

# 2<sup>nd</sup> European Freshwater Mollusk Conservation Society Meeting

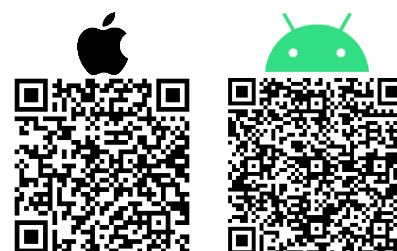


## Conference Programme Book

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**This conference will use Whova**, a web browser and smartphone app for event and conference management. Whova helps attendees navigate the conference by providing an online program to view and bookmark all conference activities, allowing for social networking, conference gamification, and postings to meeting organizers, along with other conference resources such as abstracts, logistics, local information, etc. Scan the QR code to download the app!



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# Summary schedule

Day	Start time	End time	Function	Location
17-09	0900	1030	Registration & Coffee	Karlstad University Campus Room: 1A305 'Lagerlöfsalen'
	1030	1100	Welcome to FMCS KAU	
	1100	1140	Keynote speaker: Dan Hua	
	1145	1230	Mussel breeding	
	1230	1400	Lunch	
	1400	1445	David Aldridge Keynote speech	
	1445	1545	Invasive mussels and the spread of disease	
	1545	1615	Coffee break	
	1615	1700	Invasive mussels and the spread of disease	
	1700	1745	Miscellaneous talks	
	1930	2300	Welcome reception at <i>Claesson's</i>	Sommargatan 110
18-09	0900	0945	Keynote speaker: Niklas Janz	Karlstad University Campus Room: 1A305 'Lagerlöfsalen'
	0945	1030	Host-parasite interactions	
	1030	1100	Coffee break	
	1100	1130	Host-parasite interactions	
	1130	1230	Special session: Tadeusz Zając	
	1230	1400	Lunch	
	1400	1445	Keynote speaker: Carla Atkinson	
	1445	1545	Mussel ecosystem services and functions	
	1545	1615	Coffee	
	1615	1700	Mussel ecosystem services and functions	
	1700	1745	Miscellaneous talks	
			Poster session	
19-09	0900	0945	Keynote speaker: Wendell Haag	Karlstad University Campus Room: 1A305 'Lagerlöfsalen'
	0945	1030	Threats and conservation in mussel waters	
	1030	1100	Coffee	
	1100	1230	Threats and conservation in mussel waters	
	1230	1400	Lunch	
	1400	1445	Keynote speaker: Manuel Lopes-Lima	
	1445	1545	Genetics and phylogeny and genetic methods	
	1545	1615	Coffee	
	1615	1715	Genetics and phylogeny and genetic methods	
	1930	2200	Dinner at the restaurant <i>Terassen</i>	Treffenbergsvägen 20
20-09	0900	1600	Field Excursion	Karlstad University

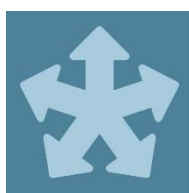
# General info

## Conference goals:

1. To bring together international experts in biology and conservation of freshwater mollusks that, through the present and forthcoming conferences and debates, will be able to create a network of knowledge with the final goal of develop collaborative projects and eventually global directives for the protection and conservation of this important faunistic group
2. To provide an incentive for non-North American freshwater malacologists to become members of FMCS and participate in planned activities, symposia, publications and workshops.
3. To set up a subcommittee within the society of local malacologists – e.g. initially from Europe, but to be expanded to other continents around the world – to provide structure and communication about resources, lobbying, collaboration and outreach. This will facilitate answering key questions and developing techniques to address the same or similar problems encountered across freshwater molluskan research.
4. To arrange joint international meetings around the world. The first four international freshwater bivalve meetings (Baltimore, Maryland, 2009; Bragança, Portugal, 2012; Buffalo, NY, 2015; and Verbania, Italy, 2018) were exciting and fun with a very good international participation. With the 2024 international meeting at Karlstad University in Sweden, we want to expand the focus on bivalves including all the freshwater mollusks and thereby further improve the building of an international network.

**The journal *Diversity*** (IF: 2.4; CS: 3.4) is preparing a special issue “Advances in Freshwater Mollusk Research”! Currently unpublished abstracts for oral and poster presentations submitted to the conference will have the opportunity to submit manuscripts to the journal special issue.

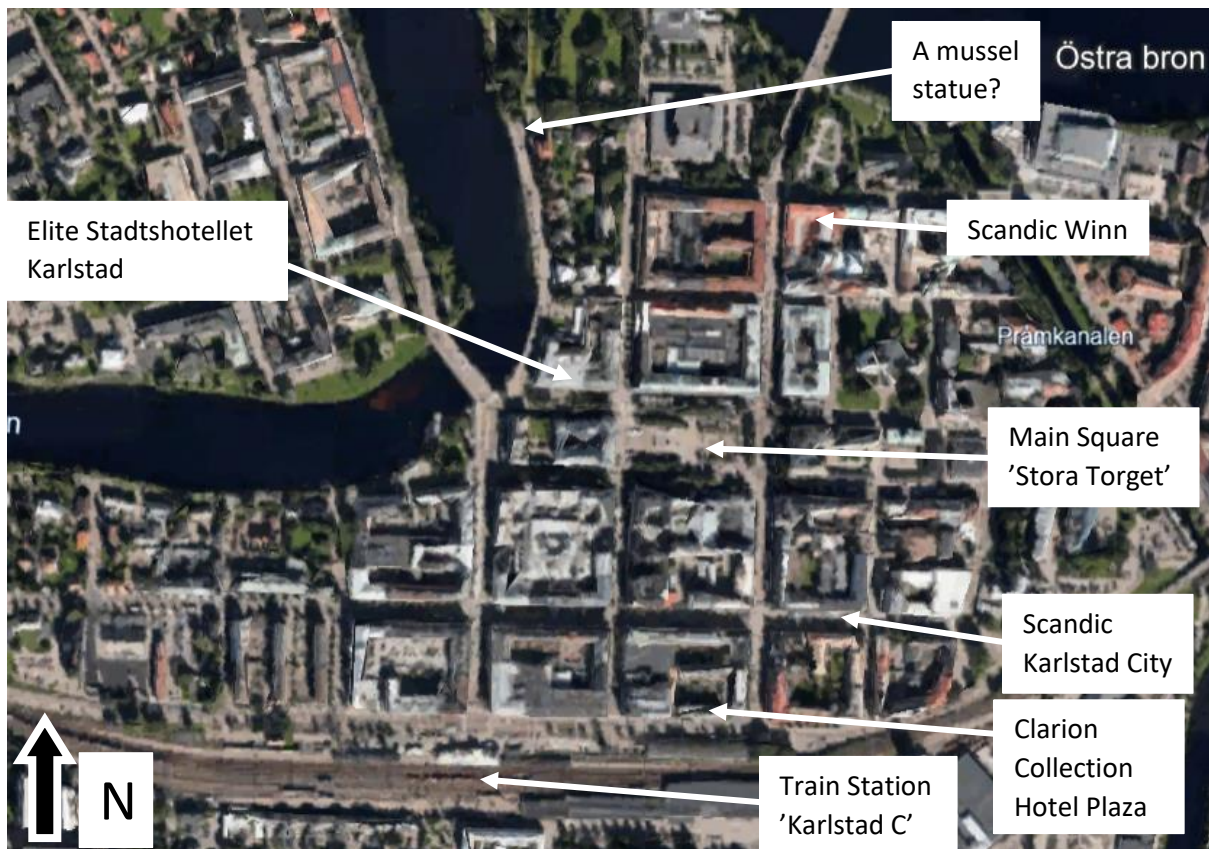
[https://www.mdpi.com/journal/diversity/special\\_issues/1R093354KE](https://www.mdpi.com/journal/diversity/special_issues/1R093354KE)



*diversity*

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**The city of Karlstad** is very sunny city located in hilly forests between Stockholm and Oslo, surrounded by a vast variety of stream and lakes. The city is located on the delta of the River Klarälven (the longest Swedish river) as it enters Lake Vänern (the largest lake in the European Union). Karlstad has been an important city since the Viking age, and their artifacts and burial mounds can be found scattered in the forests still today! The city center is small and easily navigated, below you can find an annotated map of some hotels, points for travel and an interesting attraction.

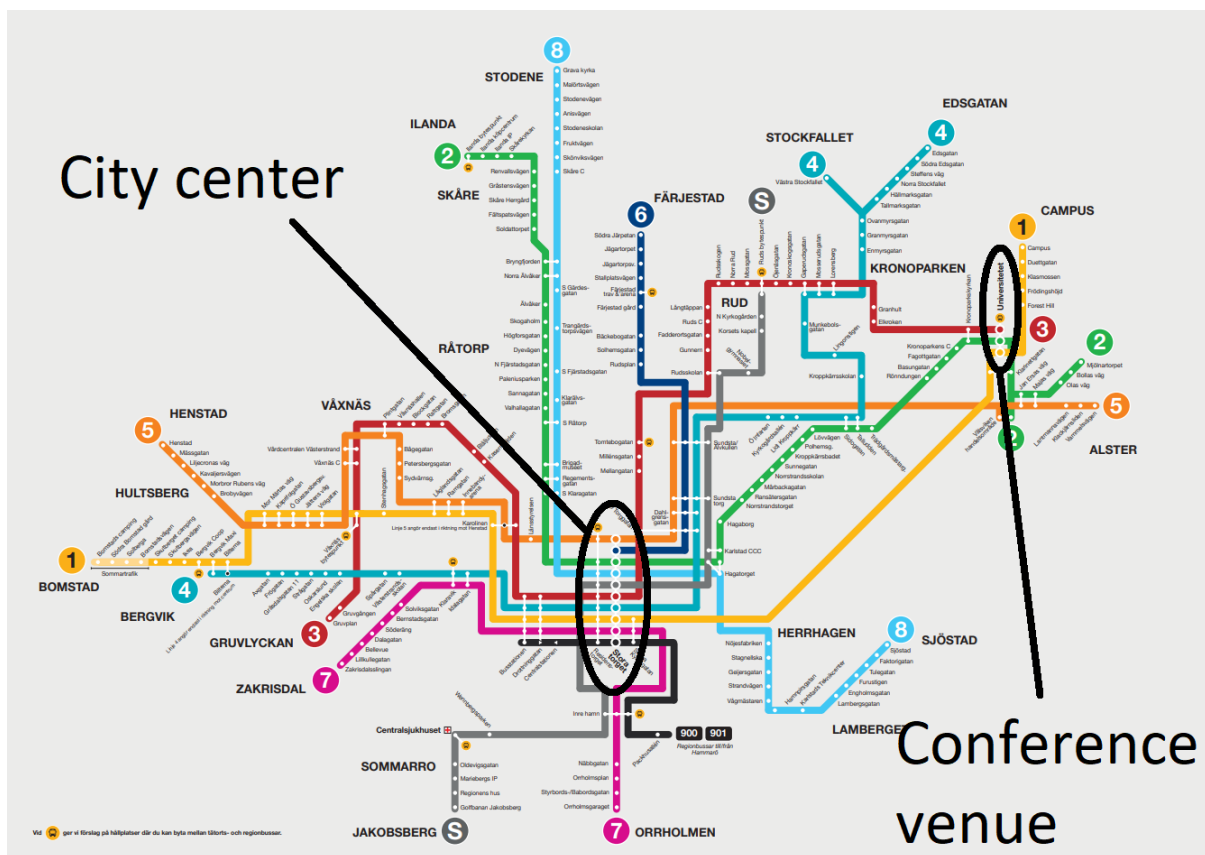


**The conference welcome reception** will be held at *Claesson's* (Sommergatan 110), a bar across the street from the conference venue. Join us there for some food and a few drinks after the first day of talks!

**The conference dinner** will be held at the Terassen Restaurant in Mariebergsskogen (Treffenbergsvägen 20), a beautiful venue just outside of the city centre, overlooking a bay in the river and next to a nature reserve. The restaurant is easily accessible with busses 10 and S, or with a 20-minute walk through the city and along the water.



The conference venue is located in the Karlstad University Campus, room 1A305 or 'Lagerlöfsalen'. Travel between hotels in the city centre and the conference venue is most easily done with the local bus service (the red buses). Bus tickets can be purchased from ticket booths next to certain bus stops, directly from the driver or, most conveniently, through the dedicated app *Karlstadbuss*. Bus numbers 1, 2 and 3 all easily connect the city centre with the university campus at the bus stop *Universitet*. See the bus map below for more details. **NB: The bus stop *Campus* is not the university campus.**



The conference field excursion will take you sight-seeing to a few different rivers to show off sites for mussel conservation, healthy populations of the freshwater pearl mussel (*Margaritifera margaritifera*), some of the local geography and woodlands and, if we keep time, some Viking sites. We will meet at the conference venue at 9:00 AM and be back around 4:00 PM. Lunch is provided with the excursion. Autumn weather is Sweden is temperamental so please check the weather before packing your bags for your trip.

# Organizing Committee

**Martin Österling**

Committee leader, Karlstad University, Sweden

**Sebastian L. Rock**

Karlstad University, Sweden

**Magnus Lovén Wallerius**

Karlstad University, Sweden

**Raviv Gal**

Karlstad University, Sweden

**Lea D. Schneider**

The Rural Economy and Agricultural Societies, Halland, Sweden

**Ted von Proschwitz**

Gothenburg Natural History Museum, Sweden

**Niklas Wengström**

Swedish Anglers Association, Gothenburg, Sweden

**Alexander Gustavsson**

Södermanland County Administrative Board

# Scientific Committee

**David Aldridge** – University of Cambridge, United Kingdom

**Carla Atkinson** – University of Alabama, USA

**Arthur Bogan** – North Carolina Museum of Natural Sciences, USA

**Megan Bradley** (president of the FMCS) – Fish and Wildlife Service, USA

**Karel Douda** – Czech University of Life Sciences Prague, Czech Republic

**Jürgen Geist** – Technical University of Munich, Germany

**Dan Hua** – Tennessee Wildlife Resources Agency, USA

**Anna Łabęcka** – Jagiellonian University, Kraków, Poland

**Manuel Lopes Lima** – CIBIO, Porto University, Portugal

**Amy Maynard** (president elect of the FMCS) – Neosho National Fish Hatchery, USA

**Teresa Newton** – U.S. Geological Survey in Wisconsin, USA

**Paz Ondina** – University of Santiago de Compostela, Spain

**Martin Österling** – Karlstad University, Sweden

**John Pfeiffer** – Smithsonian Institution, Natural Museum of natural History

**Vincent Prie** – Institute of Systematics, Evolution, Biodiversity (ISYEB)

**Nicoletta Riccardi** – CNR – ISE, Italy

**Astrid Schwalb** – Texas State University, USA

**Mary Seddon** – IUCN, Mollusc Specialist Group

**David Strayer** – Cary Institute of Ecosystem Studies, USA

**Ronaldo Sousa** – Minho University, Portugal

**Jouni Taskinen** – University of Jyväskylä, Finland

**Maria Urbańska** – Poznan University of Life Sciences, Poznan, Poland

**Tadeusz Zając** – Institute of Nature Conservation, Polish Academy of Sciences, Poland

**Alexandra Zieritz** – University of Nottingham Malaysia Campus, Malaysia



# Keynote speakers

## **Prof. David Aldridge**

University of Cambridge, UK

Presenting: *Invasive freshwater bivalves: Impacts, management and future perspectives*



## **Dr. Carla Atkinson**

University of Alabama, USA

Presenting: *Freshwater mollusk conservation in the face of global change: Identifying challenges and solutions*



## **Dr. Wendell Haag**

U.S. Forest Service, USA

Presenting: *Assessing stream health and mussel fitness with in site exposures and shell thin sections*



## **Dr. Dan Hua**

Tennessee Wildlife Resources Agency, USA

Presenting: *Propagation, Culture, and Monitoring of Critically Endangered Freshwater Mussels for Restoration*



## **Prof. Niklas Janz**

Stockholm University, Sweden

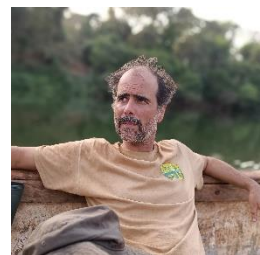
Presenting: *“The Parasite Paradox”*



## **Dr. Manuel Lopes-Lima**

University of Porto, Portugal

Presenting: *Integrating Phylogeny, phylogeography, and genetic diversity: A comprehensive approach to freshwater mussel conservation*



## Detailed schedule

Day	Start time	End time	Function/talk	Speaker	Authors
17-09	09:00 AM	10:30 AM	Registration		
	10:30 AM	11:00 AM	Welcome to the second European FMCS meeting		
	11:00 AM	11:45 AM	<i>Keynote: Propagation, Culture, and Monitoring of Critically Endangered Freshwater Mussels for Restoration</i>	Dan Hua	<a href="#">D. Hua</a>
	11:45 AM	12:00 PM	High levels of multiple paternity and its importance in captive breeding of freshwater pearl mussels	Sebastian Wacker	<a href="#">S. Wacker</a> , B. M. Larsen, P. Jakobsen and S. Karlsson
	12:00 PM	12:15 PM	Genes or environment? What influence the size at maturity, growth rate and shell shape in thick-shelled river mussel <i>Unio crassus</i> ?	Adam M. Ćmiel	<a href="#">A. M. Ćmiel</a> , A. Lipińska, K. Zając, K. Douda, A. Kilińska, M. Mioduchowska, A. Wysocka, A. Kaczmarczyk-Ziemba, J. Rychlińska, P. Ivinskis, J. Sell, T. Zając
	12:15 PM	12:30 PM	Metabolomics as a tool for assessing multiple stressors in juveniles freshwater pearl mussels ( <i>Margaritifera margaritifera</i> )	Louise Lavictoire	<a href="#">L. Lavictoire</a> and C. West
	12:30 PM	02:00 PM	Lunch		
	02:00 PM	02:45 PM	<i>Keynote: Invasive freshwater bivalves: Impacts, management and future perspectives</i>	David Aldridge	<a href="#">David Aldridge</a>
	02:45 PM	03:00 PM	Living in a thermal plume: life history traits of the invasive Chinese pond mussel ( <i>Sinanodonta woodiana</i> Lea, 1834)	Anna Maria Łabecka	<a href="#">A. M. Łabecka</a> and M. Czarnoleski
	03:00 PM	03:15 PM	Invasion of the Asian clam in naturalized canals: Implications for freshwater mussel conservation	Keiko Nakamura	<a href="#">K. Nakamura</a> , B. Gimeno, J. Guerrero-Campo, E. Elbailek, C. Salinas, J. Boix, M. Á. Muñoz, R. Sousa and F. Mesquita-Joanes
	03:15 PM	03:30 PM	Enemy Release Hypothesis: Parasitism in invasive and native freshwater bivalves	Binglin Deng	<a href="#">B. Deng</a> , R. Nicoletta, P. Nie, M. Urbańska, T. J. Marjomäki, W. Andrzejewski, F. Ercoli, M. Ożgo, K. Nakamura, J. M. Choo and J. Taskinen
	03:30 PM	03:45 PM	Identification of a new gregarine parasite associated with mass mortality events of freshwater pearl mussels ( <i>Margaritifera margaritifera</i> ) in Sweden.	Anders Alfjorden	<a href="#">A Alfjorden</a> , N. Wengström, D. Persson and F. Burki
	03:45 PM	04:15 PM	Coffee break		

Day	Start time	End time	Function/talk	Speaker	Authors
17-09	04:15 PM	04:30 PM	Exploring mysterious mussel die-offs in the Clinch River, USA, using an epidemiological framework	Jeronimo G. Da Silva Neto	<a href="#">J. G. D. Neto</a> , R. H. Hardman, A. C. Engman, G. R. Dinkins, T. W. Lane, M. M. Fry, C. Rines, A. Bisenieks, S. Rajeev and M. M. Dennis
	04:30 PM	04:45 PM	Mass mortality among freshwater pearl mussels ( <i>Margaritifera margaritifera</i> ) in Norway	Jon H. Mageroy	M. Hanssen, J. Vullum and <a href="#">J. H. Mageroy</a>
	04:45 PM	05:00 PM	Investigating Scandinavian freshwater pearl mussel ( <i>Margaritifera margaritifera</i> ) mass mortality events	Daniel A. Cossey	<a href="#">D. A. Cossey</a> , J. H. Mageroy, N. Wengström, O. Kolmakova, S. L. Rock, A. M. Labecka, M. Österling, A. Erlandsson, A. J. Tanentzap, J. Richard, D. C. Aldridge
	05:00 PM	05:15 PM	Towards a network of protected sites for the conservation of freshwater bivalves in South America	Cristhian Clavijo	<a href="#">C. Clavijo</a> , I. Miyahira and A. Bassó
	05:15 PM	05:30 PM	Enhancing Freshwater Mussel Conservation Through Collaboration: Lessons from 20 Years	Stefan A. Fernandes	<a href="#">S. A. Fernandes</a> , S. Heumann, C. Reuter, M. Frisch, T. Eybe, F. Thielen
	05:30 PM	05:45 PM	The project „MARA – Margaritifera Restoration Alliance“: a joint conservation effort for the freshwater pearl mussel ( <i>Margaritifera margaritifera</i> ) in Germany	Marco Denic	<a href="#">M. Denic</a>
	07:30 PM	10:00 PM	Welcome reception		
18-09	09:00 AM	09:45 AM	<i>Keynote: The Parasite Paradox</i>	Niklas Janz	<a href="#">Niklas Janz</a>
	09:45 AM	10:00 AM	Vital allies, not on the menu: freshwater bivalves as regulators and hosts of zoonotic nematode <i>Angiostrongylus cantonensis</i>	Karel Douda	J. Hradečný, D. Pandian, L. Annetová, A. Šípková, J. F.e Escobar-Calderón, M. Mohammadi, K.a Gregarová, D. Modrý and <a href="#">K. Douda</a>
	10:00 AM	10:15 AM	A habitat and a parasite: The impact of parasitic freshwater mussels on habitat choice and predator-prey interactions of a host fish and its prey.	Sebastian Rock	<a href="#">S. L. Rock</a> , A. M. Elmlund, P. A. Nilsson, J. Watz, O. Calles, M. Österling
	10:15 AM	10:30 AM	Exploring Life-History Strategy Linkages Between Freshwaters Mussels and Fish	Irene Sánchez González	<a href="#">I. Sánchez González</a> and K. A. Capps
	10:30 AM	11:00 AM	Coffee break		
	11:00 AM	11:15 AM	Ciliophora endosymbionts of the freshwater mussel <i>Unio crassus</i>	Katarzyna Zając	<a href="#">K. Zając</a> , M. Mioduchowska and T. Zając

Day	Start time	End time	Function/talk	Speaker	Authors
18-09	11:15 AM	11:30 AM	Characterizing the Microbiome of Scottish Freshwater Pearl Mussels ( <i>Margaritifera margaritifera</i> ): A Metabarcoding Analysis using Third Generation Nanopore Sequencing.	Alanna-Jo McCallum	<a href="#">A.J. McCallum</a> , V. Pritchard, B. Morrissey and B. Häenfling
	11:30 AM	12:30 PM	The power of cooperation: from science to policy, from CONFREMUS project to European branch of FMCS	Tadeusz Zajac	<a href="#">T. Zajac</a>
	12:30 PM	02:00 PM	Lunch		
	02:00 PM	02:45 PM	Keynote: Freshwater mollusk conservation in the face of global change: Identifying challenges and solutions	Carla Atkinson	<a href="#">C. Atkinson</a>
	02:45 PM	03:00 PM	Testing mussels' efficiency as a bioremediation tool for eutrophic water bodies: lab experiments	Nicoletta Riccardi	<a href="#">N. Riccardi</a> , S. Kazmierczak, V. Modesto, S. Rinaldi and M. Urbańska
	03:00 PM	03:15 PM	Testing mussels' efficiency as a bioremediation tool for eutrophic water bodies: mesocosm experiments	Sandra Kaźmierczak	M. Urbańska, <a href="#">S. Kaźmierczak</a> , S. Runowski, J. Golski, W. Andrzejewski and N. Riccardi
	03:15 PM	03:30 PM	Mussel-generated dissolved nutrient fluxes promote benthic primary production but denitrification depends on light.	Matthew B. Lodato	<a href="#">M. B. Lodato</a> , T. C. Ledford, J. W. Lopez and C. L. Atkinson
	03:30 PM	03:45 PM	Self-contained benthic chambers allow in situ studies of freshwater mussels' ecosystem impacts	Jonathan W. Lopez	<a href="#">J. W. Lopez</a> , M. B. Lodato and C. L. Atkinson
	03:45 PM	04:15 PM	Coffee break		
	04:15 PM	04:30 PM	Effects of the freshwater mussels <i>Anodonta anatina</i> and <i>Margaritifera margaritifera</i> on water quality, phytoplankton (and zooplankton)	Jouni Taskinen	<a href="#">J. Taskinen</a> , K. Begun and M. Chowdhury
	04:30 PM	04:45 PM	Effect of environmental changes on the selective feeding and fatty acid retention of freshwater pearl mussel	Mahsa Hajisafarali	<a href="#">M. Hajisafarali</a> , M. L. Calderini, M. Kiljunen, N. Moser, S. Nykänen, J. Litmanen, S. Taipale and J. Taskinen
	04:45 PM	05:00 PM	Comparison of three unionid mussel species in removing green microalgae grown in recirculating aquaculture system effluent	Ville Julkunen	<a href="#">V. Julkunen</a> , Č. Stevčić, J. Pirhonen and K. Pulkkinen
	05:00 PM	05:15 PM	Benthic communities and macroinvertebrate-mediated litter decomposition along a mussel density gradient	Raviv Gal	<a href="#">R. Gal</a> , B. McKie, L. Greenberg and M. Österling
	05:15 PM	05:30 PM	Revive, Revitalise, Restore for a more Resilient River Kent: How the LIFE R4ever Kent is tackling a critically low mussel population in Cumbria, UK	Samantha Bonny	<a href="#">S. Bonny</a> , L. Lavictoire, V. Burton, B. King, R. Park, J. Rigby, J. Warren, M. West, E. Wright and M. Barrie
05:30 PM	05:45 PM	Growth and longevity of the endangered freshwater pearl mussel ( <i>Margaritifera margaritifera</i> )	Sabrina Nykänen	<a href="#">S. Nykänen</a> , J. Taskinen, M. Hajisafarali and A. Kuparinen	

Day	Start time	End time	Function/talk	Speaker	Authors
18-09	05:45 PM	07:30 PM	Poster session		
19-09	09:00 AM	09:45 AM	<i>Keynote: Assessing stream health and mussel fitness with in site exposures and shell thin sections</i>	Wendell Haag	<u>W. Haag</u>
	09:45 AM	10:00 AM	Thick Shelled River Mussel ( <i>Unio crassus</i> ) as indicator of pollution	Grita Skujienė	<u>G. Skujienė</u> , J. A. Skuja and I. Nedveckytė
	10:00 AM	10:15 AM	Trace metal uptake in freshwater mussels: untangling biological factors and environmental effects	Kristi Dobra	<u>Kristi S. Dobra</u> , R. Capo and B. Stewart
	10:15 AM	10:30 AM	Analysis of the microstructure of bivalve shells from the Unionidae family – applying new techniques in ecotoxicological research	Maria Urbańska	K. Szpotkowski, J. Golski and <u>M. Urbańska</u>
	10:30 AM	11:00 AM	Coffee break		
	11:00 AM	11:15 AM	Does inshore retention concept explain mussel behavioural preference towards channel margins?	Tadeusz Zajac	<u>T. Zajac</u> , K. Zajac, P. Adamski, W. Bielański, A. Ćmiel, J. Dołęga, J. Florek, A. Strużyński and M. Wyrębek
	11:15 AM	11:30 AM	Frost resistance of <i>Anodonta anatina</i> in the face of climate change.	Anna M. Lipinska	<u>A. M. Lipinska</u> , P. Adamski, A. M. Cmiel, M. J. Golab, D. Halabowski, P. A. Idczak-Figiel, M. Lopes-Lima, J. H. Mageroy, M. Österling, S. Sniegula, A. Teixeira, S. Varandas, M. A. M. Huayas and N. M. M. Zenteno
	11:30 AM	11:45 AM	The impact of fish ponds and land use on freshwater pearl mussel streams	Rebecca Hoess	<u>R. Hoess</u> and J. Geist
	11:45 AM	12:00 PM	Updates on the current conservation status of the two highly endangered freshwater mussels <i>M. margaritifera</i> and <i>U. crassus</i> in Bavaria, Germany	Andreas H. Dobler	<u>A. H. Dobler</u> , P. Hoos and J. Geist
	12:00 PM	12:15 PM	Conservation plan for the endemic freshwater mussel of Morocco: <i>Pseudunio maroccanus</i> (Pallary, 1918)	Fatima M'hamdi	<u>F. M'hamdi</u> , H. Benaissa, F. A. Boumallassa and M. Ghamizi
	12:15 PM	12:30 PM	Living on the edge: challenges for freshwater mussel conservation in Mediterranean-type temporary rivers.	María García Álvarez	<u>M. G. Alvarez</u> , F. Rolo, M. Gonzalez-Gil, F. Godinho, P. Pinheiro, D. Pires, A. M. Ilhéu, F. Banha, M. Gama, P. Anastácio, C. Sousa-Santos, C. Lima, C. Cardoso, C. Carrapato and J. Reis
12:30 PM	02:00 PM	Lunch			

Day	Start time	End time	Function/talk	Speaker	Authors
	02:00 PM	02:45 PM	<i>Keynote: Integrating Phylogeny, phylogeography, and genetic diversity: A comprehensive approach to freshwater mussel conservation</i>	Manuel Lopes-Lima	<u>M. Lopes-Lima</u>
	02:45 PM	03:00 PM	Genetic assessments of endangered freshwater mussels aim to inform conservation management in the Midwestern United States	Claire Waterhouse	<u>C. Waterhouse</u> and K. Inoue
	03:00 PM	03:15 PM	Herbert D. Athearn and the Museum of Fluvial Mollusks	Arthur E. Bogan	J. M. Smith and <u>C. M. Bogan</u>
	03:15 PM	03:30 PM	Conservation genetic units under future climate change scenarios: A case of the threatened Freshwater Pearl Mussel ( <i>Margaritifera margaritifera</i> )	Ilya Vikhrev	<u>I. V. Vikhrev</u> , R. Kuehn, J. Geist, A. V. Kondakov, E. P. Ieshko, O. A. Chelpanovskaya and I. N. Bolotov
	03:30 PM	03:45 PM	The Swedish fauna of freshwater snails – an overview with special attention to red-listed species	Ted von Proschwitz	<u>T. von Proschwitz</u>
	03:45 PM	04:15 PM	Coffee break		
	04:15 PM	04:30 PM	Conservation genetics of the Thick Shelled River Mussel <i>Unio nanus</i> Lamarck, 1819 in Germany with an outlook on <i>Unio crassus</i> Philipsson, 1788	Ira Richling	<u>I. Richling</u> and C. Krause
	04:30 PM	04:45 PM	The freshwater mussels of Central America: Recent research efforts and future conservation plans	Kentaro Inoue	<u>K. Inoue</u> , E. Barraza, V. Melara, Y. Camacho, K. Soto, J. García-Pérez, J. Ortíz, M. Barrios, J. Pfeiffer, S. Keogh, J. Bogantes, A. Sharpe, Y. Quintana
	04:45 PM	05:00 PM	Unraveling Genetic Tapestry: Haplotype Network Analysis of <i>Pomacea canaliculata</i> (Gastropoda: Ampullariidae) in the Seven Lakes of San Pablo City, Laguna, Philippines	Kinsley Meg G. Perez	<u>K. Meg G. Perez</u> , I. K. C. Fontanilla and E. R. C. de Chavez
	05:00 PM	05:15 PM	Building the basis for conservation: Exploring unknown microgastropod diversity in New Caledonia	Nina Tombers	<u>N. Tombers</u> , M. Haase, P. Bouchet and I. Richling
	07:30 PM	10:00 PM	Conference Dinner		
<b>20-09</b>	09:00 AM	04:00 PM	Field Excursion		

# Oral Abstracts

(In schedule order)

## **High levels of multiple paternity and its importance in captive breeding of freshwater pearl mussels.**

Sebastian Wacker<sup>1</sup>, Bjørn Mejdell Larsen<sup>1</sup>, Per Jakobsen<sup>2</sup>, Sten Karlsson<sup>1</sup>

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Multiple paternity can increase within-population genotypic diversity, allowing selection to act on a wider spectre of genotypes, and potentially increasing effective population size. While the genetic mating system has been studied in many species with active mating behaviour, little is known about multiple paternity in sessile species releasing gametes into the water. In freshwater mussels, males release sperm into the water, while eggs are retained and fertilized inside the female (spermcast mating). Mature parasitic glochidia are released into the water and attach to the gills of fish where they are encapsulated until settling in the bottom substrate. We used 15 microsatellite markers to detect multiple paternity in a wild population of the freshwater pearl mussel (*Margaritifera margaritifera*). We found multiple paternity in all clutches for which more than two offspring were genotyped, and numbers of sires were extremely high. Thirty-two sires had contributed to the largest clutch (43 offspring sampled). This study provides the first evidence of multiple paternity in the freshwater pearl mussel, a species that has experienced dramatic declines across Europe. Multiple paternity in freshwater pearl mussels may be central for maintaining genetic variability in small and fragmented populations and for their potential to recover after habitat restoration. Multiple paternity may also play an important role in maintaining genetic integrity and genetic diversity of target populations in captive breeding.



## **Genes or environment? What influence the size at maturity, growth rate and shell shape in thick-shelled river mussel *Unio crassus*?**

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Shell shape is a very characteristic feature of mollusks, which highly varies among taxa. Enormous morphological plasticity of the shells remains a problem in need of explanation, as it still leads to many cases of misidentification, hampering research on species distribution, conservation, and invasiveness. On the basis of shell morphology, a confusingly large number of Unionida species were described in the past, ca. 4000 in 19th century, 1300 species at the beginning of 20th century (more than 200 separate species in *Unio crassus* in 19th century). However, recently the number of species has been greatly reduced.

The aim of presented study was to test the differences in size at maturity, growth rates and shell shape in *Unio crassus* populations inhabiting 22 European rivers located in 5 European countries, at multiple spatial scales: 1) catchment scale, 2) basin scale, 3) river scale, including also 4) genetic differences between mussel populations to show how much of variance in size at maturity, individual growth and shell shape is explained by either genetic and environment factors.

Statistical analysis (Elliptical Fourier Descriptors, PCA, hierarchically nested GLM, hierarchically nested repeated measures GLM) showed, that *U. crassus* shell size at maturity is influenced by both genetic and environmental factors, but genetic distance between clades was the least important factor, and explained only 1% of the variation, while both river and catchment influences explained about 40% of the variation in shell length. The variation in the growth rate of individuals was explained by the individual's age (80% of variation), and by significant interactions between the age of individuals and the river, and between the age of individuals and the catchment area (26% and 18% of the variation in growth rate, respectively). The results suggest, that both shell size and growth rate in *U. crassus* depend mainly on environmental conditions in specific rivers and catchments, and should be attributed to phenotypic plasticity rather than to genotype.

Statistical analysis also showed, that environmental factors have a significant effect on all of the Principal Components describing the shape of *U. crassus* shells, and that the genetic factor was always the factor that explained the smallest proportion of variation in a given Principal Component. The results suggest that *U. crassus* shell shape also depends mainly on environmental conditions in specific rivers and catchments and should be attributed to phenotypic plasticity rather than to genotype.

## **Metabolomics as a tool for assessing multiple stressors in juveniles freshwater pearl mussels (*Margaritifera margaritifera*).**

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Freshwater mussel populations have been in sharp decline for several decades following pressures such as degraded habitats, altered flow regimes, pearl fishing, host fish declines, declining water quality and, most recently, the complex effects of climate change. Many European countries have well-established conservation breeding programmes for imperilled mussel species, but what are the effects on propagated juveniles of sub-optimal habitat conditions?

This study used untargeted metabolomics to examine the metabolic effects of high temperature and suspended solids (singly and in combination) on juvenile mussels (10 years old, mean shell length = 27.9 mm). Mantle tissue was analysed using liquid chromatography mass spectrometry and features identified using compound discoverer. From a list of over 8000 potential features, ultimately 19 metabolites were identified as potentially useful biomarkers for stress. Here we examine a sub-set of the metabolites identified in this study and explore the potential mechanisms of stress in juvenile mussels.

**Living in a thermal plume: life history traits of the invasive Chinese pond mussel (*Sinanodonta woodiana* Lea, 1834).**

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Human impacts create habitats that mimic conditions naturally occurring in other climate zones of the Earth. In temperate climates, water bodies that receive cooling water from water plants form heat islands, providing habitable conditions for non-native warm-adapted species. This facilitates the spread of tropical and subtropical water invertebrates, such as the Chinese pond mussel (*Sinanodonta woodiana*), to higher latitudes.

We studied a European population of *S. woodiana* in the channel of a Polish power plant that supplies cooling waters to the Oder River. For this channel, the lowest water temperature was recorded in February (5.9 °C) and the highest in August (30.4 °C). Our results show that the studied population has female-biased sex ratio. Mussels showed signs of reproductive maturity at shell lengths of ~3 cm. Individuals of both sexes continued to grow after sexual maturity, but females grew slower and ultimately reached larger body size than males. We documented the year-round activity of gonads, with the continuous production of mature oocytes and spermatozoa. Two pathways of spermatogenesis were recorded but with different levels of typical and atypical spermatozoa production. Gravid females were observed all-year round and showed multiple tachytictic brooding patterns. Broods consisted of either only eggs, only mature or immature glochidia, or a mixture of offspring at various developmental stages. Individuals with either larger or more convex shells brooded more offspring. The proportion of incubating females, brood size and glochidia size were linked to gonadal activity and this changed seasonally.

## **Invasion of the Asian clam in naturalized canals: Implications for freshwater mussel conservation.**

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Naturalized canals such as Canal Imperial de Aragón (CIA) and Canal de Tauste (CT) (Ebro River basin, Spain) may function as stable habitats for native freshwater mussel species, including the Giant Freshwater Pearl mussel *Pseudunio auricularius* (= *Margaritifera auricularia*, Spengler, 1793), a critically endangered species. Three other species used to live in these canals: *Anodonta anatina*, *Potomida littoralis*, and *Unio mancus*. However, since 2000, gradual declines in mussel densities in the aforementioned canals have been observed. In 2013, a mass mortality episode of *P. auricularius* was recorded in CIA. Furthermore, no living specimens of the other three species have been recorded ever since. Several hypotheses are being considered to explain this rapid decline in mussel density, including, among others, the high density attained by the Asian clam in the last two decades. Samples to assess density and size structure of the Asian clam were collected from 2011 to 2023 using surber samplers (930 cm<sup>2</sup> each). The overall median density of live specimens of the Asian clam in CT increased from 21.5 ind/m<sup>2</sup> in 2011 (n=67) to 842.3 ind/m<sup>2</sup> in 2016 (n=29), and then dropped to 532.1 ind/m<sup>2</sup> in 2022 (n=30). In CIA the median density values in 2016 (n=64) was 813.4 ind/m<sup>2</sup>, decreasing in 2021 (n=24) to 630.7 ind/m<sup>2</sup> and to 369 ind/m<sup>2</sup> in 2023 (n=29). We suggest that the increase in the density of Asian clams may be one important cause for the disappearance of native mussels.

## **Enemy Release Hypothesis: Parasitism in invasive and native freshwater bivalves.**

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The invasion of exotic bivalves can threaten native freshwater mussels (Unionida) and freshwater ecosystems. The Enemy Release Hypothesis (ERH) proposes that reduction of natural enemies will bring competitive advantage to invaders and contrite to the success of invasion. In this study, ERH was tested on the invasive freshwater bivalves *Sinanodonta woodiana*, *Corbicula fluminea* and *Dreissena polymorpha* with perspective of parasitism. First, parasite pressure, measured by parasite taxon richness and sum of prevalences was compared between native and invasive freshwater bivalves living in sympatry in two separate field studies covering a total of 11 European. In the native bivalves the mean site-specific parasite taxon richness was 2.3–3.4 times and the mean sum of prevalences of infection of different parasites was 2.4–2.6 times that of those in the invasive bivalves. Second, the parasite pressure in the invasive bivalves *S. woodiana* and *C. fluminea* was compared between populations in the original range (China, 5 waterbodies) and the invaded range (Europe, 11 waterbodies). For *S. woodiana*, the average site-specific parasite taxon richness in China was 2.1 times and the sum of prevalences of infection was 3.0 times of those in Europe. For *C. fluminea*, the average site-specific parasite taxon richness was 1.3 and the sum of prevalences of infection was 13.8 in China, while all the studied European *C. fluminea* populations were free of parasites. These results indicate a reduction in the parasite pressure among invasive species, and thus support ERH.

## **Identification of a new gregarine parasite associated with mass mortality events of freshwater pearl mussels (*Margaritifera margaritifera*) in Sweden.**

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Bivalves are key players in freshwater systems, largely contributing to good health of these ecosystems. The freshwater pearl mussel, *Margaritifera margaritifera*, is found in Europe and on the East coast of North America. Once common in oxygenated streams, *M. margaritifera* is facing rapid declines in several parts of Europe. Deterioration of water quality or anthropogenic factors has been considered the main causes for these losses. However, parasitic infections have often been neglected and not investigated. In this presentation we report on the discovery of three novel protist lineages found in Swedish populations of *M. margaritifera* experiencing mass mortalities. All lineages are phylogenetically placed in one of the terrestrial groups of gregarines (Eugregarinorida, Apicomplexa) also known to infect many different groups of insects. We found cysts containing single vermiform zoites in the gills and other organs of diseased individuals using microscopy, in situ hybridization and sequencing. This represents the first report of an apicomplexan parasitic infection in *M. margaritifera*, that also may be linked to the observed declines of this freshwater pearl-mussel population. We will present a tentative life cycle from the finding also describing the parasite distribution within the mussel host of different gregarine stages and its potential exit back into the environment.

## **Exploring mysterious mussel die-offs in the Clinch River, USA, using an epidemiological framework.**

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Freshwater mussels are a vital component of riverine ecosystems, playing a crucial role in maintaining their health and functionality. Recently, freshwater mussel die-off events with unknown causes have occurred in two biodiversity hotspots in Tennessee and Virginia, USA. These mortality events reflect a broader trend of the loss of freshwater mussel populations across the United States that is a major concern for biodiversity and ecosystem function. To address these conservation challenges, we are adopting a comprehensive approach that integrates in-situ experiments, extensive field surveys, disease diagnostic techniques, next generation sequencing, and epidemiological analysis to identify drivers of freshwater mussel die-offs. I will discuss ongoing projects and highlight interesting results regarding mussel health and disease. Particularly, I will highlight a study result that revealed a significant association between *Yokenella regensburgei* and lesions characterized by severe multisystemic and multifocal infiltrative hemocytosis with necrosis, consistent with sepsis. Lesions associated with yokenellosis were of sufficient severity and physiological significance to explain mortality in infected hosts. Although our study does not explain the cause of these infections, it confirms that *A. pectorosa* at our study sites are ultimately dying from an infectious disease and that *Y. regensburgei* can be pathogenic in free-living mussels. I will also highlight study results suggesting that microcystin, a toxin produced by cyanobacteria, is not associated with mussel mortality at these sites. By investigating risk factors such as bacterial infections and exposure to environmental toxins, we aim to inform effective strategies for monitoring, diagnosing, and managing diseases in freshwater mussels.



## **Mass mortality among freshwater pearl mussels (*Margaritifera margaritifera*) in Norway.**

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In 2020, freshwater pearl mussels (*Margaritifera margaritifera*) were collected from the rivers Lakselva, Svankilelva and Vollaelva in central Norway, as brood stock for the Norwegian cultivation program. In 2021, the mussels were returned to the rivers. In 2022, mass mortality was discovered in Svankilelva and Vollaelva. Surveys showed that mortality only was elevated downriver from where the brood stock was returned (73.1 and 93.3 % mortality, respectively). In Lakselva, mass mortality was not observed. These findings resulted in surveys in all locations included in the cultivation program. In 2016-2020, mass mortality had been observed in Haukåselva in western Norway and attributed to pollution. Two more had concerning mortality levels. The rest (>20) showed no signs of elevated mortality. Monitoring in 2023, showed that mortality increased to 84.5 % and 99.9 % in Svankilelva and Vollaelva, respectively. In Lakselva, mortality increased from 2.2 to 11.6 %. Lyngstadelva in (north)western Norway was also surveyed and mortality in affected areas was 24.7 %. Later in the year, great numbers of newly dead mussels were reported from both the latter rivers. In Lyngstadelva, the mortality also moved upstream. Since the mortality initially was restricted to the areas downstream from brood stock restocking sites, it strongly indicates that the mass mortality is tied to the cultivation program. All five rivers with confirmed mass mortality, had broodstock collected in 2019-2020. This suggests that the disease agent, likely causing mass mortality, has been introduced into the program recently, possibly from Haukåselva.

## **Investigating Scandinavian freshwater pearl mussel (*Margaritifera margaritifera*) mass mortality events.**

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Unnaturally high levels of freshwater pearl mussel (*Margaritifera margaritifera*) mortality have been observed in four Norwegian rivers since 2022. Mussels from all four populations were used as brood stock in the Austevoll Cultivation Facility for the production of juvenile mussels to be used for restoration purposes. After the completion of breeding in summer 2021 the mussels were released in the upper sections of the respective rivers they were collected from. Since then, mortality rates of up to 99.9% have been observed downstream of the location where mussels were returned. Similarly devastating MMEs of pearl mussel populations are currently occurring in rivers in Sweden as well. Following the extraction of haemolymph samples from mussels, we have undertaken a metagenomic analysis to search for potential pathogens. We are also targeting a hypothesised gregarine parasite previously reported in Sweden with 18S sequencing. We are conducting histology to better characterise the pathology of the disease and search for potential etiological agents. By September we will have some of these results ready to discuss. Not only does this work investigate specific populations but it also begins to fill the general knowledge gap surrounding the drivers of mass mortality events of critically important freshwater mussel populations in Europe.

## **Towards a network of protected sites for the conservation of freshwater bivalves in South America.**

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Freshwater bivalves are a diverse and endangered group worldwide, and South America is no exception. In La Plata Basin, the second largest basin in South America, there are 83 species of bivalves, 29 of which are endemic. In order to identify potential sites for bivalve conservation, we carried out several field surveys throughout the basin, reaching more than 250 locations in Paraguay, Brazil, Argentina, and Uruguay. In each site, bivalves were collected by hand, identified, measured, weighed, and photographed, and water quality parameters (e.g., pH, conductivity) and landscape characteristics, such as forest cover and land use, were registered. An index was created to identify sites whose protection is strategic, considering richness, catch per unit effort, rare species, and species recruitment. Conservation is also a social process. Thus, the support of the local government and people is critical. The areas that bring together ecological relevance and social support were selected to start the network. For the definitive entry of these areas into the network, as well as the next ones, we proposed a workflow that includes a definition of a site for the conservation of bivalves, an admission form, a guide for evaluation, and a handbook for the implementation of the sites. Until now, four sites have been proposed, one in each country surveyed. Contact with stakeholders, local diffusion, and monitoring are some activities we implement at each site. An effective network of protected sites for bivalve conservation is now moving from a distant dream to a possible reality.

## **Enhancing Freshwater Mussel Conservation Through Collaboration: Lessons from 20 Years.**

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Conservation and reintroduction efforts for endangered freshwater mussels have increased in recent years. However, there are still unanswered questions about the biology and ecology of these species. The knowledge gained over the years by researchers and conservationists has been invaluable in improving conservation methodologies. In this regard, the 20 years of experience and expertise at the Mill of Kalborn are particularly valuable to the freshwater mussel conservation community.

During this period, significant data have been collected on the life traits, habitat suitability, and best rearing practices for the thick-shelled river mussel and the freshwater pearl mussel, both reared at the Mill of Kalborn. This information has been crucial in refining rearing practices and reintroducing over 45,000 juvenile mussels across five streams, Our, Sauer, Rulles, Anlier, and Perlenbach, in the Ardennes region of Luxembourg, Belgium, and Germany.

These achievements would not have been possible without the sharing of data and collaboration with other national and international actors in the field. By fostering stronger partnerships, we can share best practices, leverage diverse expertise, and coordinate efforts more effectively to address the complex challenges facing freshwater mussel conservation worldwide.

This talk will highlight the successes and challenges of 20 years of freshwater mussel conservation in Luxembourg, emphasizing the vital importance of collaboration for the future preservation of these species.

**The project „MARA – Margaritifera Restoration Alliance“: a joint conservation effort for the freshwater pearl mussel (*Margaritifera margaritifera*) in Germany.**

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The freshwater pearl mussel (*Margaritifera margaritifera*) is threatened by extinction in Germany. For the first time, all German conservation activities are consolidated within one joint project „MARA – Margaritifera Restoration Alliance“. The ambition of the seven project partners is to stop and invert the negative trend in order to re-establish healthy, self-sustaining populations. A propagation program aims at the stabilization and rejuvenation of the presently small and overaged populations. A thorough monitoring secures genetic integrity of breeding cohorts and tagging experiments document on-site establishment of released juveniles. So far, more than 6,000 juveniles were released to target rivers. Habitat deficits are addressed on various levels mainly targeting reduction of fine sediment loads, restoration of microhabitat structures and increase of water retention rates to buffer effects of climate change. In the densely populated, agricultural German catchments successful restoration strongly depends on a good cooperation with various kinds of stakeholders and on innovative solutions for water retention. The cooperation with a large dairy farm in Saxony allows to test and assess effects and costs of different mitigation measures including among others controllable drainages or sediment traps. In addition to classic public relations work the topic of freshwater pearl mussel conservation is processed artistically, with an art exhibition and a play. Some naturally bred juveniles in single river stretches are a first sign of positive restoration effects. However, many deficits remain to be removed and single adverse events demonstrate the difficulty of controlling complete catchments and the importance of stakeholder awareness.

**Vital allies, not on the menu: freshwater bivalves as regulators and hosts of zoonotic nematode *Angiostrongylus cantonensis*.**

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Freshwater mollusk conservation is crucial to prevent biodiversity loss, including the preservation of vital functional roles of mollusks in aquatic ecosystems. This contribution explores the roles of freshwater bivalves in the transmission dynamics of the globally invasive zoonotic nematode *Angiostrongylus cantonensis* (AC). We performed comparative analyses of four bivalve species native and invasive in Europe (*Anodonta anatina*, *Corbicula fluminea*, *Sinanodonta woodiana*, *Dreissena polymorpha*) and two control gastropod species (*Biomphalaria glabrata*, *Planorbella duryi*) to assess their capacity to eliminate two different AC larval stages from the water column. Moreover, we studied potential nematode development within these mollusks. Both the native and invasive bivalves significantly eliminated the first stage larvae from water and showed no development to the next larval stages (they were not suitable intermediate hosts), unlike both control gastropods. However, all mollusk species, particularly *S. woodiana*, were susceptible to a secondary infection by the more advanced AC larval stage (serving as suitable paratenic hosts). This dual role highlights the importance of freshwater bivalves in terms of ecosystem services, including zoonotic nematode regulation, while also addressing potential health risks for areas where freshwater bivalves are consumed, informing global strategies for waterborne disease management amidst the ongoing species range shifts. Effective conservation strategies will ensure maintenance of ecosystem complexity providing valuable services with impact on biodiversity and human health. The study was supported by Czech Science Foundation [22-26136S].

## **A habitat and a parasite: The impact of parasitic freshwater mussels on habitat choice and predator-prey interactions of a host fish and its prey.**

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Parasitic freshwater mussels are important and highly endangered ecosystem engineers with an array of functions on multiple trophic levels at multiple life stages. While the impacts of adult mussels on separate trophic levels have been studied independently, few have directly tested how the presence of adult freshwater mussels can impact predator-prey interactions (PPI), and fewer still have investigated the impacts of the mussel's parasitic larvae (glochidia) on these interactions. We present a three-pronged lab study in circular flow tanks to mimic two typical stream habitats for the endangered thick-shelled river mussel (*Unio crassus*): a pebble dominated habitat and a rock dominated habitat. Firstly, the preference of bullheads (*Cottus gobio*) for mussel dominated habitats was compared between fish infested or not by glochidia. Secondly, the preference of a gammarid (*Gammarus pulex*) for mussel dominated habitats was tested in the presence or absence of chemical cues from bullheads. Thirdly, the effect of infestation on bullhead-gammarid PPI was assessed in the presence and absence of adult mussels.

We demonstrate that bullheads only preferred mussel habitats in rocky conditions when infested, and that gammarids only preferred mussel habitats in the absence of predators. The presence of adult mussels significantly reduced bullhead predation on gammarids, whereas infestation did not affect bullhead-gammarid PPI. Adult mussels provided significant refuge for gammarids and significantly reduced rates of predation. Infestation did not affect bullhead-gammarid PPI, but altered bullhead habitat preference. As infestation typically reduces the swimming performance of host fish, our results suggest that adult mussels may offset the negative effects of glochidiosis. This study is of relevance as it is the first investigation into the cross-trophic community impacts of multiple freshwater mussel life stages simultaneously, and adds further insight into how future freshwater communities may change as global freshwater mussel populations continue to decline.



## **Exploring Life-History Strategy Linkages Between Freshwaters Mussels and Fish.**

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Freshwater mussels and fish are highly diverse and interdependent organisms with contrasting life histories. These taxa can be categorized within a triangular life history framework (opportunistic, periodic, equilibrium) to predict community changes based on population responses to stress, competition, and disturbance. While previous studies have independently assessed mussels and fish within this framework, there is a gap in integrating their host-parasite relationships. This study aims to enhance our understanding of life history relationships between these taxa to improve conservation strategies. We compiled data on 169 mussel species with known fish hosts and 247 host fish species, classifying them into three life history strategies based on trait data and phylogenetic relationships. Our analysis revealed an inverse relationship between mussel and host life history strategies. Equilibrium mussels primarily utilized opportunistic hosts, whereas periodic and opportunistic mussels predominantly used equilibrium fish. Leuciscidae (minnows) were key hosts for equilibrium mussels, with the genus *Cyprinella* being particularly significant. Centrarchidae (sunfishes) were the most frequent hosts for opportunistic mussels, with *Lepomis* as the most utilized genus. Percidae (perches) were prevalent hosts for periodic mussels, with *Etheostoma* being the most common genus. This study provides a foundation for understanding host-mussel interactions within the life history strategy framework, highlighting how fish and mussels may have contrasting habitat preferences based on their life history strategies. Understanding these strategies is crucial for developing integrated co-management approaches for mussels and their fish hosts, representing a critical step towards holistic freshwater mussel conservation.

## **Ciliophora endosymbionts of the freshwater mussel *Unio crassus*.**

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Infections caused by symbiotic single-celled organisms belonging to the Ciliophora group are frequently observed in various species of invertebrates. Freshwater mussels also serve as hosts for these organisms, contributing to their biodiversity. However, our understanding of these relationships is limited. Freshwater mussels are facing high risks of extinction, with ciliate endosymbionts potentially causing diseases and mass mortalities, especially among commercially farmed and restocked populations. The relationship between Ciliophora parasites and commensals in freshwater mussels remains largely unexplored, with many species likely to disappear before they are even discovered. Recently, two genera of Ciliophora, *Conchophthirus* sp. and *Trichodina* sp., were identified in the mantle cavity of the endangered *U. crassus*. Research focusing on the impact of endosymbiotic ciliates on *U. crassus* populations requires a comprehensive understanding of their species diversity, infection rates, intensity, and distribution in different river types. This study involves cloning, sequencing, and functional analysis of Ciliophora genetic material through NGS analysis targeting the hypervariable V4 and V9 regions of the 18SrRNA gene. This approach allows for the identification of taxonomic groups, genera, and species. The majority of observed operational taxonomic units (OTUs) are common cosmopolitan species, although some commensals and potential parasites were also identified. The hierarchical biocomplex formed by Ciliophora in the mantle cavity of freshwater mussels presents an intriguing subject for further investigation into their interactions with the host species.

## **Characterizing the Microbiome of Scottish Freshwater Pearl Mussels (*Margaritifera margaritifera*): A Metabarcoding Analysis using Third Generation Nanopore Sequencing.**

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Bivalves, such as the freshwater pearl mussel (*Margaritifera margaritifera*), are ecological powerhouses, serving as ecosystem engineers, biological indicators, and flagship species in freshwater environments. Despite their significant role in enhancing water quality and benthic habitats, these species are critically endangered and rapidly declining. A major concern is their elusive reproductive success, evident by a lack of juveniles surviving into subsequent generations. Future conservation efforts could benefit from addressing a notable research gap, specifically understanding the role of host-associated microbes in mussels' physiology and ecology. In particular, the Scottish Highlands are crucial for the global preservation of this species, as they harbour many of Europe's remaining viable pearl mussel populations. This study aims to taxonomically characterize the microbiome of these mussels, by addressing two key questions: Does the microbiota composition vary within and between Scottish populations? Do these mussels selectively retain a 'core' microbiome compared to the bacteria found in their aquatic environment. Bacterial species will be sequenced using a metabarcoding approach targeting the 16S rRNA gene, utilizing the MinION (Oxford Nanopore Technology) and Miseq (Illumina) platforms for comparative analysis. Samples will be collected through non-invasive DNA swabbing and eDNA water samples of the surrounding seston. By employing molecular methods, these findings could provide critical insights into the interactions that exists between pearl mussels and their microbial communities. Ultimately, this research aims to shape conservation strategies towards a more holistic approach for protecting this imperilled species, advancing our understanding of freshwater bivalves, and revealing the potential importance of their microbiomes.

## Testing mussels' efficiency as a bioremediation tool for eutrophic water bodies: lab experiments.

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Experiments were performed to evaluate freshwater mussels' potential use as tools to reduce eutrophication/pollution in the frame of Horizon SYMBIOREM Project (ID 101060361) aimed at developing integrated bioremediation solutions capable of removing multiple pollutants at the same time. FMs were chosen for their ability to filter large volumes of water, removing suspended particles (phytoplankton, bacteria, viruses) and contributing to the sequestration and/or transformation of pollutants (e.g., heavy metals, pesticides, microplastics, pharmaceuticals). Since FMs can be effective in promoting plant growth and nutrient absorption within 'Constructed Wetlands' (an ecological engineering solution for removing and transforming pollutants from wastewater) SYMBIOREM aims to develop a floating wetland system with symbiotic cultivation of FMs suspended in the submerged rhizosphere to more efficiently biodegrade or accumulate nutrients and pollutants. Estimation of mussels' effects on water quality was obtained by coupling lab scale and field scale experiments and testing common species (*Unio elongatulus* and *U. tumidus*) with a broad distribution range encompassing Northern, Central and Southern EU. Lab experiments to test mussels' clearance and excretion rates at different temperatures were performed by supplying either mixed natural algae or toxic and non-toxic *Microcystis aeruginosa* strains. To facilitate the extrapolation of laboratory results to the natural context experiments in semi-natural conditions (mesocosms) were performed to evaluate the effects of environmental variables.

**Testing mussels' efficiency as a bioremediation tool for eutrophic water bodies: mesocosm experiments.**

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The use of freshwater mussels has been advocated to improve water quality in eutrophic lakes and to control the dominance of large colonies of phytoplankton and/or cyanobacteria that are beyond the control of planktonic grazers. Due to their ability to reduce algae, zooplankton, heavy metals and regulate the nutrients balance, they can be used as bioremediation tools in polluted waters. Before their practical application it is necessary to quantify their effects in the specific context to verify both the positive (seston removal) and negative (excretion) effects, and to estimate the time and number of mussels necessary to obtain an improvement in water quality. The opportunity was realized with the Horizon SYMBIOREM project (ID 101060361) which aims to combine different approaches for bioremediation. To quantify the effects of mussels on nutrient and algae concentrations we first conducted laboratory experiments under different conditions. To facilitate the extrapolation of laboratory results to natural conditions we subsequently conducted experiments in semi-natural conditions. To this aim we installed specifically designed mesocosms in an eutrophic lake (Kierskie Lake, near Poznań, Poland) that served as a model for environments with similar trophic and climatic conditions. Mesocosm (16 m<sup>2</sup> and 500 kg of weight) was consisting of 6 individual chambers which were used as control (3 chambers without mussels) and treatments (3 chambers with mussels). *Unio tumidus* was chosen because it is a common, generalist, abundant and native species of the area in which the project activities take place. Water samples were taken regularly from the mesocosms to measure the effects of the mussels over time and from the lake to verify that conditions within the mesocosms continued to be representative of the external environment.

**Mussel-generated dissolved nutrient fluxes promote benthic primary production but denitrification depends on light.**

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As filter-feeders, mussels can influence ecosystem function by ingesting food particles from the water-column and translocating nutrients such as nitrogen (N) and phosphorus (P) to the sediment as waste. Excretion of inorganic N and P into the sediment can alleviate nutrient limitation and stimulate primary production, while egestion of organic N in biodeposits may promote microbial-mediated denitrification (DNF)—the conversion of biologically available N to inert N<sub>2</sub> gas. We performed a field experiment to test how mussel species identity, excretion, and biodeposition influenced dissolved N and P fluxes, sediment gross primary production (GPP), and DNF. We conducted a series of light-dark incubations using self-contained benthic chambers to quantify GPP based on dissolved O<sub>2</sub> fluxes, and DNF based on N<sub>2</sub> fluxes. We also incubated individual mussels to determine their excretion rates. Excreted N and P led to increased N and P concentrations inside the chambers in the dark, but not in the light, suggesting that excreted nutrients were being taken up by photoautotrophic organisms. We observed a positive relationship between excreted N and GPP, which further supports the hypothesis that benthic autotrophs were taking up and using mussel-derived N. DNF rates were positive in the dark and negative in the light, suggesting N<sub>2</sub>-fixation by autotrophs. However, DNF did not vary among species treatments, indicating that mussel-mediated influences on DNF were negligible compared to their impacts on GPP. Measuring mussel-mediated influences on biogeochemical pathways across varying environmental contexts enables better predictions of the consequences that losing species will have on ecosystem function.

## **Self-contained benthic chambers allow in situ studies of freshwater mussels' ecosystem impacts.**

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Mollusks can drive ecosystem function and provide important ecosystem services through mechanisms tied to their ecophysiological and behavioral traits. In systems where their abundance and biomass are high, freshwater mussels can support the flow of energy and resources through the ecosystem via physiological processes like nutrient excretion and respiration, and through burrowing behavior. Here, we outline a novel benthic chamber design used to quantify mussels' ecosystem impacts in the field and present a case study demonstrating the design's use. First, we discuss the methodology and performance of the self-contained chambers, which can be installed directly into the benthic zone to evaluate mussels' impacts on freshwater ecosystems. Then, we outline an experiment we conducted to test how mussel traits mediate their influence on stream metabolism. Each chamber contained one of four monospecific mussel treatments with contrasting thermal tolerance traits. We quantified stream metabolism in each chamber during summer and fall incubations. We also tracked individual mussels' burrowing movements. We found that temperature, biomass, and excretion explained more variation in metabolism than species identity. Further, thermally sensitive treatments had higher ER than thermally tolerant treatments in the summer. Greater movement appeared to increase background ER via bioturbation. Our findings indicate that mussels' ecophysiological and behavioral traits constrain their impacts on benthic metabolism. Quantifying mussel-driven ecosystem impacts and the environmental conditions that constrain them provides scientific grounds for their conservation and helps predict the consequences that ecosystems stand to endure with the global defaunation of freshwater mollusk communities.

## **Effects of the freshwater mussels *Anodonta anatina* and *Margaritifera margaritifera* on water quality, phytoplankton (and zooplankton).**

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Freshwater mussels are effective filter feeders, performing significant global ecological functions and providing valuable ecosystem services. I present results of two studies on the effects of freshwater mussels on water quality, phytoplankton (and zooplankton). First, we examined effect of the duck mussel, *Anodonta anatina*, on phytoplankton and water quality in a 48 h mesocosm experiment. Second, we studied effect of the endangered freshwater pearl mussel *Margaritifera margaritifera* in a field study where we compared profiles of water quality parameters, phytoplankton and zooplankton along a 500-m section downstream from the headwater lake of a *Margaritifera* river and a control river. *A. anatina* substantially decreased total phytoplankton cell count to one fifth of the control, but selectively, so that the density of small Cryptophyceae flagellates (e.g. *Cryptomonas* sp.) decreased by a factor of 1:15, and density of big diatoms decreased to half in mussel containers compared to control, while no effect on blue green algae and on the rest of algae species combined. Mussels also increased nitrogen and phosphorus concentrations, as well as turbidity, and decreased pH of the water in experimental containers. In the field study, contrasting trend between the *Margaritifera* river and control river, verified with regression analysis results, indicated that a dense *M. margaritifera* bed may reduce dissolved oxygen, chlorophyll-a, blue-green algae, phytoplankton density and zooplankton density. These studies suggest important ecosystem and water quality effects by *A. anatina* and *M. margaritifera* when occurring in high density, and highlight the potential of lost ecosystem functions when mussel populations decline.



## **Effect of environmental changes on the selective feeding and fatty acid retention of freshwater pearl mussel.**

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Land use activities in a catchment can increase nutrient load in river and, therefore, affect the availability and quality of food sources for filter feeders. Freshwater pearl mussel (*Margaritifera margaritifera*, FPM) is a critically endangered keystone species of oligotrophic headwater rivers. Essential fatty acids are vital biomolecules that consumers usually need to obtain from their diet, as the body cannot synthesize them. Therefore, dietary intake is essential for maintaining overall health. Consequently, we analyzed the fatty acid compositions in the stomach content of FPM and their diet (seston) to explore how anthropogenic activities in catchments affect their availability and retention. Environmental factors (26 in total), that reflect anthropogenic pressure, such as chlorophyll-a, nutrients concentrations, intensity of forestry activities, the extent of ditched areas, the size of conservation areas, and latitude were assessed. The fatty acid compositions and content in seston were compared to the ingested fatty acids in stomach content of FPM. Anthropogenic activities increased the availability/contribution of phytoplankton fatty acids in the diet of FPM, while the contribution of terrestrial decreased. Moreover, by comparing fatty acid from seston and stomach content, we found that FPM selectively fed on phytoplankton. Additionally, rivers with less anthropogenic activities showed reduction in contribution of phytoplankton fatty acids but higher reliance on bacterial sources. FPM actively retained physiologically essential fatty acids, underscoring a strong dietary reliance on these fatty acids. Our findings revealed that filter-feeder mussels may rely on essential dietary fatty acids and anthropogenic activities can modify how these nutrients are retained.

## **Comparison of three unionid mussel species in removing green microalgae grown in recirculating aquaculture system effluent.**

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Global increase in aquaculture production has created a need to reduce its environmental impacts. Nutrients could be recycled especially at land-based recirculating aquaculture systems (RAS) by cultivating green microalgae in aquaculture effluent. However, microalgae are difficult to harvest. As a multi-trophic solution, mussels could be used in harvesting microalgae. We tested three European freshwater mussels (duck mussel *Anodonta anatina*, swan mussel *A. cygnea*, and swollen river mussel *Unio tumidus*) for filtering two common green microalgae (*Monoraphidium griffithii* and *Selenastrum* sp.) grown in RAS effluent. Mussels decreased microalgal concentrations in the tanks 42–83% over three consecutive trials. Algal concentrations at the end of each trial were lowest for both microalgae in tanks containing *Anodonta* mussels. Clearance rates were higher for *Anodonta* mussels than for *U. tumidus*. Mussels biodeposited more microalgae to tank bottoms when *M. griffithii* was filtered. Ammonium concentration decreased or did not change in tanks with *M. griffithii*, but increased in tanks containing *Selenastrum* sp. These results suggest that of the tested species *Anodonta* mussels and *M. griffithii* show best potential for RAS effluent bioremediation application. We conclude that a co-culture of microalgae and unionid mussels could be used for recycling nutrients in aquaculture

## **Benthic communities and macroinvertebrate-mediated litter decomposition along a mussel density gradient.**

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The indirect effects of freshwater bivalves on the rates of leaf decomposition, an important process in carbon cycling, have recently begun to be explored. *Margaritifera margaritifera*, like many other freshwater bivalves, was an abundant ecosystem engineer with potential effects on ecosystem functioning in many streams, but has experienced substantial losses in numbers and reproduction capacity and is currently endangered. We conducted an in-situ manipulative experiment in a Swedish stream by placing thirty 27 L mesh enclosures containing coarse gravel and fine (for measuring microbially decomposition) as well as coarse (for measuring total decomposition) mesh bags with birch (*Betula*) leaves, which were stocked with 0, 3, 6, 12 or 24 *M. margaritifera* mussels. The enclosures were deployed three months between November and January, at temperatures averaging 2.2 °C. Macroinvertebrates on the leaf litter as well as in the gravel were collected, identified and counted. We expected decomposition to be faster in proximity to mussels, due to fertilisation and priming effects by mussel excretes and biodeposits, and that the benthic invertebrate community living in proximity to mussels will differ from controls both taxonomically and in regards to species traits. We found that mussels had a subtle effect on the macroinvertebrate community, and, unexpectedly, that the presence of mussels was associated with total decomposition rates that were lower than those in control enclosures without mussels. We hypothesise that the reason for the difference in decomposition rates is caused by macroinvertebrate detritivores preferentially feeding on mussel biodeposits instead of leaf litter.

## **Revive, Revitalise, Restore for a more Resilient River Kent: How the LIFE R4ever Kent is tackling a critically low mussel population in Cumbria, UK.**

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The river Kent is designated as a Special Area of Conservation for the freshwater pearl mussel (*Margaritifera margaritifera*), white clawed crayfish (*Austropotamobius pallipes*), bullhead (*Cottus gobio*) and *Ranunculus* habitat. The freshwater pearl mussel population is now at a critically low level; fewer than 10 individuals are known to survive in the catchment. We are focussing conservation action to restore degraded habitats, carrying out mussel population translocations, improving habitat connectivity by removing barriers such as weirs, cooperating with and supporting land-owners/managers to improve water quality throughout the catchment and engaging with the public to raise awareness about the threats facing the River Kent's unique wildlife.

Here we will discuss the range of activities being carried out to restore and revitalise the River Kent catchment, and how we are approaching population reinforcements/reintroductions of the freshwater pearl mussel, considering that the critically low population number does not allow for a feasible conservation breeding programme using mussels native to the River Kent.

## **Growth and longevity of the endangered freshwater pearl mussel (*Margaritifera margaritifera*).**

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Gaining information on key life-history traits, such as growth and age, is crucial for effective conservation of imperilled freshwater mussel species. However, traditionally growth and age studies require large sample sizes covering all age classes, which can pose risks to populations of conservation concern. To avoid destructive sampling and to overcome the constraints of low sample sizes, we reconstructed the shell growth of 98 critically endangered freshwater pearl mussel (FPM) individuals from 34 populations across Finland and Sweden. We also compared the performance of six different growth models using the reconstructed size-at-age data across FPM juvenile and adult life stages. The growth reconstruction model showed reasonable skill in reconstructing FPM growth patterns. The von Bertalanffy model served as a good general descriptor of growth for FPM but systematically underestimated the asymptotic size. The power law model was the most accurate in estimating juvenile growth. FPM exhibited great variability in longevity ( $A_{\max} = 54\text{--}254$  years) and growth constant ( $0.018\text{--}0.057$  year<sup>-1</sup>) within our study area. Our results demonstrate that reasonable estimates of growth can be obtained even with extremely limited sample sizes. The results can be further applied to gain knowledge on the population's age structure, size at maturation, and recovery potential. The methodology used in our study is applicable not only to FPM but also to other freshwater mussel species of conservation concern. In summary, our approach provides a valuable alternative to destructive sampling, allowing researchers to gain insights into mussel life-history traits while minimizing risks to vulnerable populations.

## **Thick Shelled River Mussel (*Unio crassus*) as indicator of pollution.**

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Bivalves are often named as bioindicators of pollution. In Lithuania, according to the 2016 inventory data of *U. crassus*, it became clear that its condition can vary greatly even in one and the same river, and it remained unclear to which environmental parameters it reacts most sensitively. In order to clarify this, research was carried out in three Lithuanian rivers: one river with an uneven *U. crassus* population (Mituva) and two rivers at a distance of 60-100 km with abundant populations. The aim of the work: to assess and compare the condition of *U. crassus* in the Mituva, Ašva and Uošna rivers, determining which parameters could have influenced their abundance. In the Mituva River, a total of surveys were carried out and substrate and water samples were taken in nine places, in Uošna river – in 4 places, in the Ašva river – in 5 places. It was found statistically significant mean Kendall's Tau negative correlations with six parameters: the alkalinity, the concentration of dissolved solid particles, the salinity, the conductivity, the concentration of ammonium and the concentration of calcium and magnesium ions. A weaker negative correlation was found with dissolved oxygen content, nitrate and phosphorus concentration. It has been confirmed that the *U. crassus* is sensitive to various pollutants and changes in the chemical parameters of water quality: in the Mituva river, where *U. crassus* was not found, the values of all measured hydrochemical parameters were increased, compared to those places where the *U. crassus* was abundant.

## Trace metal uptake in freshwater mussels: untangling biological factors and environmental effects.

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To interpret multi-decadal records of environmental conditions preserved in the aragonite shells of freshwater bivalves, the biological factors affecting metal uptake must be understood. We measured trace metal concentrations and barium (Ba) and strontium (Sr) isotopes in freshwater bivalve shells of 7 species from 4 streams in the Ohio River watershed (Pennsylvania, USA), including the invasive *D. polymorpha* and *C. fluminea*. We also analyzed extrapallial fluid, seston, and river water to refine how trace elements are partitioned and isotopically fractionated between the environment and the shell. To control for variation in environmental conditions, different species were collected at the same time from the same stream. To control for species differences, *A. ligamentina* and *C. fluminea* were both collected from different streams of varying chemistry.

Our results show that freshwater bivalve shells are faithful recorders of the radiogenic Sr isotope composition of their home stream; however, Ba isotope fractionation between shell and river water ( $\Delta^{138}\text{Ba}_{\text{shell-water}}$ ) varies by up to 0.9‰. The magnitude of the offset increases with slower growth rates and varies with species, suggesting that Ba isotope fractionation is occurring during ion transport or shell mineralization. Preliminary results also indicate that significant differences exist in trace metal uptake among species, and that metals, including Mn, Ba, Ca, P, Zn, and Cu, are highly concentrated in extrapallial fluid relative to river water. These results provide insight into the biochemical processes that lead to shell biomineralization and the mechanisms by which different freshwater species incorporate metals from their surrounding environment.

**Analysis of the microstructure of bivalve shells from the *Unionidae* family – applying new techniques in ecotoxicological research.**

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The shell of bivalves functions as an external skeleton, but it can also be used for storing harmful substances. The shell's microstructure depends on the mussel's condition; therefore, analyzing it allows for assessing its health and the impact of various external factors on its physiology. The general microstructure of mussel shells is quite well known, but the impact of the environment on shell microstructure still needs to be better understood. Using techniques such as small-angle X-ray scattering (SAXS), wide-angle X-ray scattering (WAXS), and powder diffraction opens new possibilities in studying shell microstructure and indirectly in ecological research. SAXS and WAXS techniques give information about the structure of proteins that form the organic part of the bivalve shell and the influence of calcium carbonate on the structure of the organic part. The powder diffraction method allows for determining the structure of microcrystals of calcite and aragonite from the inorganic part of the shell. Our research used shells of *Unio tumidus* from two Polish populations. SAXS/WAXS and powder diffraction data were collected at synchrotron beamlines in Hamburg (DESY, Germany).

For analysis, fragments of shell and ligamentum were used. In addition to SAXS/WAXS and powder diffraction, protein electrophoresis (SDS-PAGE) was also used to determine the proteins' quantitative and qualitative aspects from the shell's organic part. It was determined that the applied techniques allow for a precise analysis of the microstructure of bivalve shells, demonstrate population differences, and are suitable for assessing the impact of environmental pollution on the conditions of bivalves.



## **Does inshore retention concept explain mussel behavioural preference towards channel margins?**

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High species diversity in rivers are related to zones of higher retention, where ecological processes are intensified (specific structural properties, deposition and storage, transformation and production of matter, lateral exchange). We studied the distribution and behaviour of freshwater mussels by means of mapping them on cross-sectional transects and by detailed mapping of movements of radiomarked mussels. In most of the rivers freshwater mussels occupy channel margins, i.e. retention zone. This is easy to explain: retention zone contains area of seston accumulation and is inhabited by dense populations of host fish. In mountainous rivers it can also be explained by lower risk of being dislodged by flood. However, in lowland river, where spates are much more mild this risk do not occur. In our study of *Unio crassus* in lowland river, radiomarked mussels distributed in the middle part of the channel escape towards the channel banks, where they stay and reproduce, whereas thalweg is an area of their very fast mortality. This result is very important considering the goals and design of broadly adopted practices of river training works, mostly oriented towards narrowing flow to deepen channel, i.e. intensifying conditions characteristic for thalweg.

## Frost resistance of *Anodonta anatina* in the face of climate change.

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Climate change affects species distribution, causing both contraction and expansion of geographic ranges. This is crucial for endangered species like freshwater mussels. Despite being relatively buffered, shallow freshwater environments can freeze to temperatures below the thermal tolerance of some benthic organisms, including large freshwater mussels. Resistance to low temperatures is key in regulating species ranges.

The research aimed to examine phenotypic plasticity traits—body size, condition, and supercooling point (SCP)—across environmental gradients (latitude and altitude) in the common freshwater mussel, *Anodonta anatina*. Widely distributed in the western Palearctic, this species is expected to show varied traits due to costly cryoprotectant production and differing climatic zones. We hypothesized that in harsh climates (northern Europe and mountains), nutrient deficiencies and low temperatures lead to freezing tolerance and a narrow range of reaction norms. In moderate climates (shorter, milder winters), mussels would exhibit smaller body sizes due to the temperature-size rule.

Mussels were collected in winter across lowland and upland sites, with 353 individuals sampled. Measurements included shell size and shape, supercooling point (SCP), and glycogen concentration in tissues. Results indicate that *A. anatina* shows significant variability in shell shape but not in SCP. Only 10 individuals (2.83%) could supercool: 3 from Central Europe and 7 from Northern Europe, with a freezing point around -1°C. None from Southern Europe supercooled. Preliminary results suggest that *A. anatina* may adopt variability in frost survival strategy, not only across environmental or geographical gradients but also within populations.

## **The impact of fish ponds and land use on freshwater pearl mussel streams.**

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(Fish) ponds are integral parts of river catchments, affecting water retention, sediment and nutrient dynamics as well as the thermal regimes of stream systems they are connected to. However, their impact remains controversial in the context of freshwater mussel conservation. We quantified pond effects over more than 2 years within two catchments containing both > 500 ponds and populations of the endangered freshwater pearl mussel (*Margaritifera margaritifera*) – a species which is particularly sensitive to fine sediments and changes of hydrology and temperature. Nearby ponds increased summer stream temperature by up to 5.5 °C in receiving streams, with the spatial extent of the temperature increase also strongly depending on the riparian land use and groundwater contribution via the hyporheic zone. Nutrient and fine sediment inputs from ponds over most of the time and across various discharges were typically small compared to other sources such as agricultural land use. In contrast, pond drainage - in particular during the fish harvest - released large quantities of fines to the receiving streams, especially in situations when no mitigation measures were applied. Mitigation measures such as physical settling structures and adjusted harvesting methods significantly reduced the fine sediment deposition rates in the receiving streams. In the context of freshwater pearl mussel conservation, these impact of ponds need to be accounted for to maximize benefits and minimize potential negative impacts of fish ponds on the habitat quality for mussels and their host fish.

## **Updates on the current conservation status of the two highly endangered freshwater mussels *M. margaritifera* and *U. crassus* in Bavaria, Germany.**

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The two highly endangered mussel species *Margaritifera margaritifera* and *Unio crassus* are target species of conservation. Based on a recently completed systematic state-wide monitoring of each 22 *M. margaritifera* and 22 *U. crassus* streams in Bavaria, Germany, we present an update on population trends, conservation status, habitat quality and threats for both species.

Populations status and habitat quality varied strongly between *M. margaritifera* and *U. crassus* streams, but there was also great variability within each of those groups. The population decline of *M. margaritifera* has continued, albeit higher proportions of juveniles originating from artificial breeding programmes have established in some streams. Habitat quality often did not match known requirements as evident from poor stream bed quality, lack of hosts and elevated nutrient levels.

In contrast, *U. crassus* showed a better status, with an increase in population size over all sampled streams. Successful recruitment was indicated by high proportions of juveniles. However, no mussels older than 16 years were found, probably due to predation and maintenance measures. Climate change effects, such as extreme droughts, affected both species.

This study demonstrated different needs in conservation management for both species. While for *U. crassus*, tackling direct threats such as predation and drought needs to be prioritized, restoration of habitat quality is key to enable natural reproduction in *M. margaritifera*.

**Conservation plan for the endemic freshwater mussel of Morocco: *Pseudunio maroccanus* (Pallary, 1918).**

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*Pseudunio maroccanus* is an endemic species of freshwater bivalve belonging to the Margaritiferidae family and found in Morocco in Oum Er Rbia and Sebou basin. Over the last 100 years, *P. maroccanus* has been under constant threats: habitat degradation, climate change, water pollution and decline/extinction of host fish, resulting in a massive reduction in its population size, with a continuous decline. This loss contributed to classify this species as being critically endangered by the IUCN, and is now considered as one of the 100 most endangered animal species in the world.

Our work mainly focuses on Laabid river, where most of populations are located and are subject to many threats. In this context, the work consists in a characterization of water quality using a water quality index based on physicochemical parameters and on macroinvertebrates. A riparian strip quality index is also used to evaluate the degree of habitat degradation. The work also consists of re-evaluating the distribution of the species while estimating its density using the C.P.U.E method. A statistical analysis is conducted in the form of a PCA to understand the correlations between the density, mean weight and mean length of *P. maroccanus* individuals located with the physicochemical parameters.

Given that this work is part of a funded project by IUCN SOS program and Segre Foundation and aims to undertake a similar approach carried out in Europe to preserve freshwater bivalves, the work includes activities consisting of workshops with local populations in order to raise awareness of the importance of this species and to include them in all the aspects of this conservation plan.

## **Living on the edge: challenges for freshwater mussel conservation in Mediterranean-type temporary rivers.**

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Temporary Mediterranean-type streams exhibit high natural variability in hydrological conditions, characterized by droughts in summer and flooding in winter. However, climate change trends predict decreasing rainfall and increased temperatures, leading to more frequent and intense droughts. Organisms inhabiting these ecosystems are adapted to extreme events, but drastic changes may push them to their limits, increasing extinction risks. This situation may particularly affect organisms with strict habitat requirements, such as freshwater mussels. For decades, rivers in the southwestern Iberian Peninsula have been experiencing significant alterations related to climate change. This region is home to freshwater mussels' endemic species with a restricted distribution area. The risk of water stress in these systems is increasing, which could be catastrophic for these organisms and their fish host. This study aimed to identify key factors influencing freshwater mussel community dynamics in temporary Mediterranean-type streams of the Southwestern Region of the Iberian Peninsula. Specific objectives included characterizing pool systems associated with mussel communities, assessing the role of fish hosts in mussel distribution and abundance, and determining mussel vulnerability to extreme droughts. Results highlight the critical role of fish hosts in shaping mussel distribution and abundance along streams, and the higher hydraulic instability of smaller streams, with implications for conservation strategies. Understanding the vulnerability of mussel populations to extreme droughts is crucial for effective conservation efforts in the face of climate change. These findings contribute to the broader understanding of freshwater ecosystem dynamics and inform targeted conservation actions to safeguard vulnerable mussel species in the Mediterranean Region.

## **Genetic assessments of endangered freshwater mussels aim to inform conservation management in the Midwestern United States.**

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North America is a global source for freshwater mussel biodiversity, home to around 300 species. Despite being a historic hotspot for freshwater mussels, the United States has seen widespread decline across various species resulting from water pollution, habitat and host loss, competition with invasive species, and climate change. The complex life histories of freshwater mussels can leave them extremely vulnerable to such threats in their first few years of life. Consequently, in a bid to revitalize threatened populations, conservation groups have turned to captive breeding and reintroduction programs for many freshwater mussels. However, these programs often forgo genetic assessments to ensure that natural genetic diversity is maintained. In partnership with state and federal agencies, we use ddRADseq and GT-seq methods to quantify the genetic diversity and structure of propagated populations, including broodstock, glochidia and juveniles of multiple federally endangered species (*Cumberlandia monodonta*, *Lampsilis higginsii*, *Quadrula fragosa* and *Simpsonaias ambigua*). By comparing these data with natural populations, we evaluate the amount of genetic diversity retained in propagated populations. Subsequently, we develop genetic management guidelines for conservation agencies to optimize the success of recovery programs and safeguard the natural diversity of endangered species.

## **Herbert D. Athearn and the Museum of Fluvial Mollusks.**

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Herbert D. Athearn (1923-2011) was an avid amateur shell collector. Today he would be a citizen scientist. He was born in Fall River, Massachusetts, U.S.A. Herb began keeping fieldnotes on his freshwater collecting in 1941, interrupted by service during World War II. After the war, Herb returned home and, for seven years, volunteered with Dr. William J. Clench in the Mollusk Section of the Museum of Comparative Zoology, Harvard University, learning the basics of museum curation. In 1955, he and his family moved to Cleveland, Tennessee where he spent the rest of his life working primarily for the U.S. Postal Service. All his spare time, weekends, and holidays were spent either collecting freshwater mollusks across the southeastern United States or curating his collection, which he called the Museum of Fluvial Mollusks. His collection was well curated and spans between 1850 and 2005, with 23,344 cataloged lots containing over 3,000 lots of imperiled and extinct taxa. He traded extensively with collectors worldwide obtaining specimens from 84 countries. His collection was donated to the North Carolina Museum of Natural Sciences in 2007. Approximately 60 % of this collection has been digitized, totaling an estimated 585,000 specimens. The collection consists of freshwater bivalves, primarily Unionidae and Margaritiferidae, and the gastropods, represented by 40 families, with the greatest abundance representing the freshwater Pleuroceridae.



**Conservation genetic units under future climate change scenarios: A case of the threatened Freshwater Pearl Mussel (*Margaritifera margaritifera*).**

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Freshwater Pearl Mussel is a keystone species that facilitates a number of important ecosystem functions in coldwater oligotrophic streams, while is currently facing habitat alteration and degradation due to human activities. Global climate change is amplifying these negative impacts. Population decline is directly connected with transformation of population genetic structure and may result in significant loss of genetic diversity. In this study, we identified populations of *M. margaritifera* for conservation prioritization in the north-eastern part of the species' range and investigated the potential impact of future climate scenarios on its conservation genetics of the species. We used nine microsatellite loci to examine genetic diversity and differentiation in 17 populations of the species from rivers of the Barents, White, and Baltic Sea basins. To forecast the effects of climatic changes on the dynamics of genetic structure, we applied the Bayesian clustering algorithm. Genetic simulations were conditioned to mean summer temperatures estimated under future carbon emission projections, i.e., representative concentration pathway (RCP) scenarios 4.5 and 8.5. Obtained data allowed us to delineate three conservation units that play critical roles in securing the genetic diversity of the species currently and in the future: 1) populations of the Barents Sea basin with the highest contribution to the overall genetic diversity; 2) populations of northern Karelia which will lose significant part of their diversity under the climate change; and 3) populations of the Baltic Sea basin, of which their unique gene pool of which can be threatened by climate change and other anthropogenic pressures.

## The Swedish fauna of freshwater snails – an overview with special attention to red-listed species.

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The Swedish freshwater snail fauna comprises 49 species: 37 Hygrophila (Eupulmonata); 8 Neritimorpha and Caenogastropoda and 4 Valvatoidea. 43 species are indigenous, and 6 introduced by man, of the latter 4 have so far only been found in artificially heated waters.

8 species can be considered as having a throughout distribution in the country. Approximately 17 species have their northern limit in Middle Sweden, more or less pronounced, in the biogeographical transition zone known as *limes norrlandicus* (e.g. *Stagnicola corvus*, *Planorbis carinatus*, *Acroloxus lacustris*, *Martoniopsis insubrica*). A few species also have their southern limit or becomes rare in the same zone (e.g. *Gyalus acronicus*). Local conditions, especially water chemistry, limit some species to eutrophic areas, and hence these show a split-up distribution picture. Examples of such species are *Segmentina nitida* and *Valvata macrostoma*. A few species are extremely local and today possibly extinct (*Anisus vorticulus*, *Bithynia transsilvanica*). In some cases, the unclear, or only recently cleared taxonomy, makes the distribution and abundance unclear (e.g. the *Radix/Peregriana/Ampullaceana*- genera group, *Stagnicola* spp. and *Gyalus acronicus*/*G. stromi*).

The latest Swedish red-list (2020) comprises six freshwater snail species, they are generally geographically restricted (e.g. *Omphiscola glabra*, *Valvata macrostoma*), and/or ecologically restricted with special demands, and are threatened by habitat destruction (e.g. *Valvata sibirica*).

## **Conservation genetics of the Thick Shelled River Mussel *Unio nanus* Lamarck, 1819 in Germany with an outlook on *Unio crassus* Philipsson, 1788.**

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The Thick Shelled River Mussel *Unio crassus* s. l. is one of the most endangered unionid species in Europe. Providing a baseline for the conservation of genetic diversity, potential reintroduction and enforcement of populations, the genetic constitution of almost all surviving populations of *Unio nanus* in Southwestern Germany was investigated. With 48 populations from Baden-Württemberg and seven from Hesse and Rhineland-Palatinate totalling in >1.500 individuals from >200 sites, the sampling covered the drainage systems of Rhine, upper Danube and Weser.

Molecular methods applied included sequencing the maternal mitochondrial COI haplotypes, analyses of microsatellites as well as Next Generation Sequencing with RADseq.

Overall, the different datasets revealed similar patterns and results regarding genetic diversity, diversification and connectivity of populations as well as the differentiation across the range with an expectedly diverging depth of resolution for the three methods at the different levels of analysis from individuals to drainage systems. Data split into four major clusters reflecting the drainage systems of Weser, Upper to Middle Rhine, the confluences of Lake Constance, and the Danube with the exception of the populations of the High Rhine and upper Neckar River which cluster with the Danube populations.

Results presented will focus on the RADseq data and as outlook a comparison will be given to the preliminary results on the populations of *Unio crassus* from Northern Germany.

The project in Baden-Württemberg was supported by the Stiftung Naturschutzfond Baden-Württemberg and the „Fischereiabgabe des Landes“; for N-Germany by the EU project (LIFE Bachmuschel) and Stiftung Naturschutzfonds Brandenburg.

## **The freshwater mussels of Central America: Recent research efforts and future conservation plans.**

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Central America has a unique assemblage of freshwater mussels, serving as a faunal transition zone between North and South America. This region has species from Unionidae and Mycetopodidae, many of which are believed to be endemic. However, outdated taxonomy and a lack of contemporary field surveys continue to impede efforts to understand the total species diversity and distribution of mussel fauna in the region. This lack of information also hinders the assessment of the conservation status of these species and the development of effective conservation strategies. To address these gaps, we have conducted multiple field excursions in rivers and lakes in Costa Rica, El Salvador, Guatemala, and Panamá to document the presence, abundance, and distribution of freshwater mussels and collect voucher specimens, including genetic materials. Preliminary phylogenetic analyses indicate that taxonomic reviews and revisions are likely required for many Central American species. Furthermore, as part of our capacity-building activities for mussel conservation, we hosted the first regional workshop in Guatemala. This workshop showcased our survey efforts, including hands-on field surveys in local rivers, and trained undergraduate students and early-career scientists in specimen vouchering and molecular genetic techniques using an Oxford Nanopore MinION sequencer. Our future plans include expanding our research to encompass further field surveys, molecular genetics, and taxonomy, and completing the IUCN Red List assessments for Central American species. We also aim to enhance our capacity-building activities through continued training and mentoring of students and early-career scientists and forging partnerships with local and regional conservation organizations.

**Unraveling Genetic Tapestry: Haplotype Network Analysis of *Pomacea canaliculata* (Gastropoda: Ampullariidae) in the Seven Lakes of San Pablo City, Laguna, Philippines.**

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Invasive alien species are threats to native biodiversity, and one of the world's 100 worst is the golden apple snail, *Pomacea canaliculata*. In more than 40 years after its introduction in the Philippines, the possible origin and genetic relationship with other known populations is still poorly understood. To address this research gap, phylogenetic and haplotype analysis of *P. canaliculata* from the seven maar lakes of San Pablo City, Laguna was conducted. A total of 150 sequences (50 cytochrome c oxidase I/cox1 gene barcodes from the seven lakes and 100 GenBank sequences) were used to generate a phylogenetic tree and median-joining haplotype network map. Basic Local Alignment Search Tool (BLAST) analysis confirmed 98-100% similarity of snails from the seven lakes with those from China, Malaysia, and Papua New Guinea. Around 31 haplotypes were obtained from 14 countries. Median-joining haplotype network map revealed four haplotypes (Hap 2, 22, 30, and 31) from the seven lakes. Hap 22 is a unique haplotype observed in Pandin Lake which clustered with specimens from China. Distinct haplotypes were also detected in Calibato Lake (Hap 30) and Mohicap Lake (Hap 31). Hap 2 is shared among all lake samples except for Pandin, and has clustered with those from South America, Asia, and Australasia. In this study, we demonstrated that DNA barcodes could aid not only in accurate species identification, but also provide clues on the possible geographic origin which can be a basis for the control and management of *P. canaliculata* in tropical lake ecosystems.

## **Building the basis for conservation: Exploring unknown microgastropod diversity in New Caledonia.**

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The island of New Caledonia in the South Pacific is known as a biodiversity hotspot with a high level of endemism due to its combination of ancient elements and island radiations. The “Lacs du Grand Sud” region with tree-dominated wetlands, shrub-dominated marshes, and an intricate network of permanent and seasonal freshwater bodies has been listed as a RAMSAR site for its unique pseudokarst geomorphology on peridotite bedrock supporting a highly specialized endemic flora and fauna.

Invasive species, wildfires, and an opencast mining environment pose significant threats to this unparalleled flora and fauna, and unprotected drainages surrounding the RAMSAR site are at risk of conversion into mining in the future. Despite being highly vulnerable, the freshwater microgastropods of the family Tateidae remain essentially unstudied in the region and may become extinct even before leaving a record in the scientific literature.

We are studying samples from 144 locations in the Grand Sud within and around the RAMSAR site, collected during several expeditions as part of the “Planète Revisitée” program. Utilizing shell characters alongside an initial cytochrome oxidase I phylogeny, we estimate that there are in the order of 100 species in the 30 x 30 km Grand Sud – nearly all undescribed –, including cryptic species and distinct genetic clades translating into unknown genera – all endemic to New Caledonia.

Documenting these new species and analyzing their potential range sizes, we assess the degree of microendemism and estimate the threat levels faced by these microgastropods, laying the groundwork for future conservation measures.

# Poster Abstracts

(Alphabetical order)

## **Population reinforcement of the freshwater pearl mussel (*Margaritifera margaritifera*) in Cumbria (NW England).**

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Freshwater mussel populations are in sharp decline and are considered to be one of the most imperilled groups globally. Consequently, the number of captive breeding programmes has increased rapidly in recent years, coupled with subsequent reintroductions/population reinforcements to reverse these declines. Following a small-scale pilot release of freshwater pearl mussel (*Margaritifera margaritifera*) in a declining population in the River Irt in 2017, juvenile mussels were PIT tagged and released across four sites in the River Irt in 2021 (1,300) and 2023 (1,100). Subsequent monitoring found high levels of retention at three out of the four release sites, despite the occurrence of a significant flood event during October 2021. As part of Natural England Species Recovery Programme, 1000 further releases are planned at the same three sites in the River Irt for summer 2024 to support this population, and 350 juveniles are planned to be released in three sites on the River Ehen to reinforce this population. Methods employed to carry out a successful conservation translocation of the freshwater pearl mussel in a declining population in northwest England have been documented, which can be used to design conservation translocation strategies for other imperilled pearl mussel populations in the UK and throughout Europe.

## **Floodplain lakes: a promissory opportunity for bivalve conservation.**

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Freshwater bivalve conservation is a challenge due to 1) the reduction of population sizes and historical distribution, 2) the extension of threats, and 3) the lack of plans for invertebrate conservation in protected areas. To start addressing these issues, we aimed to identify environments priority for bivalve conservation. We compare the richness, biomass, and abundance of bivalves between floodplain lakes (FLs) and watercourses (WAs). We used two sources of information: a survey in 20 FLs and data recorded from 81 WAs distributed to represent all the basins in Uruguay. A one-hour manual sampling effort was implemented at each site, and the individuals were identified, measured, and weighed. The results were compared using Mann Whitney U test. In total, 18 species were recorded in the FLs, representing 45% of the species cited for Uruguay. Five FLs and ten WAs had no bivalves and were not considered in the analysis. The richness, biomass, and abundance were significantly higher in FLs than in WAs. Furthermore, in the WAs, the abundance and biomass of invasive species were significantly higher than native species. In contrast, in FLs, the abundance and biomass of native species were significantly higher than those of invasive species. Another advantage of FLs over other environments is the temporary isolation from threats of the basin. Thus, these environments are reservoirs of species that can contribute to the diversity and recovery of the main watercourses. In this context, we propose FLs as environments priority for bivalves conservation in Uruguay.



## **A tool for standardised, rapid mussel health assessment in the field.**

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To address the precipitous declines of many freshwater mussel populations globally, it is necessary to apply informative tools for measuring mussel health, understand baseline measurements with these tools, and develop a systematic programme for monitoring populations to detect and take steps towards alleviating potential stressors. To do this, a suite of stress biomarkers has been applied to mussels; however, baseline measurements of these biomarkers are often not available, no framework exists for the translation of these methods from research into routine mussel health monitoring, and they are impractical for giving rapid health measurements in the field. We developed a standardised measure of valve closure strength and showed that it is correlated with well-established bioindicator assays for stress. Our method utilises a specially designed pair of tongs that can bend when opening a mussel to give a numerical reading of tong displacement. We have shown that this value is loosely correlated with acetylcholinesterase activity, a common stress indicator, in mussels at physiological conditions. We are currently testing this correlation in mussels that have been heat-stressed and will have these results ready for the presentation. Our method takes only a few seconds per mussel so can be applied rapidly in the field. It provides a standardised reading that can be compared amongst mussels in a population over time, allowing the detection of stressed mussels. We envision the wide application of this technique in rapid mussel health monitoring in the future.

**A global comparison of the microbiome of an invasive clam (*Corbicula fluminea*).**

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One hypothesis explaining the success of invasive species is their relationships with microorganisms (their microbiome), which include both their mutualistic symbionts and the pathogens they might carry and transmit to competitive native species. Although the importance of the microbiome of invasive species is well established in plants, it has received little attention in animals. To work towards filling this gap, we aim to improve our understanding of the dynamics of the microbiome of *C. fluminea* with regard to its spread around the globe. We will compare multiple invaded sites and native sites to determine if there is a core microbiome common to *C. fluminea* from across the globe or if the microbiome changes completely to match the environment. We also intend to investigate if any specific species may be providing *C. fluminea* with success in its invaded range. Currently we have or intend to collect samples from New Zealand, the United Kingdom, France, Portugal, Italy, the USA, the Philippines, Canada, and Argentina for our global analysis. We will have some preliminary method testing results to share along with outlining our plans in a poster format for the conference. This work is not only of academic interest but has the potential to generate predictions regarding where *C. fluminea* may be able to spread in the future and could be applied to control measure development.

## **Interactions of invasive and native freshwater mussels.**

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Biological invasions are major threat for biodiversity including freshwater mussels. Effects from biotic interactions such as competition for food, space or host fish, physiological impairment or predation are often not fully understood, yet such knowledge is crucial for both invasive species control and native mussel conservation.

We examined these interactions, focusing on the non-native Chinese pond mussel, *Sinanodonta woodiana*, in Bavaria, Germany. Laboratory experiments were used to assess predation effects from native and invasive crayfish on three native and one invasive mussel species.

Field monitoring revealed a widespread distribution of *S. woodiana* in Bavaria, mainly associated with fish ponds. The results indicate that *S. woodiana* has a broad ecological niche. In laboratory experiments, invasive signal crayfish showed stronger predation on native mussels, such as the thick-shelled *Unio pictorum*, than on *S. woodiana*. *Sinanodonta woodiana* was also less susceptible to predation by native crayfish. The observed functional interactions of *S. woodiana* likely have a profound effect on native unionids, as they often overlap in habitat and host fish use.

We propose that research and monitoring efforts should more intensively include non-native mussel species and their interactions with native fauna. Such work is not only essential for a better understanding and mitigation of biological invasions, but also highly relevant for conservation of native mussel communities.

## Higher mobility of freshwater mussels in response to unsuitable habitat conditions

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Although typically considered sedentary, unionid mussels can move and their burrowing and crawling behavior is an integral part of their ecology. The European *Unio crassus* is usually found in faster flowing streams with coarse substrates, while *Anodonta* sp. tends to be more abundant in calmer waters with finer substrates. The objective of this study was to examine whether movement distances and burrowing depth of *U. crassus* and *Anodonta* sp. would vary between three different habitats in a first order stream in southern Germany with a longer (8 weeks) and shorter (48 hours, controlled for substrate) field experiments. We hypothesized that mussels should be the most mobile (i.e., larger movement distances) in presumably unsuitable habitat where they are rare. Both species moved the longest distances and were most mobile at a sidearm location where both species were rare or absent, and that had the lowest depth and flow velocity. While *U. crassus* burrowed significantly deeper at the other faster flowing sites, burrowing of *Anodonta* sp. was more varied and did not significantly differ between sites. The results suggest that low water depth may have triggered higher mobility in both species. This could be an adaptation to cope with drying of parts of the water body with higher mobility allowing the mussels to encounter deeper water. A higher mobility of *U. crassus* at the sidearm location compared to *Anodonta* sp. suggests a stronger response to unsuitable habitat conditions which may be typical for lotic species in disturbance-prone systems.

## **One species' gain, another's pain? Effects of dreissenid fouling on a native mussel species under different thermal conditions**

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Native freshwater unionids are among the most endangered species in North America and Europe. With the ongoing spread of invasive species, functional interactions such as fouling and competition for food by the invasive zebra mussel *Dreissena polymorpha* can pose major threats to native species. In this study, the physiological impacts of *D. polymorpha* fouling on the native European unionid *Anodonta cygnea* was tested at different water temperatures of 12, 19, and 25 °C. The laboratory experiment was conducted over a 28-d period, including three treatments: fouling of *A. cygnea* by *D. polymorpha*, both species present but spatially separated, and a control in which *A. cygnea* and *D. polymorpha* were placed in separate tanks. Decreased tissue glycogen, glucose, lipid and protein concentrations indicated that higher water temperatures caused physiological stress in *D. polymorpha*. In contrast, *D. polymorpha* benefited from fouling unionids as evident from increased storage substances. However, elevated temperatures did not aggravate the competitive effects of *D. polymorpha* over *A. cygnea*. Possible confounding factors were glochidia release during the experiment, lower infestation intensity, and physiological stress of *Dreissena* at higher temperatures. This study demonstrates the usefulness of considering physiological endpoints in analyzing consequences of species interactions, especially in the context of assessing future climate change impacts.

## **DECOMPOSITION OF LEAF LITTER ENRICHED WITH ALGAE AND MUSSEL BIODEPOSITS**

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In freshwaters, the enrichment of recalcitrant material with labile organic matter can have positive or negative effects on decomposition rates, depending on the context. How the presence of freshwater bivalves and their provisioning of organic matter to the benthic environment via biodeposition affect leaf litter decomposition has likewise had contrasting results in field studies. We conducted a microcosm study using 90 microcosms, each containing two birch (*Betula*) leaf litter portions, one of which was exposed to the generalist detritivore *Asellus aquaticus*. Microcosms were enriched with dead algae, dead algae mixed with a mineral component, and/or biodeposits produced from the same mixes by the freshwater mussel *Unio tumidus*, or not enriched at all, and were deployed for 69 days at ca 14.5 °C. Exposure to feeding by *A. aquaticus*, as well as its interaction with the algae treatment both increased decomposition rates, while enrichment on its own did not increase microbially mediated decomposition.

## **Does the presence of *Sinanodonta woodiana* and mussel macroparasites influence the pattern of use of European mussels by the European bitterling?**

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Parasitic infections, are ubiquitous, and their impact on hosts may play a role in ecosystem processes and affect many species, including freshwater mussels. Additionally, parasite-host relationships in the presence of alien species and their impact have not been well qualified. The following assumptions were tested: (1) the presence of the alien freshwater mussel *Sinanodonta woodiana* influenced the pattern of use of mussels by the bitterling, itself a parasite of unionid mussels, (2) there is a negative relationship between the parasitism of the bitterling and the presence of non-bitterling parasites. Polish populations of all native and alien species of mussels from the Unionidae family were examined. The overall prevalence of bitterling eggs and embryos was 18.8% with high variability among mussel species. The number of bitterlings was significantly higher in species of the *Unio* genus. All native mussel species were used by the bitterling (larger individuals were used more often), while in the examined individuals of *S. woodiana*, no eggs and embryos of the bitterling were found. Among other macroparasites water mites were most frequently found in mussels, followed by trematodes, oligochaetes and non-biting midges. Among native species, the presence of the bitterling in mussel gills was most strongly affected by host species identity. There was no significant influence of the occurrence of *S. woodiana* and macroparasites on both the use of native mussel species by the bitterling and the number of bitterlings in mussels. However, the presence of glochidia had a negative impact on the bitterling clutch size.

**EU Project “LIFE Bachmuschel” - Conservation and reintroduction of the thick-shelled river mussel (*Unio crassus*) in Brandenburg (Germany).**

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The population of the once widespread thick-shelled river mussel (*Unio crassus*) has been declining dramatically, to the extent of being listed as “critically endangered” in Germany and its state Brandenburg respectively. Through the EU project "LIFE Bachmuschel", the Foundation Naturschutzfonds Brandenburg and its partners, the Institute for Inland Fisheries e. V. Potsdam-Sacrow and the State Museum of Natural History Stuttgart, will conserve, increase, or reintroduce the population of *Unio crassus* in Brandenburg's rivers.

The aim of the project is to improve the living conditions and to expand suitable habitats for *Unio crassus* in rivers and streams of the Elbe, Havel and Spree catchment areas. By creating sediment traps in the water, planting hedges and riparian trees, threats such as fine sediment and nutrient influx are reduced. Adding gravel and dead wood to the river restores structure to the bed and leads to flow diversity.

Rivers are upgraded as spawning habitat for mussels and their host fish. The relocation of water bodies to old course structures are also planned.

The genetics of the occurrent populations will be considered as a basis for targeted population support and reintroduction for *Unio crassus* during the 2023-2032 project period. By artificially infesting host fish with glochidia, *Unio crassus* is reintroduced and/or reinforced in areas where the species has become extinct, or still exists but in low and declining numbers. Implementation of host fish stock support and reintroduction activities improve the abundance of local fish populations and thus the reproduction success of *Unio crassus*.



## **Freshwater mussels of Republic of North Macedonia (Bivalvia: Unionidae): an introduction to their distribution, status and conservation**

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Knowledge about the distribution, habitat preferences, conservation status, and primary threats to unionid mussels across Europe is highly inconsistent, showing substantial variations in data quantity between countries. Moreover, some parts of Europe like R. North Macedonia, are completely omitted in the previous investigations. In this study we present the results from the comprehensive biological investigations on the Unionidae species in R. North Macedonia conducted over the last decade. The presence of the unionid mussel was investigated by detail search of the bottom of rivers, river banks and lakes all over the country. All collected specimens were returned to their natural habitat shortly after counting and photographing them. Special attention was dedicated to the thick-shelled river mussel *Unio crassus* as it is considered a species of community interest in the country, listed on the Annex II of the EU Habitats Directive and requires designation of special areas of conservation (SACs). Unfortunately, bearing in mind that unionid species like the habitats in which they are encountered are still being in desperate need of conservational attention, this investigation aims to provide information about these species’ distribution in Republic of North Macedonia and the threats their populations are facing in order to internationally engage in managing freshwater mussel diversity, applying incentives to freshwater ecosystems protection.

## **Using movement patterns of captive-bred freshwater pearl mussel to evaluate habitat suitability of release sites**

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The ongoing declines in freshwater pearl mussel (*Margaritifera margaritifera*) populations have led to the initiation of captive breeding programs across the species' distribution range. However, most pearl mussel streams have been impacted by anthropogenic degradation of channel morphology and hydrologic regimes, making the selection of suitable release sites difficult. We explored the potential of using the movement patterns of released captive-bred freshwater pearl mussels as indicator of habitat suitability in restored and unrestored stream areas. Movement of captive-bred freshwater pearl mussels was tracked with PIT (passive integrated transponder) tags over one year after their release into three different habitat types of two streams: a) areas with colonies of adult mussels, b) randomly chosen sites with monotonous flow pattern over mobile sand, and c) restored sites with introduction of boulders and gravel. Detection rates remained high at all three sites for both streams, even after strong winter floods. Movement of released mussels was lowest at sites already occupied by adult mussels, indicating a preference of such areas. In contrast, mussels at the random stretches moved largest distances, with some individuals recovered more than 20 m downstream. Some mussels released in the restored stretches moved longer distances than in the habitats with adult mussels, but their movement was moderate compared to randomly chosen stretches, representing a search for optimal micro-habitats. The results of this study demonstrate that mussel movement can be a powerful indicator for assessing habitat quality, evaluating restoration success, and guiding the selection of release-sites.

## **Reproductive success of *Pseudunio auricularius*' juveniles (Spengler 1793): first data**

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*Pseudunio auricularius* is one of the most endangered freshwater mussel species in the world. In Spain, it has suffered a strong decline since 2013, when a massive mortality episode was recorded. Captive breeding developed by the Aragón Government (Zaragoza), allow the production of  $>10^6$  juveniles yearly, out of which 95% are release into the Ebro River and naturalized canals, and 5% remain in the laboratory under controlled conditions. Ten juveniles from 2017 and 2019 cohorts (mean shell length: 6 cm) were transferred in March 2023 to the glochidia collection system used for the adults specimens. They were monitored all together during one month and for the first time we have verified the start of the hitherto unknown reproductive cycle of *P. auricularius*. In 2024, 13 juveniles were monitored separately or in pairs, recording the release of glochidia in one specimen from the 2017 cohort and four from the 2019 cohort. The maximum estimate of glochidia released by one specimen was 35,085. The glochidia were used to infest six Siberian sturgeons (*Acipenser baeri*), which produced 48,835 viable juveniles that continue their growing process in captivity.

## **Effects of parasitic freshwater mussels on their host fishes: a review.**

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Freshwater mussels in the order Unionida are highly adapted to parasitize fish for the primary purpose of dispersal. The parasitic larval stage affixes itself to the gills or fins of the host where it becomes encysted in the tissue, eventually excysting to develop into a free-living adult. Research on the parasitic interactions between unionids and their host fishes has garnered attention recently due to the increase in worldwide preservation efforts surrounding this highly endangered and ecologically significant order. With the exception of heavy infestation events, these mussels cause minor effects to their hosts, typically only observable effect in combination with other stressors. Moreover, the range of effect intensities on the host varies greatly with the species involved in the interaction, an effect that may arise from different evolutionary strategies between long- and short-infesting mussels; a distinction not typically made in conservation practices. Lower growth and reduced osmotic potential in infested hosts are commonly observed and correlated with infestation load. These effects are typically also associated with increases in metabolic rate and behaviour indicative of stress. Host fish seem to compensate for this through a combination of rapid wound healing in the parasitized areas and higher ventilation rates. The findings are heavily biased towards *Margaritifera margaritifera*, a unique mussel not well suited for cross-species generalizations. Furthermore, the small body of molecular and genetic studies should be expanded as many conclusions are drawn from studies on the ultimate effects of glochidiosis rather than proximate studies on the underlying mechanisms.

## **Post-parasitic survival and growth of juvenile *Unio crassus* in semi-natural and laboratory conditions.**

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Understanding the in-situ survival of unionoid mussels after parasitizing their host fish is vital for conservation of threatened species. However, there is limited research on the survival and growth of juvenile thick-shelled river mussels (*Unio crassus*), particularly regarding adaptation to local conditions, differences between semi-natural and laboratory environments, and the impact of specimen handling. This study examined the survival and growth of 1,550 three-week-old *U. crassus* juveniles over three months, using a combined field and laboratory approach. The juveniles were placed in modified Whitlock-Vibert boxes (TuBox) in three rivers: their original river Bråån, the habitat-restored Klingavälsån in the same catchment targeted for mussel re-introduction, and Vramsån, a river outside the catchment with vital populations of *U. crassus* and their primary host fish. Additionally, a subset of juveniles was kept in laboratory conditions using water and detritus from the three rivers. Contrary to expectations, the highest survival rate (72%) occurred in the laboratory, with water from Vramsån. These mussels also exhibited the most significant growth. Field-survival was highest in the warm Klingavälsån, where juveniles showed a 50% survival rate and increased shell growth (up to 30%). However, their survival dropped with frequent handling. This study highlights the need to consider temperature regimes of donor and recipient rivers, monitor juvenile survival in the field, and use low-impact methodologies for successful unionoid re-introduction efforts.

**Novel measures of cumulative sequential beta and gamma diversities reveal effects of environment and human pressure on freshwater mollusk communities along a gradient.**

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Along a river's course, line transect, or time series the beta ( $\beta$ ) diversity usually characterizes the overall species turnover. But the habitats' features and anthropogenic pressures rarely run linearly or monotonically; most often, they vary in disruptive, recursive, and even chaotical ways, mirrored by the trajectories of community dynamics. We introduced the sequential  $\beta$  diversity earlier, which measures the successive magnitude of species turnover and ecological changes along a gradient. Here, we define the cumulative sequential  $\beta$  diversity, which assesses gradient changes by adding and comparing the effect of each new sample to those already registered before or upstream. All structural changes (common, absent, and newly encountered species) are used in the formula, and the values are arranged within a matrix of dissimilarities, which in turn is related to environmental and spatial descriptors using uni- and multivariate canonical ordination analyses and corresponding diagrams. Adding communities' alpha ( $\alpha$ ) diversity, we show a novel way to compute the newly introduced sequential cumulative gamma ( $\gamma$ ) diversity index as a synthetic approach in gradient analysis of life-environment dynamics. Using data on freshwater mollusks along a river, we show how to relate these measures to different predictors (environmental, spatial, and human impact descriptors) and how to compare and graphically illustrate the actual trajectory with extreme scenarios. The use and significance of variation partitioning methods are also shown. The new measures and models might be used in diversity patterns analysis and have recognizable importance in malacological research, human impact assessment and biodiversity conservation.

## **Sensitivity of juvenile *Margaritifera margaritifera* to hypoxia – a laboratory experiment**

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The freshwater pearl mussel is critically endangered. Low survival of juvenile individuals is one of the main reasons for decline of *M. margaritifera* populations. Low survival of young *M. margaritifera*, on the other hand, is attributed to oxygen depletion in the stream substrate due to anthropogenic siltation. However, the actual hypoxia tolerance of juvenile *M. margaritifera* has remained almost unstudied. To effectively conserve this endangered species, we need a better understanding of the effects of hypoxia on juvenile mussels. Juvenile FPM (7–8-month-old) were exposed to different levels of dissolved oxygen at 5, 10 and 17 °C and their viability was monitored for 10 days to assess the acute hypoxia tolerance of juvenile FPM. Dissolved oxygen concentrations ranged between 11.67 mg/l and 6.79 mg/l in the high oxygen treatments (control), 6.47–2.16 mg/l in the medium treatments, and 1.07–0.03 in the low oxygen treatments. The viability of juvenile FPM was significantly affected by the oxygen concentration and temperature, such that all juveniles in high and medium oxygen concentrations survived, while in low oxygen treatments 100 % of juveniles at 17 °C, 80 % at 5 °C and 13 % at 10 °C became non-viable. Thus, very low oxygen concentrations, near anoxic conditions, are lethal to *M. margaritifera* when temperature is high, but in lower temperatures some of the juveniles can survive up to 10 days even in near anoxic conditions.

## Behavioural responses of Iberian freshwater mussels to drought conditions

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Freshwater mussels are one of the most endangered faunal groups worldwide. In Mediterranean areas, anthropic impacts and, recently, climate change have been contributing to the decline of their populations. This study assessed the behavioural responses, through movement and habitat use, of 4 native mussels, *Anodonta anatina*, *Potomida littoralis*, *Unio delphinus* and *Unio tumidiformis* to drought conditions. Experiments were carried out **1) in the laboratory**, using ramps, to simulate the slope of the riverbanks, and manipulating the water level through 3 dewatering rates a) slow: 4 cm/day; b) moderate: 8 cm/day; and c) fast: 4 cm/hour); **2) in situ**, through daily and seasonal monitoring of the movement and habitat use of tagged individuals. Significant differences were found in horizontal and vertical movements between species, with *A. anatina* and *U. delphinus* moving more actively than *P. littoralis*. These movements were more pronounced for the fast treatment. Daily field monitoring revealed immediate movements from the bank to deeper areas, while seasonal monitoring highlighted the importance of available microhabitats for the mussel movement. Efficient water management and protection/restoration of freshwater ecosystems are essential for the conservation of threatened mussels in Iberia. This work was supported by FCT - Fundação para a Ciência e Tecnologia, I.P. by project " EdgeOmics- Freshwater Bivalves at the Edge: Adaptation genomics under climate change scenarios " with the reference PTDC/CTA-AMB/3065/2020 and DOI identifier <http://doi.org/10.54499/PTDC/CTA-AMB/3065/2020>



## **The environmental bill of Mediterranean “Liquid Gold” to riverine ecosystems-ecotoxicology tools to act as early warning signals on crustaceans and bivalves.**

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Nowadays, 80000t of olive pomace is processed in one single extractor located in Northeastern Portugal, producing olive mill wastewaters (OMWW) which are highly phytotoxic. The lack of management measures for OMWW raises concern about the ecological integrity of surrounding ecosystems. Therefore, eight sites along Tua River (Douro basin) and its tributaries were seasonally monitored for a year (2021-2022), following Water Framework Directive standards for biologic and physicochemical elements. Toxicant role of OMWW was assessed using *Daphnia magna*, *Unio delphinus* (native) and *Corbicula fluminea* (invasive) as bioindicator organisms. *D. magna* was subjected to acute tests (24 hours) while bivalves were subjected to chronic (14 days) exposures, respectively. When comparing the obtained results, sites located downstream of oil extraction industry showed a significant decrease in the water quality (higher values of oxygen demand and particularly of phenols) and aquatic habitats (anoxic sediments). Several ecological metrics and multivariate analyses detected the ecological impact of industrial discharges. Additionally, the conducted ecotoxicological assays revealed high toxicity for different trophic levels, causing the death, of at least 50% of the tested *D. magna* population, when exposed to 0.88% OMWW(v/v). After 14 days of exposure, it was observed that OMWW presence led to cellular membrane disruption, confirmed by higher LPO levels. The increase of reactive oxygen species may have triggered the antioxidant enzymes (SOD) and inhibited neuronal activity (AChE). Additionally, invasive species are revealed to be more likely to cope with continuous organic contamination, such as OMWW discharges.

## **Mussels in peril – investigating the effects of a massive nickel discharge in river Kokemäenjoki, SW Finland**

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River Kokemäenjoki runs 121 km from Lake Pyhäjärvi to the Gulf of Bothnia in the Baltic Sea. An accidental release of 66 tons of nickel and 94 tons of sulfate occurred in the river in 2014 from the plant of Norilsk Nickel Harjavalta Ltd in SW Finland.

As a consequence, many thick shelled river mussels *Unio crassus*, a protected species listed in the annex IV of the EU Habitats Directive, among three other unionid species, perished downstream of the discharge point. A national survey was established to find out the mortality and reproduction capacity of the *U. crassus*. Other key questions were whether the mussel populations will recover and what is the role of metals in the mortality.

The survey consists of 36 dive transect across the river downstream of the discharge point and 9 cross transects above it as control sites. The recent mortality for *U. crassus* in 2014 was 15,6 %, or about 1 million individuals. In 2015, mortality had fallen to a few percent. The high mortality of the mussels was caused by nickel poisoning, which affects ion pumps in the mussels.

The fecundity of mussels in 2015 was not significantly lower in the impact area, suggesting that the population may recover in future. Furthermore, there has been a significant positive trend in the reproduction success of *U. crassus* in the latest survey completed in 2023. The population size has increased by a staggering 106 % from 2014. The reason for such a strong increase is unclear.

## **COST Training School - a suitable place to improve new methods**

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Freshwater mussels are endangered worldwide, and their population is still decreasing. One of the reasons cited is the castration of molluscan hosts by larval trematode parasites that destroy or alter gonadal tissue. In this presentation, we compare the efficiency of two techniques to find trematode infection. It can be done by dissection or non-destructive biopsy. The latter is more straightforward and quicker, but the accuracy of the examination depends significantly on the researcher's experience (the valid positive rate is 94.7% for experts vs. 50% for novices). However, when doubts arise in both approaches, histological analysis can resolve them, although with some limitations. However, the likelihood of recognizing an infection at an early phase, even by an experienced researcher, is small. When the infestation is advanced and leads to the destruction of gonads, histological analysis is then needed to recognize sex. These conclusions are based on observation with participants of a training school dedicated to recognizing parasites. The relation between researchers' experience and correct recognition was evaluated using statistical techniques in the examination method. Researchers beginning their careers need more practice to gain experience and confidence in analyzing images before attaining the capacity to produce reliable diagnoses. Unfortunately, there is still a lack of a large dataset of images that can be used for training purposes.

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## Understanding Patterns of Antibiotic Resistance in *E. coli* Isolated from *Anodonta anatina*: An Emerging One Health Concern

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The freshwater bivalve *Anodonta anatina*, due to its ecobiological traits (filters), serves as an effective bioindicator for environmental biomonitoring of the contaminant's effect. This study aimed to characterize the prevalence of antibiotic resistance, genetic diversity, and virulence traits in *Escherichia coli* strains from *A. anatina* collected in 2022 from two sites in Tua River basin, a tributary of Douro River, Portugal, with different levels of human disturbance. Antibiotic susceptibility testing (AST) was conducted using the disk diffusion method on Mueller-Hinton Agar medium, following EUCAST 2023 guidelines. Identification focused on phylogenetic characteristics of *E. coli* through the eight phylogroups using multiplex PCR (Clermont et al., 2013).

Results revealed a prevalence of antibiotic resistance with 60% of isolates exhibiting resistance. Among the 23 antibiotics tested, Site 2 (Barcel) showed the highest resistance incidence. Multidrug resistance was common, with one strain resistant to five antibiotics. Phylogenetic analysis identified diverse groups among the *E. coli* isolates: B1 (55%), D/E (15%), A (10%), E/clade I (10%), with 10% unclassified. No isolates demonstrated pathogenicity.

These findings underscore the importance of *A. anatina* as a bioindicator for monitoring antimicrobial resistance (AMR) in freshwater ecosystems. This study highlights the utility of integrating multidisciplinary methods for comprehensive environmental monitoring, contributing to sustainable freshwater ecosystem management and public health.

Continuous monitoring and innovative strategies are essential to combat AMR, aligning with the One Health approach to mitigate the spread of antimicrobial resistance in aquatic environments, stressing the need for further research and preventive measures to protect public and environmental health.

**Who works best in *ex situ* conservation programmes for *Margaritifera margaritifera*: a sympatric or allopatric host?**

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The host-parasite relationship in naiads is subject to evolutionary pressures that affect the dynamics of the species involved. Subtle alterations can affect the life cycle and cause local adaptations that impact on their viability and need to be understood.

The critically endangered species *Margaritifera margaritifera* is characterised by a long parasitic phase in the gills of native salmonids. In culture programmes, the choice of host species is generally a practical decision, using captive-bred or wild specimens, without assessing whether their sympatric or allopatric origin in relation to the naiad population may influence the success.

Within the *ex situ* conservation plan developed in Galicia (NW Spain), and with the aim of finding out whether the origin of the fish influences its ability as a host, an experiment was carried out by infesting 0+ Atlantic salmon from the same and different basins of origin as the naiad population.

Throughout the parasitic phase, a series of controls were carried out to analyse the prevalence, infestation rate and growth of glochidia in each group. In addition, the survival and growth of reared juveniles was monitored for 70 days and the impact of infestation on the fish was assessed using the condition factor.

Although the results show that cyst growth was significantly higher in the sympatric infestation, the success of the cycle and the viability of the juveniles obtained did not depend on the specificity of the glochidia. The condition factor of the fish was not affected by infestation.

## Habitat functions of dead freshwater mussel shells for macroinvertebrates

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Freshwater mussels are considered keystone species in aquatic ecosystems and the presence of living mussels can enhance ambient macroinvertebrate biodiversity. However, due to a lack of empirical data, the functional role of dead freshwater mussel shells as habitat for other species remains unclear. Drawing primarily from research in marine ecosystems, we hypothesized that mussel shells enrich riverbed structure by providing diverse microhabitats, especially for macroinvertebrates. In a defined field experiment, shells of three native freshwater mussel species (*Margaritifera margaritifera*, *Unio crassus*, *Anodonta* spp.) were exposed in wire cages into three mussel streams over two months, and colonization by macroinvertebrates was compared to controls without shells. At the macroinvertebrates community level, the differences between seasons and streams were greater than the impact from the mussel shells. In addition, there was no general trend across all streams regarding the macroinvertebrate feeding types. However, within streams, there was a positive trend for abundance in the cages with mussel shells for some feeding types like predators. Notably, there was a clear positive effect of mussel shells on specific species, such as the two crayfish species *Astacus astacus* and *Pacifastacus leniusculus*. The findings of this study show that dead shells can provide important microhabitat functions, and therefore should not be removed. Depending on the microhabitat availability of a stream, mussel shells can e.g. be utilized to promote specific target species such as crayfish.