analysis. This will help us understand how environmental changes influence bacterial recycling of dead organisms and consequently, the state of river ecosystems.

FLORIAN LEESE
I am a biology professor at University of Duisburg-Essen in Germany, where I focus on molecular ecology and its applications in multiple-stressor research. I have held editorial roles and received awards like the Water Resource Prize and the University Teaching Award. In 2022, I became the Dean of the Faculty of Biology and chairman of the Water Science Alliance at the University of Duisburg-Essen. I am passionate about science and want to make a positive impact on the world with the results of my research. *florian.leese@uni-due.de

IRIS MADGE PIMENTEL
I am a river ecologist from Germany and my research focuses on the diverse group of macroinvertebrates: insect larvae, water bugs and beetles, worms, crustaceans, mussels, snails, water spider and mites—they all are part of this fascinating group. During my Ph.D., I will use genetic methods and automatic image recognition to characterize macroinvertebrate communities after stressor exposure and after recovery from stress. With my research, I want to contribute to the conservation of our precious river ecosystems. *iris.madge-pimentel@uni-due.de

NTAMBWE A. SERGE MAYOMBO
I am an aquatic ecologist and study diatoms, a specific group of algae. I am interested in understanding how different diatom species interact with their environment, in their diversity, and in understanding how they indicate water quality. My ongoing research project investigates the responses of diatom communities to multiple stressors, both in the field (streams and rivers) and using experiments like the ExStream. To decipher the complex responses of microalgae to multiple stressors, I combine a variety of laboratory methods.

JEREMY PIGGOTT
I am a professor in aquatic biology with interests in the combined influence of multiple human-created stressors on communities and ecosystems, and the management and conservation of biodiversity and ecosystem services in the face of global change. I perform my research predominantly in streams and rivers, combining "natural" field experiments with experiments at a range of scales, from whole streams to systems like ExStream. By advancing knowledge and connecting

science to policy, my research seeks to improve the management, conservation, and restoration of aquatic ecosystems in the face of global change.

SEBASTIAN PRATI

I am a freshwater ecologist. My research investigates the effects of stream degradation and restoration on parasite communities. Therefore, parasites of fish, snails, and bottom-living invertebrates are investigated in near-natural, degraded, and restored streams. I also investigate the responses of infected and uninfected organisms with the ExStream system. The combination of experimental and field observations will provide insights into the effect of multiple stressors on parasite communities and the subsequent effects on the organisms in which they live.

TOM LENNARD STACH

I am a microbiologist with a focus on freshwater bacteria and viruses. My current project investigates the effects of degradation and restoration of river ecosystems on these microbes, individually and in interaction with other organisms. To answer this complex question, I use the ExStream system in cooperation with other Ph.D. students. Aside from classical laboratory-based techniques, I am mainly using computational methods to analyze the genetic material of microbes. With my research, I hope to increase awareness of the benefits of a diverse microbial community in rivers.

JÖRN STARKE

I am a microbiologist at the University of Duisburg-Essen who studies the effects of human-caused stressors on the microbes living in freshwater streams. While most scientists in our project focus on streams in Germany, I want to investigate how general principles of stressor interactions hold true across river ecosystems worldwide. To answer this difficult question, I will visit ExStream systems all around the world to collect samples and subsequently use computational methods to analyze the diversity and functions of the microbes living in each. With my research, I hope to contribute to the understanding of how human impacts can disrupt microbial freshwater ecosystems.

ANNA-MARIA VERMIERT

I am a freshwater ecologist at the Ruhr University Bochum, Germany, studying predator-prey interactions in river ecosystems. These interactions, which strongly influence the behavior, growth, and survival of prey, play an important role in the structuring of natural communities. The introduction of single or multiple stressors into a system can disrupt predator-prey interactions, resulting, for example, in the inability of prey to respond appropriately to the predator. To determine whether different stressors or stressor combinations can change avoidance behaviors toward predators, the ExStream system will be used as the basis for my research.