



XXII AIL MEETING  
VIGO 2024



XXII Congress of the Iberian Association of Limnology  
(AIL 2024)

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**23-28 June 2024**

VIGO

## Welcome message

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Dear friends and colleagues,  
Welcome to “Aquatic ecosystems under threat: improving our knowledge to mitigate the effects of global change, promoting adaptive management and involving citizens”, an event joining the XXII conference of the Iberian Association of Limnology (AIL) that will be held in Vigo, (Spain) in June 2024.

This meeting that will be hold for the first time in Galicia aims to gather in one place the limnologist from Spain and Portugal, and wider afield to exchange knowledge and expertise from other European and Latin-American countries. Policy makers and environmental managers, industry members, and all others who take an interest in ecological research and building an environmentally sustainable future are also welcome to join us.

We are facing constant threats and changes of the water ecosystems and we feel like there is an increasing need for limnologist to participate and to be part of the solutions in order to support societal decision-making regarding the current climate of overexploitation of natural resources, habitat loss and degradation, pollution, invasive species, urbanization, climate change, and overall global biodiversity crisis. Ecological research-derived scientific knowledge has played a crucial role in forging socio-environmental links, preserving biodiversity, and promoting cleaner environments for sustainable utilization and development of our natural resources.

In light of the integrative and transdisciplinary nature of ecological research, this gathering actively welcomes contributions from scholars involved in various ecological sub-disciplines, ecosystems, and biological levels of organization. Within this context, our aim is to explore theoretical and experimental approaches that facilitate comprehension of intricate interactions between organisms and their environment.

Our collective intention is to address one of today's most urgent societal concerns: the preservation of functional and resilient ecosystems amidst ongoing change.

With the implementation of all necessary safety protocols, this assembly will provide a valuable opportunity for camaraderie among peers and colleagues, fostering diversity and inclusivity while supporting and motivating students and young researchers within our community. We wish you a productive meeting and pleasant stay here in Vigo!

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## Scientific Committee

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# Sessions

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## Ordinary Sessions

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- OS1** Microbial Ecology
- OS2** Processes and Functioning of Ecosystems
- OS3** Community Ecology
- OS4** Monitoring, Management, and Restoration of Aquatic Ecosystems
- OS5** Exotic and Invasive Species
- OS6** Biogeography and Evolution of Species
- OS7** Ecology of Wetlands, Lakes, and Estuaries
- OS8** Global Change and Aquatic Ecosystems
- OS9** Extreme Aquatic Ecosystems
- OS10** Ecohydrology and Groundwater
- OS11** Aquatic Ecotoxicology and Environmental Risk Assessment
- OS12** Ecosystem Services
- OS13** Urban Aquatic Ecosystems
- OS14** Biodiversity and Conservation of Aquatic Ecosystems

## Special Sessions

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- SS1** Advances in eDNA for the study of freshwater ecosystems: from technical to ecological aspects
- SS2** Exploring ways of Integrated land and water resource management to achieve temporary pond conservation
- SS3** Water management of reservoirs: the challenges of the future generations
- SS4** Research and management challenges on non-perennial rivers
- SS5** Justice, Equity, Diversity, and Inclusion (JEDI) in Limnology
- SS6** Exploring the effects of global change on freshwaters: The crucial role of LTER studies
- SS7** Adaptive approaches for water resources management and wetland restoration



## **Introduction to meta- -analysis in limnology**

**Dr. Verónica Ferreira**

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## **Essentials of eDNA-based freshwater biodiversity as- sessments - the god, the bad, the ugly**

**Dr. Florian Leese**

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## **Scientific illustrations and an introduction to Inkscape**

**Dr. Katrin Attermeyer**

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**Sunday, June 23rd**

**10:00 - 16:00 h**

**Redeiras Building**

## **Workshops**

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## Keynote Speakers

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**Sunday, 23rd June**

19:30 - 20:30

**Beyond hype: Towards the reliable use of DNA-based biodiversity data in freshwater research & application**

**Dr. Florian Leese**

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**Monday, 24th June**

11:00 - 12:00

**Speed bumps and highways: aquatic carbon processing from source to sea**

**Dr. Katrin Attermeyer**

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**Monday, 24th June**

15:00 - 16:00

**Plastic pollution research in aquatic ecosystems**

**Dr. Sonja Ehlers**

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**Tuesday, 25th June**

12:30 - 13:30

**Building blocks for upscaling freshwater restoration: The role of science in a trans-disciplinary challenge**

**Dr. Sebastian Birk**

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## Keynote Speakers

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**Wednesday, 26th June**

11:00 - 12:00

### **Alteraciones ambientales y sus efectos en la biodiversidad de insectos acuáticos en la Amazonia**

**Dr. Leandro Juen**

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**Thursday, 27th June**

11:00 - 12:00

### **Career Award Senior Scientist**

**Dr. María Rosario Vidal-Abarca**

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**Thursday, 27th June**

15:00 - 15:30

### **PhD Award 2022-2023: Hydrological alteration, critical swimming speed and life history in inland fish**

**Carlos Cano Barbacil**

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**Thursday, 27th June**

15:30 - 16:00

### **PhD Award 2023-2024: Linking biodiversity, ecosystem services, and ecological stability for river ecosystem sustainability**

**Ana Paula Senra Portela**

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# Oral Communications

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# OSI

## Microbial Ecology



# Effects of intensity and temporal occurrence of multiple stressors on river biofilm diversity and functions

Javier Ortiz<sup>1</sup>, Paula Siñeriz<sup>1</sup>, Anna Freixa<sup>1</sup>, Sergi Sabater<sup>1,2</sup>

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Mediterranean rivers are exposed to co-occurring stressors, either from anthropogenic activities such as agriculture or industry, or from alterations in temperature and hydrological regime associated to climate change. These multiple stressors may lead to a negative impact on the aquatic biota and their ecological functions in relation to their relative intensity and order of occurrence. We performed a field experiment to assess the effect of hydric stress and urban pollution on river biofilms to evaluate their composition and diversity, extracellular enzyme activities, and biomass. We colonized artificial substrata (ceramic tiles) in four unimpacted streams (2nd - 3rd order) for one month. Afterwards, one set of substrates were left in the river channel (as control), while other two sets were exposed to two different levels of hydric stress, transferred to the stream bank (pulse: short and repeated and press: prolonged and continuous drying). Finally, all substrates were relocated downstream of an urban wastewater effluent (pollution stressor), and we analyzed how biofilm responded to these stressors individually and in combination. Our findings indicate that hydric stress negatively affects microbial communities structure and functions (i.e. Leu-aminopeptidase activity), with no discernible differences between the two levels of drying. Conversely, exposure to the pollution stressor promoted a faster recovery of biofilms in those treatments previously subjected to hydric stress. While we observed changes in structural variables, functional responses remained unaffected. This study underscores the importance of accounting for the combined impacts of multiple stressors when assessing their effects on biofilms and their resilience to disturbances.

# Disentangling the ecology of microalgae in extreme environments: Insights from Iberian inland saline shallow lakes

Rafael Carballeira<sup>1</sup>, Antonio Picazo<sup>1</sup>, Carlos Rochera<sup>1</sup>, Carla Morales<sup>1</sup>, Daniel Morant<sup>1</sup>, Antonio Camacho<sup>1</sup>

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Photosynthetic microorganisms contribute significantly to ecosystem's functioning and overall biodiversity, even at the limits of life. The natural complex of saline inland wetlands of the semi-arid Central Iberian Peninsula offers, in the same geographical area, a complete gradient of extreme saline environments, and even different degrees of eutrophication due to human alteration. Understanding the ecology of microalgae in saline lakes can be very informative for effective management and conservation efforts, especially considering the increasing pressures from human activities and climate change on these unique and fragile ecosystems. We analyze the environmental parameters of water and sediment, characterize the limnology and extracted DNA of water samples across a strong gradient of salinity and human disturbance in Iberian inland saline shallow lakes. High-throughput sequencing of 16S rRNA genes in water and sediment samples was conducted to evaluate the composition and structure of the photosynthetic microbial assemblages, and the DNA metabarcoding data were taxonomically assigned using a customized reference database. We used multivariate statistics to understand the intricate interplay between abiotic and biotic parameters along this pronounced salinity gradient, unveiling the primary ecological patterns and interspecific composition based on data derived from zero-radius Operational Taxonomic Units (ZOTUs) of photosynthetic microorganism. Salinity conditioned the organic matter content of the sediment, and in turn, this affects the availability of nutrient concentration in water column and the structure of microalgae communities. This work was supported by the project CLIMAWET-CONS (PID2019-104742RB-I00), funded by Agencia Estatal de Investigación and the Ministerio de Ciencia e Innovación (Gobierno de España).

# Freshwater mussel holobionts show a high diversity of symbiotic microorganisms across Iberian rivers

Félix Picazo<sup>1</sup>, Andrés Tirado<sup>1</sup>, Joaquim Reis<sup>2</sup>, Ignacio Peralta-Maraver<sup>1</sup>, Silke Martínez-Moreno<sup>1</sup>, Elizabeth León-Palmero<sup>3</sup>, Pedro Abellán<sup>4</sup>, Manuel Jesús López-Rodríguez<sup>1</sup>, José Luis Moreno<sup>5</sup>, Isabel Reche<sup>1</sup>

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The recent advenement of mass sequencing techniques has revealed a huge variety of animal-microbiome interactions. For instance, hosts establish symbiotic relationships with both epi- and endobiotic microorganisms conforming biological entities known as holobionts. While most research done at holobiont level in aquatic ecosystems have focused on marine and transitional water environments, the endangered freshwater ecosystems and their biodiversity has received less attention. Although numerous studies have demonstrated the crucial role that freshwater invertebrates play in ecosystem functioning, we still have little knowledge on the ecosystem functions and services that take place at holobiont level. Therefore, many freshwater holobionts could become extinct before we comprehend their role in ecosystem functioning. This is the case of freshwater mussels, which are suffering one of the most dramatic declines worldwide. Namely, the Iberian Peninsula counts 10 species and most of them are catalogued as endangered because of a severe reduction in their population abundances and former ranges. We here characterize the epi- and endosymbiotic microbiome of 3 native freshwater mussels (*Potomida littoralis*, *Unio delphinus* and *Anodonta anatina*) together with the invasive *Corbicula fluminea*. We found a high prokaryotic diversity in freshwater mussel microbiome, with the epibiotic component showing an environmental signal while the endobiotic component showed a phylogenetic signal. Moreover, these microbiomes presented the potential to provide regulating and supporting ecosystem services related to the nitrogen cycle such as denitrification. In summary, our results serve as a baseline to comprehend ecosystem functioning at holobiont level in freshwaters.



# Microbial mats diversity associated with wetlands in the Canadian Arctic: microbial structure and trophic networks

Carlos Manso<sup>1</sup>, Eugenio Rico<sup>2</sup>, Samuel Cirés<sup>1</sup>, Antonio Quesada<sup>1</sup>, Ana Muñoz<sup>3</sup>, David Velázquez<sup>1</sup>

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Microbial mats are fundamental components of polar ecosystems, where plants and animals are greatly limited by the harsh environments. Microorganisms are major drivers of biogeochemical cycles, influencing both climate and ecosystem functions in polar regions and beyond. Microbial mats harbor a large and diverse pool of organisms and exhibit a vast array of metabolic functions under harsh environmental conditions. The study of trophic relationships of these microecosystems in Arctic wetlands in relation to environmental variability is still under research. In this communication, we study the effects of environmental variability on microbial diversity from two microbial mats located at two different latitudes and associated with wetlands from tundra and taiga biotopes in the Canadian Arctic. We studied the structure and the trophic relationships in microbial mats using DNA analysis by sequencing the 16S rRNA gene, stable isotopes as trophic tracers, microscopic analyses, and photosynthetic pigments analyses. Our results suggested that, based on a Bayesian mixing model, both microbial mats showed different trophic levels composed by primary producers (cyanobacteria, diatoms) and at least two more trophic levels conformed by primary consumers (rotifers, tardigrades) and secondary consumers (nematodes). Metabarcoding analysis also show that both microbial mats differ in their bacterial community's composition. Significant differences have been also found in environmental variables between microbial mats, which explain the variations in pigment contents. Furthermore, development indexes displayed similar rates between mats, suggesting that external environmental conditions shape their structure and trophic relationships, but do not seem to significantly influence their development capacity.

## Two-decades monitoring of the surface layer ciliate assemblage (Slapy reservoir; Czech Republic): What can we state from?

Miroslav Macek<sup>1</sup>, Jaroslav Vrba<sup>1</sup>, Jiří Jarošík<sup>2</sup>, Klára Řeháková<sup>2</sup>

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In the two-decade surface samples (three-weekly since 1994) from the Slapy reservoir (Vltava River, Czech Republic), a ciliate assemblage within the microbial loop was analysed to study the combined effects of climate change and eutrophication (more- A and less-eutrophic B periods were compared). Ciliates were identified using Quantitative Protargol Stain protocol to the genus level. However, we centre the analysis on feeding-behaviour groups. The most important were the nanoplankton filtering mixotrophic genera *Limnostrombidium* and *Pelagostrombidium*, peaking just when the stratification was stabilized. Since a drop of nutrients during B, they were important also until the overturn. The minute picoplankton feeders followed them in importance: *Halteria grandinella*, a mixotrophic *Pelagohalteria viridis*, minute *Rimostrombidium* spp. (some of them with kleptoplasts), and *Vorticella aqua-dulcis*. They also peaked at the end of mixing but another peak during the stagnation period was more pronounced, especially during B. Algivorous ciliates formed the third group, reaching a maximum during the spring phytoplankton peak; a higher biomass was observed during stagnation B: minute one-tailed urotrichas resembling *Urotricha agilis*, double-tailed *U. furcata* or *U. pseudofurcata* and multiple-tailed *U. castalia* or *U. pelagica*. The position of *Balanion planctonicum* and *Histiobalantium* spp. is uncertain: the ciliates repeatedly reached very high biomasses but in less than one-half of years. The results are in concordance with the enhanced (2012) PEG model. However, the surface layer analysis did not provide a reasonable water column ciliate-importance evaluation. If somebody is interested in further monitoring studies, he must include the metalimnion.

**OS2**

**Processes and Functioning  
of Ecosystems**



# Understanding the contribution of wet and dry areas to gaseous carbon exchange in a Mediterranean Intermittent Stream

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Stream hydrological regimes are experiencing pronounced alterations as a consequence of both climate change and anthropogenic influences, leading to prolonged periods of streambed desiccation in many regions like the Mediterranean. These alterations significantly impact biogeochemical processes within stream networks. Until now, mainstream research on the carbon (C) cycle within aquatic systems has focused on inundated zones, thereby neglecting the potential role of intermittently dry streambeds in biogeochemical dynamics. However, recent studies have shown substantial influence of dry streambeds on C fluxes within fluvial environments. This study aims to understand the effects of hydrological alterations on stream-atmosphere C exchange, with specific emphasis on the relative contribution of dry and wet areas. During an exceptionally dry period of over a year, we monitored the extent of wet and dry areas and measured carbon dioxide (CO<sub>2</sub>) and methane (CH<sub>4</sub>) fluxes along a 100-m reach of a Mediterranean intermittent stream using high-frequency automatic sensors and in-situ samplings every 2-3 weeks. Dry streambeds showed high areal fluxes of C, especially for CO<sub>2</sub>. Over the study period these fluxes contributed ca. 50% of the total reach-scale stream-air C exchange. These results emphasize the importance of accounting for both inundated and intermittently dry areas when analysing carbon exchange dynamics in intermittent streams. This issue becomes particularly relevant in the current context of climate change, with widespread water scarcity in many of the world's regions.

# Effects of Nanoplastic Sizes and Types on Leaf Litter Decomposition Processes in Freshwaters

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Nanoplastics (NPs) have emerged as a pressing global concern, yet their impact on freshwater ecosystems, particularly in streams, remains inadequately understood. In headwater streams, leaf litter decomposition, governed by aquatic fungi, notably aquatic hyphomycetes, is a fundamental process driving nutrient and energy transfer to various trophic levels. Though these fungi play a crucial role, the impact of NPs on litter decomposition in freshwater environments is still poorly understood.

In this study, the effects of environmentally relevant concentrations (up to 25 µg/L) of polystyrene NPs with varying sizes (100 and 1000 nm) and types (bare NPs and functionalised –COOH NPs) on leaf litter decomposition, community structure and sporulation rates of aquatic hyphomycetes were evaluated. Our results demonstrate significant impacts of both NP sizes and types on leaf litter decomposition and fungal sporulation, with smaller-sized and functionalised NPs exerting stronger effects compared to larger-sized and bare NPs. Additionally, the community structure of aquatic hyphomycetes, as assessed by abundance, was distinctively influenced by both NP sizes and types.

This study underscores the importance of further investigating the implications of NP pollution and their interactions on critical ecological processes in freshwater ecosystems and emphasises the need for informed management strategies to mitigate its potential adverse effects.

# Nanoplastics Regulate Aquatic Fungal Responses to Metals

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Plastic pollution, including nanoplastics (NPs), and metal contamination are major environmental concerns in freshwater ecosystems. Aquatic fungi, specifically aquatic hyphomycetes, play a crucial role as primary decomposers in these ecosystems. Therefore, understanding the interactions and impacts of these pollutants on key organisms like aquatic fungi is vital for freshwater ecosystem functioning. This study investigated the impact of environmentally relevant concentrations (up to 25 mg L<sup>-1</sup>) of bare and functionalised (–COOH) polystyrene (PS) NPs, along with copper (Cu; up to 50 mg L<sup>-1</sup>), on *Articulospora tetracladia* (ARTE). The worldwide distributed ARTE is a dominant aquatic hyphomycete responsible for litter decomposition in streams. Results show that both bare and –COOH NPs caused intracellular accumulation of reactive oxygen species (ROS) and disruption of the fungi's plasma membrane. Notably, simultaneous exposure of functionalised NPs with Cu induced a pronounced adverse effect, characterised by heightened cellular response and inhibition of fungal growth. This effect could be attributed to the enhanced adsorption of Cu onto the surface of functionalised NPs, as indicated by spectroscopic analyses. Conversely, exposure to Cu alone uncovered stimulatory effects on fungal growth. Overall, these findings underscore the complex interplay among NPs, metals and organisms in freshwater ecosystems. Further investigation, specifically at environmentally relevant concentrations, is crucial for informing environmental agencies and safeguarding freshwater biodiversity and health.

# Nitrogen dynamics under natural salinity in semiarid fluvial ecosystems

Yolanda García de Fuentes<sup>1</sup>, Rosa Gómez<sup>1</sup>, Eliot Mompean<sup>1</sup>, Anna Roman<sup>2</sup>, Joan Ferriol<sup>2</sup>,  
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Nitrogen (N) is a key nutrient for ecosystems; however, it is also one of the main driver of aquatic eutrophication. Water salinity is described as a stress factor affecting the structure and functioning of rivers and streams, and there are evidence of a potential effect altering nitrogen dynamics.

By studying natural semiarid saline streams, we examined the influence of salinity on the concentrations and dynamics of the different inorganic-N forms, and specifically on the relative importance of N-NO<sub>3</sub><sup>-</sup> and N-NH<sub>4</sub><sup>+</sup> species. Our findings from field survey and laboratory microcosms, which included a N spike (both in form of N-NH<sub>4</sub><sup>+</sup> and N-NO<sub>3</sub><sup>-</sup>) as well as the monitoring of N dynamics over 48 hours, confirm that not only N-NH<sub>4</sub><sup>+</sup> concentrations but also their dominance over N-NO<sub>3</sub><sup>-</sup> tend to be higher in streams with higher salinity. These results are in line with previous findings that, although scarce, suggest that salinity promotes those processes that increase N-NH<sub>4</sub><sup>+</sup> concentration in streams over those that reduce it. We found evidence that suggest limited nitrification, increased cation exchange from sediments, or increased N leaching from senescent plant material and riparian soils, as possible responses to salinity increases. Overall, results suggest high N-NH<sub>4</sub><sup>+</sup> concentration may be exacerbated by salinity increases, which in turn can affect aquatic organisms and the ecosystem function.

Because salinity is expected to increase in fluvial ecosystems worldwide as consequence of both the climate change and anthropogenic activities, the binomial water salinity-nitrogen requires much more future attention.

# Response of ecosystem-level stream metabolism to light

**Maite Arroita**<sup>1</sup>, Charles B. Yackulic<sup>2</sup>, Robert O. Hall<sup>3</sup>, Alison P. Appling<sup>2</sup>, Bridget Deemer<sup>2</sup>, Bryce A. Mihalevich<sup>2,4</sup>, Arturo Elosegi<sup>1</sup>

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The relationship between light and gross primary production (GPP) is well understood for individual plants in controlled settings, but less well understood for ecosystems composed of individuals of multiple species exposed to heterogeneous light environments. Most studies of river GPP assume that instantaneous GPP is either a linear function of light, or a nonlinear function based on studies of individual plants. However, many factors affect this relationship, including shading, light attenuation, and nonuniform distribution of primary producers. These misspecifications can cause large errors in GPP estimates. Here we modified a widely used river metabolism model and developed a new approach that accounts for process error (PE) during the day (versus generic PE during day and night) to allow a flexible means of addressing light saturation in the face of heterogeneous communities of primary producers exposed to heterogeneous light conditions. We compared three models: linear GPP-light relationship and generic PE; saturating GPP-light relationship and generic PE; and daytime PE. We compared their performance by simulating  $[O_2]$  data under different light scenarios and evaluating each model's success in recovering the simulation parameter values. We also fit the three models to  $[O_2]$  data measured at four case studies. Overall, the daytime PE outperformed the other models examined when light was misspecified, suggesting that even the generic PE term can be misspecified, leading to errors in metabolism estimates. Additionally, the daytime PE provided insights into ecosystem-level light saturation and hysteresis, providing a basis to improve our understanding of how light controls ecosystem-level stream metabolism.



# ECOSYSTEM SIZE IS RELATED WITH A GRADIENT IN PHOSPHORUS AVAILABILITY IN HIGH MOUNTAIN LAKES AND PONDS

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The limnological features of high mountain ponds and shallow lakes remain less understood compared to larger lakes. Typically, phosphorus availability is lower than nitrogen availability in freshwater environments, yet in highly productive environments, nitrogen may become the limiting factor for phytoplankton growth. This study deepens into the limnological attributes of 74 lakes and 134 ponds in the Pyrenees, specifically focusing on nitrogen and phosphorus availability and their impact on phytoplankton composition. The size of the ecosystem was related with a gradient of phosphorus availability in relation to nitrogen, influenced by factors such as vegetation coverage along the shore and water temperature. Major ions were not significantly related with lakes size, but alkalinity, silicon, and calcium varied with catchment lithology. Additionally, Cl<sup>-</sup> and K<sup>+</sup> levels were higher in ponds than in lakes, likely due to the effect of livestock. Lakes exhibited low concentrations of total phosphorus (TP) and total nitrogen (TN), characteristic of ultra-oligotrophic systems. Conversely, ponds displayed a wider concentration range for both TP and TN, indicative of a trophic gradient from oligotrophic to eutrophic conditions. Organic nitrogen was more abundant in ponds, while lakes predominantly featured inorganic nitrogen. Based on TN/TP and DIN/TP ratios, water bodies likely limited by phosphorus exhibited significantly lower concentrations of Chl-a compared to those limited by nitrogen. Chlorophyte and cryptophyte biomass were significantly lower in phosphorus-limited systems compared to nitrogen-limited ones among the different algal groups. Other significant variables explaining algal group composition included lake temperature and the dominance of minnows.

# Impact of multiple stress factors on the metabolic balance of Mediterranean mountain lake

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Global climate change has caused new combinations and levels of intensity of multiple stressors in ecosystems, such as temperature, nutrients, UVR, and CO<sub>2</sub> concentration. The metabolic balance and algal-bacterial interactions of aquatic ecosystems are complex processes and are regulated by these factors. The individual effect on ecosystem functioning of these stressors has been studied previously, however, there was a knowledge gap regarding how a simultaneous increase in these factors will impact ecosystems. This study aimed to quantify the interactive effects of these four abiotic controllers to measure the potential impact effects of global change conditions predicted for the year 2100. Temperature and nutrients were classified as the dominant drivers. For that reason, their impacts were evaluated in both isolated conditions and, also in conditions where current and future UVR and CO<sub>2</sub> environmental conditions were combined. A collapsed design was utilized including natural planktonic communities from a mountain lake to evaluate the effects of these factors. Based on our results, the interactive effect of these four drivers enforces the C-flux through the microbial loop, favors autotrophic processes, and increases the carbon sink in high mountain lakes. These findings stress how important it is to comprehend the entire perspective of the complex environmental scenarios we are expecting in the near future. This study serves as a great foundation to construct more models that can help predict such impacts on more complex microbial diversity dynamics.

# Multiple stream ecosystem processes responses to anthropic and natural drivers at regional scale

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Stream ecosystems are dynamic and highly interactive with their surroundings, making them particularly vulnerable to anthropogenic impacts that affect their structure and functioning. Ecosystem functioning is based on biological processes that are affected by different environmental variables. Therefore, their response to anthropogenic impacts complex. Our study aimed at identifying the key drivers of key ecosystem processes (nutrient uptake, organic matter decomposition, biomass accumulation and ecosystem metabolism) at a regional scale. We measured these processes across 63 stream reaches in Gipuzkoa and related them to 38 natural and anthropogenic variables. The results show the complex direct and indirect relationships between environmental drivers and ecosystem processes. Generally, nutrient uptake rates were mainly controlled by nutrient availability, which was significantly enhanced by artificial land-use. Artificial land-use also affected the rest of the studied processes, mainly through its effects on temperature and turbidity. However, these processes were also influenced by the area of the drainage basin and their lithology. Metabolism, more specifically GPP, was negatively related to forest land-use. Our study provides empirical evidence of the decline of multiple functions associated with the impacts resulting from human activities in the basin. Overall, our findings emphasize the significance of investigating and monitoring stream ecosystems processes on a broad spatial scale to comprehensively understand their dynamics and control mechanisms.

# Leaf-litter decomposition in headwater streams depends on litter traits and digestive enzyme profile of key macroinvertebrates

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Headwater streams are characterized by their dependence of allochthonous organic matter, introduced into these ecosystems from riparian vegetation, which represent their main source of energy and nutrients. The composition of these riparian plant communities is modified due to warming, and consequently, the quality of litter inputs to the stream ecosystem. We investigated the influence of leaf quality on the decomposition process mediated by shredder macroinvertebrates. A reciprocal incubation of high quality leaf litter of *Alnus glutinosa* (mountain streams) and low-quality leaf litter of *Phragmites australis* (lowland semiarid regions), was carried out in six headwater streams. Moreover, we investigated digestive enzyme profile of shredders for determining which dietary components are most effectively metabolized. We measured the activity of digestive enzymes in dominant shredder taxa from streams. In general, higher decomposition rates were found for high-quality litter, and were higher in the warmer lowland region, whereas litter decomposition corrected for the effect of temperature was higher in mountain streams, where shredders dominate. Density and biomass of shredders in leaf-litter bags were also investigated. *Echinogammarus obtusidens* and *Protonemura* sp. have the most balanced enzymatic profiles, whereas trypsin activity was dominant in *Tipula lea* and both cellulolytic activities in *Melanopsis praemorsa*. The results indicate that, in semiarid lowland streams, organisms from other functional groups, i.e., scrapers, perform this same function, becoming key species in the fluvial food web. Identifying these species that play a key role in essential processes for the functioning of the ecosystem is fundamental for its conservation and protection.

# Hydromorphological changes and vegetation succession after flood events in Cantabrian rivers

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Extreme flood events are a major driver of riparian vegetation succession and river morphology configuration. Increasing our understanding of the interplay between flow, sediment and vegetation to shape fluvial habitats is paramount to build scenarios of river habitat change linked to restoration or to explore global change effects. In this context, rivers from the Cantabrian Cordillera have experienced significant flooding over the past decade, resulting in visible channel shifts and vegetation cover regression, followed by ecological succession and recovery in some circumstances. In this study, we aimed to semi-automatically quantify these changes throughout the 2016-2023 period, using aerial photographs and Sentinel-2 images processed in Google Earth Engine. We considered a linear bidirectional sequence that includes five classes: deep and shallow water, unvegetated sediment bars, and sparse and dense vegetation. Class shifts along this sequence were quantified by comparing random forest classifications of summer satellite images (10-m resolution) in four rivers. As a result, we were able to map sediment sinks (accretion zones) and sources (erosion zones), which can also be described in terms of ecological succession and regression. Our results illustrate the effects of year-to-year flow pattern changes, locating them in time and space. Furthermore, we could distinguish between mature and young riparian vegetation patches. The developed methodology may constitute a straightforward cloud-based tool to assess hydromorphological and ecological changes in dynamic river systems, paving the way to future research on channel-vegetation interactions, and the role of flood magnitude, river channel form and riparian vegetation in shaping these processes.

# Effects of snow avalanches on alpine lakes: water column changes, zooplankton resurrection and sediment hiatus

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When snow avalanches reach alpine lakes with enough energy, they produce an impulse wave that affects the entire water body. We present a detailed description of the effects of this impulse wave on an alpine lake. The avalanche broke the ice cover (around 50 cm thick) and caused the lake to overflow with a wave of at least several hundred cubic meters. The impulse wave altered the water column stratification and physicochemical properties (dissolved oxygen, conductivity) in the short (hours) and mid-term (days and weeks). It also caused the removal of a large amount of sediment in the deepest part of the lake (330 m<sup>3</sup> in 2025 m<sup>2</sup>), and its decompaction and deposition in shallower areas (1035 m<sup>3</sup> in 4615 m<sup>2</sup>). These changes in the sediment increased the sedimentation rate in the lake and caused important modifications in the zooplankton density and composition in the next four years after the avalanche, including the resurrection of a cladoceran species (*Daphnia pulicaria*) that had disappeared from the lake decades ago, thanks to the re-exposure of its resistance eggs on the sediment surface. The paleolimnological implications of these events may be dramatic since we have now recorded the formation of a present-day sedimentary hiatus (loss of 75 cm of sediment), and we have documented the past almost complete removal of sediment from the deepest part of the lake around 260 BC.

# Dark Carbon Uptake in Emerged Sediments: Seasonal and Daily CO<sub>2</sub> Flux Patterns in a saline wetland

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Inland water bodies are currently shrinking due to alterations of the water cycle, exposing extensive areas of previously submerged sediments to the atmosphere. Consequently, the carbon cycle is altered, leading to an increased emission of CO<sub>2</sub>. To study this, we performed monthly surveys throughout one year to measure CO<sub>2</sub> fluxes from dawn to dusk in the calcite-rich emerged sediments of the endorheic saline wetland of La Laguna de Fuente de Piedra, Málaga (Spain). Consistent with existing literature, CO<sub>2</sub> emission fluxes predominated in these sediments. However, at dawn and dusk fluxes were considerably lower, and even CO<sub>2</sub> uptake into the sediment occurred.

Consequently, we conducted three campaigns over a full 24-hour cycle to measure CO<sub>2</sub> fluxes in the emerged sediments and their most influential environmental drivers. This approach revealed a net CO<sub>2</sub> emission from the sediment during daylight hours, while a net CO<sub>2</sub> uptake by the sediment was measured during darkness. The magnitude of CO<sub>2</sub> fluxes exhibited seasonality with both highest uptake and emission fluxes in summer, reaching a maximum emission and uptake value of 0.29 g(CO<sub>2</sub>)m<sup>-2</sup>h<sup>-1</sup> and -0.14 g(CO<sub>2</sub>)m<sup>-2</sup>h<sup>-1</sup> respectively. In contrast, winter showed the lowest fluxes, with a maximum emission value of 0.04 g(CO<sub>2</sub>)m<sup>-2</sup>h<sup>-1</sup> and a maximum uptake value of -0.01 g(CO<sub>2</sub>)m<sup>-2</sup>h<sup>-1</sup>.

Our results reveal that, in addition to seasonal variability of CO<sub>2</sub> fluxes magnitude in Fuente de Piedra, a daily trend in CO<sub>2</sub> fluxes exists. This trend shows that CO<sub>2</sub> emissions from sediments increase towards midday but decrease towards midnight resulting in CO<sub>2</sub> uptake by the sediment.

# Impacts of Eucalyptus Plantations on Macroinvertebrate Communities in Galician Streams

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In this study we compare the structure of macroinvertebrate communities in streams that differ in the amount of Eucalyptus leaves accumulated in the stream bed. Our goal was to assess if increased amounts of Eucalyptus leaves affect macroinvertebrates and community composition. To address this question, we sampled 20 sites in Galicia, NW Spain in autumn and spring of 2020-2021, and classified as plantation, mixed and reference (=autochthonous forest).

Compared to reference sites, streams running through Eucalyptus plantations exhibited higher water conductivity, temperature and were more shaded during autumn. In spring, streams running through Eucalypt plantations had lower pH and higher CPOM. However, there were no differences in macroinvertebrate densities between plantations and reference site neither in autumn (ANOSIM,  $p=0.32$ ) nor in spring (ANOSIM,  $p=0.599$ ). Functional feeding groups did not differ significantly between vegetation types in either season. Instead, the abundance shredders in streams flowing through Eucalyptus was lower.

The effects of plantations were limited, perhaps due to the seasonal variations in leaf litter inputs and water flow, which may have masked the effects of Eucalyptus litter on macroinvertebrate communities. The reduced diversity of shredders in streams running through Eucalyptus plantations is likely due to the low quality of Eucalyptus leaf litter, which may contain feeding deterrents and have lower nutrient content compared to native deciduous species.



# Layered in Time: Exploring the Impact of Nearly 8 Months of Stratification on Carbon Dynamics in a Mediterranean Reservoir

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Ecosystem metabolism determines its role as a CO<sub>2</sub> source or sink. In lakes and reservoirs, metabolism is conditioned by the phytoplankton community. This relationship was studied throughout a hydrological year in El Gergal, a Mediterranean mesotrophic and warm monomictic reservoir used for water supply to the city of Seville, Spain. Aerobic metabolic rates (respiration and primary production) were modeled using high-resolution data, including temperature, PAR, oxygen, salinity, and meteorological variables. The size structure of the phytoplankton community was examined using flow cytometry. Thermal stratification persisted for 7.7 months. During this period the aerobic ecosystem metabolism was autotrophic, with a net O<sub>2</sub> production of  $0.06 \pm 0.03 \text{ mg l}^{-1} \text{ day}^{-1}$ , and ultraphytoplankton (an intermediate size group between pico- and nanophytoplankton) dominated the community. Notably, the epilimnion was the only layer exhibiting net O<sub>2</sub> production, while the metalimnion remained almost neutral and the hypolimnion was heterotrophic. During the mixing period, El Gergal was heterotrophic (net O<sub>2</sub> consumption:  $0.12 \pm 0.03 \text{ mg l}^{-1} \text{ day}^{-1}$ ) with picophytoplankton (the smallest size group) dominating the community. On an annual scale, El Gergal reservoir was slightly heterotrophic, raising concerns about its role as a CO<sub>2</sub> sink.

## Saltpan as carbon sink

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Part of the Marchamalo salt pans (Murcia, Spain) has been abandoned for 28 years. The project “RESALAR” aims to restore 8 ha of this saltpan to extract salt in a traditional way while preserving biodiversity and historical heritage. The principal primary producers in hypersaline wetlands are microorganisms such as cyanobacteria and chlorophytes. Once the microorganism’s biomass becomes part of the decomposing sediment organic matter, the high sediment salinity and moisture levels leads to an anoxic environment, allowing it to be buried in the sediments. In this context, our study, previous to the restoration, aims to assess the carbon stock accumulated during the period of disuse as well as measure the carbon dioxide (CO<sub>2</sub>) efflux from the saltpan to the atmosphere and the effect of various physicochemical variables on soil CO<sub>2</sub> emissions. To determine the carbon stock in the first 0.5 m we extracted a core from every tank, analyzed the bulk density, organic carbon, inorganic carbon and salinity in 5 cm sections. CO<sub>2</sub> fluxes were measured using a soil respiration chamber connected to an infrared gas analyzer, and samples were taken from the first 10 cm of soil to analyze physicochemical variables. Our preliminary results shows that mean carbon stock value was  $1349.71 \pm 251.39$  Mg ha<sup>-1</sup> and mean CO<sub>2</sub> flux was  $0.55 \pm 0.84$  g m<sup>-2</sup> h<sup>-1</sup>. Temperature was the main CO<sub>2</sub> flux driver, while sediment moisture was the main carbon burial driver. Our results will be used to suggest management strategies to make the salt extraction compatible with carbon burial.

# OS3

## Community Ecology



# Succession of zooplankton in a Mediterranean pond using taxonomic and functional approaches

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Habitat restoration offers a unique opportunity to recover aquatic biodiversity and study several ecological processes in new habitats, such as successional changes in communities. In restored habitats, an increase in species richness over time and a decrease in beta-diversity between consecutive samplings are expected, and several ecological mechanisms can drive these temporal variations. An approach using functional traits can reveal how communities are assembled through time. In l'Albufera Natural Park (Valencia, Spain), a permanent interdunal pond has been created in a restoration project, and zooplankton samples were bimonthly taken over the first four years after pond creation (2007 – 2011). A set of taxonomic and functional indices were estimated, including Hill numbers, functional richness, originality and redundancy, and taxonomic and functional beta diversity, and their trends over time were analyzed, as well as the relationship with environmental variables. The results showed an increasing trend in taxonomic diversity and functional richness along the four-year succession. Regarding beta diversity between samplings, a decreasing trend was observed for taxonomic and functional values. A minor role of local environmental conditions in driving both community and traits assembly was found. Contrarily, the positive correlation between taxonomic and functional richness showed that niche differentiation was the primary mechanism in community assembly. In addition, regional factors, such as colonization through passive dispersal, were probably more important than local ones.

# Analysis of the planktonic food web structure of three lakes in Lagunas de Ruidera Natural Park (Central Spain)

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This study is part of the DAMOLAKE project, which aims to address the anaerobic oxidation of methane coupled with denitrification in lakes by means of two differentiated but complementary disciplines: one focused on biogeochemical processes and the other on the responses of the food web. From this second approach, we studied, in winter and summer 2022, the main constituents of the planktonic food web and their isotopic signature of three lakes (Conceja, Colgada and Cueva Morenilla), in Lagunas de Ruidera Natural Park (Castilla-La Mancha). The planktonic community consisted of 38 phytoplankton and 22 zooplankton taxa. Phytoplankton community presented low densities; being Cyanophyta and Chlorophyta, the most diverse taxa. Zooplankton appeared dominated by the rotifers *Keratella cochlearis* and *K. quadrata*, the cladoceran *Bosmina longirostris* and the copepod *Tropocyclops prasinus*. In Conceja and Colgada, zooplankton biomass was dominated by juvenile copepods while in Cueva Morenilla, cladocerans dominated, although all showed low densities. The taxa were grouped into nodes, based on their morpho-functional features, creating interaction networks. Differences in the structure and function of the networks between winter and summer periods, as well as among the three lakes, were observed, highlighting biological dynamics influenced by environmental factors and available resources. However, global network properties (i.e. connectance, modularity, and nestedness), remained relatively stable both among periods and lakes. This underscores the importance of functional diversity and adaptability of the studied trophic networks. The trophic network approach is crucial for understanding the functioning of these ecosystems as well as to protect the biodiversity they host.

## Size distribution shifts in fluvial networks of different level of anthropization

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Anthropogenic stressors, such as land-use changes, heavily impact aquatic ecosystems as they alter biodiversity and community properties in multiple ways. The body size structure of aquatic communities for instance, can respond to nutrient inputs, to pollutants and to changes of the quality and quantity of resources, which can more strongly affect individuals of a given body size, and thus, affect the energy flow along the food web. In this work we assess changes in the size spectra of aquatic communities (i.e. relationship between body size and density of organisms) in fluvial networks mildly impacted by alterations in land use change (tree plantations and urban areas). We expected the moderate increase of nutrient pollution in the most urbanized areas to increase the amount of energy flowing from the base to the top of the food web, and thus, to increase the abundance of big individuals. To test this hypothesis, we sampled fish and macroinvertebrate communities in several sampling locations distributed along 5 fluvial networks with different level of naturalness in their catchments and obtained their body mass to construct size spectra for each sampling site. Preliminary analyses point that size spectrum was very conservative in highly preserved headwater catchments. On the other hand, anthropization of the land use seemed to be related to more energy being channeled to the top of the food web, as shown by the presence of the largest fish in urban areas.

# Analyzing changes in functional traits structure and complexity in spatial and temporal gradients

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Analyses of species traits provide information at the functional level (i.e. on the role that species play within the ecosystem) thereby complementing taxonomic analyses in community ecology, ecosystem management and conservation studies. Most studies so far have focused on the analysis of local trait diversity and the replacement of traits along natural and human impact gradients. However, changes in trait composition might also reflect species interactions and spatiotemporal dynamics. In this regard, the analysis of traits using a network perspective allows us to infer the interactions of functional networks and its complexity. In this study, we investigated the temporal and spatial variation of functional networks in aquatic ecosystems focusing on macroinvertebrates communities. We used trait data from 56 lakes and 36 rivers in Sweden, collected over 20 and 11 years, respectively. We designed an iterative procedure merging samples through space (samples from different sites taken the same year), and through time (samples from the same site but sampled in different years). After merging the selected samples, we calculated their functional structure and obtained several functional and network metrics (e.g. functional richness, centrality). We illustrate how our framework can successfully capture changes in functional complexity at both temporal and spatial scales. This can help to better understand how ecosystems will respond to global change by providing insights into key ecological questions such as the relationship between biodiversity and ecosystem functioning or the spatiotemporal stability of ecological networks.

## Drivers of distance-decay of similarity in headwater macroinvertebrate communities

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Headwater streams make a great contribution to the macroinvertebrate diversity at the catchment scale, as they show high variation of the communities among headwaters ( $\beta$ -diversity). They are usually well preserved and only suffer from mild anthropic perturbations; thus, communities vary responding to more subtle environmental changes and the spatial connectivity between them.

Distance-decay of similarity is a common approach when analysing  $\beta$ -diversity, which assumes that taxonomic or functional (i.e. traits) differences between pairs of biological communities is positively related to the geographical or environmental distance between them. Although, patterns of decay rates are known at large spatial scales, they are highly scale dependant so, they presumably will differ in freshwater communities at the regional scale.

We sampled benthic macroinvertebrate communities in 50 reaches from five headwater stream networks, located in protected areas in the Basque Country and Navarre. In each site, we assessed drivers that can act as environmental filters for macroinvertebrates (water properties, habitat quality and land use) and barriers that limit dispersal among the reaches. We analysed the taxonomic and functional similarity of macroinvertebrate communities among reaches and tested whether the decay rates responded more to geographical or environmental distances. We expect environmental distance-decay to be stronger than geographic distance-decay, as some sites suffer from mild perturbations, while others show nearly natural environmental conditions. We foresee this approach to be a good framework for identifying both, high value and problematic locations, by using the mean difference of every point involving each site from the distance-decay curve as proxy.



# Unravelling Trophic Networks Using Isotopic Signatures and Graph Analysis

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Understanding the intricate trophic dynamics within communities is crucial for effective ecosystem management and conservation. This study employs a novel integration of isotopic analysis ( $\delta^{13}\text{C}$  &  $\delta^{15}\text{N}$ ) and graph tools to unlock the structural organization of trophic interactions within food webs. To do so, we have sampled 45 aquatic communities (from microcrustaceans to amphibians and fishes, if present). These communities were located at 4 different ponds (10 to 12 ponds of different sizes were sampled in each pondscape). Taxa were sorted by feeding type and body size for isotopic analysis. Subsequently, we constructed undirected, weighted graphs for each pond to delineate food web networks, and estimated modularity and topological roles within the web.

The number of nodes in the analyzed communities ranged from 4 to 27. Modularity ranged from 0.069 to 0.286, with 2 to 4 modules identified per community. The main roles identified among nodes (proxy of taxa), corresponded to the peripheral (i.e. specialists) and connectors (i.e. which are nodes connecting modules) types, and only in a few webs, ultra peripheral nodes were detected (i.e. super specialists).

Our findings open a new approach to analyze the hierarchical arrangement of species interactions, highlighting central hubs and peripheral nodes within the web. This will be invaluable for gaining a deeper understanding of food web functioning. Indeed, our approach elucidates the functional roles of taxa, which ultimately defines their contributions to ecosystem dynamics.

# Impact of environmental stress on macrofaunal communities in Mediterranean ponds

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The increase in human activities in recent years has generated significant environmental stress in aquatic ecosystems, which are experiencing progressive and increasing deterioration. To evaluate this impact on aquatic macrofauna communities in Mediterranean ponds, two aspects have been measured: pond's integrity and water trophic state. Pond's integrity was measured by using the ECELS index, which provides information on the wetland condition status considering aspects such as pond morphology, human pressure (e.g., frequency of visitors, agricultural activities, or the presence of nearby infrastructures), water appearance and vegetation composition and structure. To analyze the trophic state, several indexes were used (TRIX and TSI) based on the data of nutrients, dissolved oxygen, chlorophyll-a and total phosphorous concentration. We selected 30 ponds situated in five locations in Catalonia and classified them considering both aspects: habitat condition and trophic state. Then, the richness and the diversity of the macrofaunal communities present in each pond was analyzed to test the effect of anthropic stress on aquatic diversity in Mediterranean ponds. Our findings emphasized the significance of managing and reducing the impact of human activities on aquatic ecosystems to preserve their biodiversity.

# Functional diversity of macroinvertebrate communities under the influence of environmental filters: salinity and altitude

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Environmental filters are habitat features that determine the possibility of species to persist in a local community. Only those species adapted to certain environmental filters will be present in the local community. Therefore, strong environmental filters could imply the exclusion of species and, consequently, a functional trait sorting in the local community. Thus, understanding how strong environmental filters affect functional traits is crucial to comprehend functional diversity patterns at metacommunity level. We collected macroinvertebrate samples from 57 temporary ponds distributed across five pondscape in the Mediterranean region. Two of the pondscape are subject to harsh environmental conditions: one is situated at an altitude of approximately 2100 m (Eastern Pyrenees), and the other is a brackish water pondscape located at sea level on the coast (Empordà Wetlands Natural Park). The three remaining locations do not experience similar strong environmental conditions, as they are freshwater ponds located in lowlands. Therefore, they will be considered as reference pondscape to compare the variations in functional diversity with and without the influence of the mentioned environmental filters. We quantified functional diversity indices using a functional-trait based approach. We investigated whether communities occupying pondscape with stronger environmental filters exhibit higher levels of functional redundancy, and whether this results in a decrease in the functional diversity of macroinvertebrate communities.

**OS4**

**Monitoring, Management,  
and Restoration of  
Aquatic Ecosystems**



# Monitoring the biological quality of an urban stream using a learning-by-doing approach with higher education students

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This study assessed the longitudinal and temporal trends in the biological quality of the stream Ribeira de Covões (Coimbra, Portugal) using a learning-by-doing approach with higher education students under supervision. From 2004 to 2014, benthic macroinvertebrates were collected nine times in five sampling campaigns to calculate the scores of the biotic index IBMWP and of the multi-metric Portuguese index IPtIS. The two indices provided similar information, but the scores were more constrained for IPtIS than for IBMWP. The biological quality was moderate along most of the longitudinal profile and time. Scores decreased downstream attaining a poor condition at the two lower reaches. The overall quality of the stream increased temporally from 2004 to 2013, decreasing in 2014 due to the disruption of the benthic macroinvertebrate communities by strong flash floods. This learning-by-doing approach provided useful information on the spatial and temporal trends in the biological quality of the stream and detected invasion by the New Zealand mud snail. Data collected by higher education students may fill data gaps and complement information gathered by national authorities.

# Proposed solutions for disruption caused by hydroelectric production in the Cabriel valley

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The work is situated in the Conquense area of the ‘Valle del Cabriel’ Biosphere Reserve, where a sustainable management model for heritage and natural resources is promoted. With this approach in mind, the hydroelectric production scheme has been thoroughly analysed to propose a correction that aligns the production system with a more natural flow regime.

Electricity production systems are one of the installations that can significantly impact the operation of these systems. The work scheme comprises the following steps:

- Identifying the hidroelectricity plants under study
- Classifying the power station to detect the fluvial environment alteration.
- Design studying of the plant (equipment and operation) and its production method.
- Analyzing the hydrological alteration produced by the plant and other environmental effects.
- Reviewing the e-flow proposals for the water bodies affected by the hydroelectric production of these plants

The focus of the work was on the Contreras and Lucas Urquijo plants. The Lucas Urquijo plant has a complex installation that takes water from one river and turbines it in another. The review aimed to correct the alteration of the flow regime by applying hydraulic simulation models. The correction includes the maintenance of a minimum flow in the intake sections and the reduction of hydropeaking in the release sections. The new flow release proposals are designed to preserve natural habitats and key hydraulic processes.

# Laboratory tests of a HAB bioremediation method based in allelopathy

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Harmful Algal Blooms (HABs) of the toxic cyanobacteria *Microcystis aeruginosa* are the most damaging to the human economy and recreation in freshwater environments worldwide. This species is favored by prolonged periods of stable water column, high temperatures, high radiation levels and eutrophication. Consequently, climate change and human-induced eutrophication have led to a worldwide increase in the geographic distribution, abundance and intensity of these HABs. Although many remediation methods, including mechanical, chemical and biological methods, have been tried, there is none that is effective, widely applicable, affordable and does not affect other ecosystem components. The aim of this work was to test a bioremediation method for *M. aeruginosa* HABs based on the allelochemical portoamide, produced by the filamentous cyanobacteria *Phormidium* sp. This method showed strong effectiveness in long-term monocultures of this toxic cyanobacterium. However, with natural phytoplankton communities in the long-term, while the allelochemical significantly reduced the total biomass of the phytoplankton, *M. aeruginosa* indirectly benefited from the decay of the more sensitive species. These results underline that complex community dynamics are a key element to consider, and the success of the method depends in optimizing the intensity and dosage of the allelochemical.

# Decarbonizing Hydrological Landscapes Through Dam Removal

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Reservoirs are globally recognized as a significant anthropogenic source of greenhouse gases (GHG) to the atmosphere, particularly methane. However, the significance of employing dam decommissioning as a strategy to remove these emission hotspots in the river network has been largely overlooked. Here, we assess how dam removal reduces GHGs emissions by comparing CO<sub>2</sub> and CH<sub>4</sub> emissions before, during, and after the demolition of a 7m tall dam located in a Mediterranean stream in Spain. CO<sub>2</sub> and CH<sub>4</sub> flux measurements were carried out during one year in stream reaches upstream and downstream the reservoir, in the reservoir itself, and in a negative control in a reservoir within the same catchment. Our findings reveal variations during the dam removal process: CH<sub>4</sub> and CO<sub>2</sub> emissions were higher before the decommission in water, although the CO<sub>2</sub> released by sediments exposed to the atmosphere was considerably high during decommission. Furthermore, we observed a pronounced seasonal effect on GHG emissions, for both CO<sub>2</sub> and CH<sub>4</sub> fluxes, being higher in summer compared to winter across all three study sites. Emissions from the new dry areas and stream generated after dam demolition were more similar to those from upstream and downstream river reaches than emissions from the control reservoir. Overall, our results support the hypothesis that dam removal imply a reduction in the carbon emissions from the river network. In a context of global change, we anticipate that our findings will constitute an additional incentive for future dam removal projects as they reduce anthropogenic GHG emissions.



# Restoration proposal of a Mediterranean eutrophic temporary wetland by adding magnetic particles: a laboratory approach

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Mediterranean temporary wetlands face threats from climate change and land use changes, leading to eutrophication. This study explores the feasibility of using magnetic particles to reduce phosphorus and emerging contaminants in water column. In this context, the objective of this work is to evaluate whether the subsequent addition and removal of magnetic particles in dry sediment reduces the later (after reflooding) internal phosphorus load to the system. Sediment cores were collected (along transects extending from the wetter zone to a distance of 72 m) during July 2023 at Laguna de los Juncas which discharge in the RAMSAR wetland Fuente de Piedra (Málaga, Spain). Once in the laboratory, the incubation experiment was run by using a resuspension chamber. For each sampling station, two different treatments (five replicates each) were applied: (i) control, no addition and (ii) magnetic particles addition. After a contact time of 24 h, magnetic particles were removed by applying a magnetic field gradient and later, lake water was added to simulate the effect of reflooding conditions. Water samples (for nutrients and emerging contaminants analysis) from each core were taken at different times. Results indicate a significant reduction in phosphate fluxes when magnetic particles were added. Additionally, significant reductions have been found in some of the emerging contaminants after the addition of magnetic particles. Overall, our results suggest that magnetic particles can be considered as a promising technique for improving water quality in temporary Mediterranean wetlands not only by reducing internal phosphate load but also emerging contaminants after reflooding.

# New metabarcoding-based protocol for Saprolegniales

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In the last few years, the number of studies using metabarcoding to assess diversity in ecosystems has increased. The quality of this assessment depends on the taxonomical precision barcoding markers can achieve. Ideally, metabarcoding should rely on species level discrimination to achieve maximum accuracy, which implies using fast evolving marker genes.

Saprolegniales (Stramenopiles; Peronosporomycetes) are fungi-like protists and include about 20 genera and more than 100 species. Some of these organisms are well-known as pathogens for aquatic animals and plants threatening wildlife, aquaculture and crops. Most studies developed on these organisms are focused on particular species and rely on strain isolation. However, barcoding-based studies provide quickly and accurately information on the distribution patterns of the entire diversity and allow predicting zoonotic episodes. Indeed, reaching species-level accuracy is especially important to assess the organism's pathogenicity, but also their ecological contributions due to their association to organic matter and their role in freshwater ecosystems.

In that purpose, we developed a metabarcoding protocol specific to Saprolegniales, ensuring fast, cost effective and reliable estimation of environmental diversity. We designed two primers that amplified specifically the 5.8S gene and the variable region ITS2 of Saprolegniales. We tested their efficiency on 18 pure cultures, 10 water-filtered samples and 8 sediment samples. We extracted DNA, amplified it specifically and sequenced each sample using Illumina Next Seq. We provided an evaluation of Saprolegnialean diversity in human impacted (aquaculture basins) and pristine sites. We propose this new protocol to be used to track emerging diseases and infection dynamics.

# A biophysical simulation of cyanobacteria migration in water bodies

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In order to face the increasingly precarious state of water bodies due to the appearance of Cyanobacterial Harmful Algae Blooms (CyanoHABs) and to mitigate their effects, new efficient solutions for monitoring and preventing their growth are being developed. A key challenge in these efforts is to obtain reliable information about the studied water bodies, since the data collected must provide an accurate and comprehensive picture of the water system's current state. Traditionally, this information is gathered from specific points of the water body with fixed probes, or in-situ direct human-collected measurements and sampling. However, these methods do not provide enough data to accurately assess the present and future state of the aquatic systems. In this presentation, we introduce a simulation model that can provide information about the migratory movements of CyanoHAB clouds, as well as about their growth or death within the studied water body. The main aim of this model is to support the determination of the optimal locations for gathering measurements and samples. The system factors in real biological behavior such as the vertical migration of CyanoHAB, through density changes dependent on the depth, and the daylight cycle, all of which evolve in real-life geographic scenarios.

This system has been developed within the DEVS-BLOOM framework, which aims to create a virtual twin of the selected waterbody taking into account real-life data to facilitate inference and preventive actions according to the expected evolution of the water body's state.

# Evaluation of an Artificial Aeration Strategy for the Rehabilitation of an Urban Eutrophic Lake in Uruguay

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Urban eutrophic lakes face severe nutrient accumulation, leading to cyanobacteria blooms, low transparency, anoxia, and fish kills. Rehabilitation strategies like artificial aeration aim to improve conditions, but many attempts fail due to insufficient understanding of the lake processes involved. We evaluated the first hypolimnetic aeration system installed in a deep urban eutrophic lake in Uruguay. The experience was a community-driven initiative with local government support, and the scope of this study was focused solely on the limnological assessment aspect.

Monitoring was conducted from September 2020 to February 2024 to assess the effects of the aeration intervention on thermal regime, nutrient dynamics, water transparency and other key limnological variables.

The lake exhibited warm monomictic behavior, stratifying in summer with persistent hypolimnetic anoxia, and mixing in winter, seemingly unaffected by the intervention. Interestingly, water transparency significantly increased in mid-August 2023, around 2 years and 8 months after aeration commenced, coinciding with decreased chlorophyll-a concentration. However, by late February 2024, transparency declined again. Although a trend toward decreasing hypolimnetic phosphorus was observed during stratification periods, the system remained eutrophic overall.

Results so far indicate the aeration system yielded limited water quality improvements, potentially due to being undersized or operational issues. Although to fully assess the experience would be relevant to consider costs, due to the nature of the intervention, access to relevant data was not yet possible. Complementary research approaches are underway, including satellite monitoring of long-term chlorophyll-a dynamics and evaluating social perceptions to contextualize the rehabilitation strategy from another perspective.

# Boosting river citizen sciences: new approaches to identify macroinvertebrates: locomotion, behaviour, and artificial intelligence.

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Citizen science (CS) projects such as RiuNet and ProjecteRius provide valuable contributions to monitoring river ecosystems. However, a discrepancy has been observed between the biological quality of rivers assessed by CS and official data, with a general underestimation.

This can be attributed to several factors: (1) reduced sampling effort, as time constraints of citizen participants may limit the frequency and intensity of sampling, (2) technical limitations, such as the lack of professional equipment, which may hinder access to certain habitats and comprehensive collection of the invertebrate fauna, and (3) identification difficulties either because naked-eye observation can make it difficult to accurately identify small or less mobile invertebrate families, or because of potential confusion between similar families. To mitigate these discrepancies, new solutions are proposed for one of the key steps in the process: how to help identify macroinvertebrate families.

1. Creation of a new behavior-based identification key by using citizen participants' live observation. A new identification key is proposed that focuses on the movements and behavior of macroinvertebrates in controlled containers. This will allow an innovative classification of bioindicator taxa, independent of the macroinvertebrate' anatomical characteristics, which will facilitate the task for citizen participants.

2. Use of automatic image identification by supplementing the identification key with artificial intelligence algorithms and models for automatic image identification of macroinvertebrates. This will allow for rapid and accurate validation of identifications made by citizen participants.

The implementation of these proposals can contribute to more accurate and reliable data in CS projects, increasing their value for river management and conservation.

# Artificial intelligence as a tool for cyanobacterial bloom forecasting using multiparametric probes and phycocyanin fluorescence measurements

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Cyanobacterial blooms (CBs) are expected to increase due to anthropogenic activities, threatening ecosystem functioning and water quality. Recent efforts have focused on developing early warning systems to enable timely implementation of management measures, mitigating the impacts of incipient CBs. This study aims to assess the feasibility of high-frequency data and artificial intelligence to provide an effective early warning of CBs. To achieve this goal, the research utilized five years of water column data collected by a TriOS multiparametric probe in Cuerda del Pozo reservoir (Soria, Spain). This reservoir, that offers multiple ecosystem services, has also a well-described history of CBs. First, the data was pre-processed and explored towards the creation of effective time series that represented the CB problem in the reservoir. Second, five forecasting models were trained and evaluated, including simple exponential smoothing, Random Forest, and Long Short-Term (LSTM) neural networks in their autoregressive and multivariate versions. Phycocyanin fluorescence served as the cyanobacterial proxy, with chlorophyll a and temperature as additional covariates in multivariate models. The models were evaluated for seven forecasting horizons ranging from 4 to 28 days. Classic evaluation metrics were used in a hybrid approach, assessing the capabilities of the models to forecast either the phycocyanin concentration or the offset of an alert system based on phycocyanin thresholds. The multivariate LSTM neural network provided outstanding forecasts, especially at horizons beyond 16 days. Results will be discussed in relation to their applicability for the advanced monitoring and management of waterbodies affected by CBs.

# Major drivers controlling phosphorus adsorption on magnetic particles: the case of treated wastewater discharging in a Ramsar Mediterranean wetland

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Nowadays, a wide variety of phosphorus (P) adsorbents with different physico-chemical characteristics are commercially available for combating eutrophication. Among them, magnetic particles have been recently proposed for P removal from freshwater bodies as P-loaded adsorbents can be recovered by applying a magnetic separation gradient. Apart from the physico-chemical characteristics of the adsorbent (e.g. particle size, specific surface area), environmental conditions (e.g. pH and competitive ions) play an important role in the P removal efficiency. In this sense, it is expected that these chemical interferences will be especially strong in matrices as complex as treated wastewater. In this context, our objective was to determine the major drivers (e.g. silicate, emerging contaminants, major cations/anions) affecting P removal efficiency when using magnetic particles in treated wastewaters that discharges into a Ramsar wetland (Fuente de Piedra, Málaga, Spain). To get this purpose, laboratory experiments were conducted under batch conditions by using samples from two different stations: station A (high polluted) and B (less polluted). Our results have shown P removal efficiencies up to 99% and 90% in stations A and B, respectively. Additionally, strong temporal fluctuations in P removal efficiency were observed, so linear models were used to relate P removal with several physico-chemical variables (e.g. pH, major cations and anions, emerging contaminants). A relevant temporal variability has been evidenced in the efficiency of P removal associated with changes in the chemical composition of the water, which demonstrates the complexity of the interactions involved and the need to delve deeper into future research.

# Conservation planning for present and future management in drying river networks

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Drying River Networks (DRNs) are globally distributed freshwater ecosystems that provide key biodiversity values and vital ecological processes. Climate change will increase drying occurrence and intensity worldwide, threatening the conservation values of these DRNs. Despite their importance, DRNs have received little conservation attention, and efficient integrated strategies are needed to ensure good management practices and biodiversity conservation. Systematic Conservation Planning emerges as a useful tool for integrative management at the DRN scale, offering the potential to define priority areas within the river network based on values such as biodiversity, ecological functions, and ecosystem services. Here, we present a planning exercise for six European DRNs located in countries with different biogeographic, climatic, and socioeconomic characteristics. We first created a survey to be answered by experts of different DRNs to gather information that would allow us to define and prioritize conservation objectives and determine the most significant criteria to include in the planning process. Then, we developed two-time scenarios accounting for current interannual variation and future conditions, to prioritize 1) sites within the river network that are resilient and could act as refugia for species, and 2) identify river sections that have the potential to maintain biodiversity if Nature Base Solutions (NBSs) were implemented. Results obtained here can be used to assist decision-making and allocate management efforts to areas within the DRN where they are most needed. These approaches are essential for developing successful strategies and implementing actions to mitigate threats and adapt DRNs to the changing conditions they will face.



# Environmental DNA-based bioindication of *Arcellinida testate amoebae*

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Bioindication, the evaluation biological responses to environmental perturbations, is crucial for assessing environmental health. Traditionally, bioindication in freshwater ecosystems relied on macroscopic organisms such as arthropods. Nowadays, the technological leap forward brought by massive sequencing and applied to environmental DNA (eDNA) allows using protists as bioindicators without the need of morphological identification. We propose here a novel eDNA-based approach using *Arcellinida*, a group of top predators among protists, to monitor freshwater ecosystems quality. In that purpose, we firstly apply a specific eDNA protocol to evaluate their diversity and the connectivity between sites. Then, we characterized the *Arcellinida* diversity over one year at three different points of the Lake Sanabria, an ancient glacier lake known to be subjected to anthropogenic disturbances, and compared this diversity with an undisturbed control site. Results indicate limited connectivity between the different ecosystems, and an edge effect in the flood zone in the shore of Lake Sanabria. Disturbed freshwater ecosystems exhibited reduced *Arcellinida* diversity at both specific and infraspecific levels, providing valuable insights into recent perturbations. *Arcellinida*-based bioindication provides a sensitive, accurate and easy to interpret protocol for monitoring disturbances in freshwater ecosystems. It represents a valuable tool for environmental assessments and conservation strategies.

# Magnitude and variability of air-water carbon fluxes in two newly created Mediterranean coastal lagoons

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Coastal ecosystems play a significant role in biogeochemical dynamics, yet large uncertainties remain on how these systems affect the global carbon cycle. We measured monthly fluxes of carbon dioxide (CO<sub>2</sub>) and methane (CH<sub>4</sub>), in two coastal lagoons built in autumn of 2017. Measurements were conducted from February 2023 to February 2024. Our aim was to quantify the carbon gas fluxes and to examine key factors controlling variation of these fluxes in both lagoons. Although the lagoons are only 100 m apart, they highly differ from each other in physical, chemical and biological characteristics mainly due to their level of confinement and exchange with the sea during storms. The fluxes of CO<sub>2</sub> and CH<sub>4</sub> were markedly different between the two sites, being higher in the most confined lagoon, characterized by low conductivity and high macrophyte coverage, than in the most connected lagoon, characterized by high conductivity and phytoplankton dominance. Both lagoons emitted carbon to the atmosphere for nearly the entire study period. The maximum flux rates for both CO<sub>2</sub> and CH<sub>4</sub> occurred in September, when water temperature was highest. In contrast CO<sub>2</sub> uptake period were different. Macrophyte-dominated lagoon absorbed CO<sub>2</sub> during spring, whereas the phytoplankton-dominated lagoon absorbed CO<sub>2</sub> during winter. Our study sheds light on the carbon dynamics in shallow newly created coastal lagoons and the relevant role of hydrology in the carbon budget of these ecosystems.

## Comparison efi+ integrated/efi + modulated Ibi-Júcar

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Ecological Status (“ES”) is defined as “an expression of the quality of the structure and functioning of the aquatic ecosystems associated with surface waters”. The establishment of this quality involves biological indicators, among which is the ichthyofauna, the assessment of which is controversial as various indices have been tested with different results; one of the main problems detected with the fish indices is the relationship with the pressures and the level of confidence of the results.

For the years 2019-2020-2022-2023, the Júcar River Basin District compares how the use of the Integrated EFI+ (methodology included in the Guide for the Assessment of the State of Surface and Groundwater published by MITERD; applied only in those bodies where EFI+ data are available) and EFI+modulated by IBI-Júcar (Aparicio et al. 2011), where the IBI-Júcar acts as a modulator of the Confidence Level of the result, affect the assessment of the “ES”.

The results obtained in the ES by applying both methodologies for calculating the ichthyofauna indicator have been cross-checked with the significant pressures affecting fish life.

It is observed that the use of Integrated EFI+ penalises the ES assessment more, however, it allows higher levels of confidence to be obtained, nevertheless, the results obtained with the EFI+ indicator modulated by IBI-Júcar correlates better with the significant pressures affecting the ichthyofauna

Both the results of the assessment and the NCF of the results obtained will allow a better choice of indices and thus more reliable results in the assessment of the ecological status.

# Citizen Science, a valuable tool for monitoring presence and quantity of plastic residues in fluvial ecosystems. Two years of implementation of “Plastic Pirates - Go Europe!” project in Spain

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Plastic pollution is an environmental challenge of our time with a substantial knowledge gap regarding the real sources, fluxes and impacts of plastics in freshwater ecosystems. There is a lack of standardized protocols to monitor their presence and abundance and they are not included in protocols for monitoring environmental quality in freshwater ecosystems by environmental authorities. Citizen Science can serve as a tool to bridge this gap by obtaining data over large spatial and temporal scales. When young people participate as part of school and young organizations, Citizen Science directly engages them with these environmental problems at a local level, provides them an understanding of the scientific process and promotes positive habits to face this problem. This is the aim of the EU-funded project “Plastic Pirates - Go Europe!”, where young people of 12 European countries participate in sampling campaigns and data collection about plastic pollution in freshwater ecosystems. This project uses standardized methods to quantify and characterize plastic and microplastic litter in rivers and to identify possible plastic sources. The collected data is then analyzed by scientists. In Spain this project is implemented since 2022 in schools and young organizations from Castilla y León and Cataluña. Over 800 students from 38 schools and organizations are sampling 37 sections of 10 different rivers. This information is useful in identifying the most impacted sites in the studied rivers, the types of plastic residues contributing most to it and the possible sources or habits responsible for this pollution.

# Reducing nutrient and emerging contaminant inputs via semi-natural ponds system in a Mediterranean RAMSAR wetland

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Fuente de Piedra is a RAMSAR wetland (Málaga, Spain) received treated wastewater directly from two local wastewater treatment plants (FdP I and FdP II) until the late 1990s. In 2005 the wastewater's flow from FdP I was redirected to a system of channels and semi-natural ponds to reduce the inputs of contaminants into the wetland. This study aims to assess the efficacy of the semi-natural ponds system in Fuente de Piedra on improving the water quality reaching the wetland. Assessment parameters included nutrients [total nitrogen (TN), total phosphorus (TP), chlorophyll a (Chla)], and emerging contaminants (ECs). Water samples were collected from four sampling points within the semi-natural ponds system (stations A-D) to evaluate its purifying capacity, with an additional sampling point (station E) within the main water body (Fuente de Piedra Lake) to quantify external nutrient, evidenced by reductions in TN by 69.2%, TP by 35.6%, and Chla by 84.1%. Analysis of ECs unveiled the presence of 34 contaminants, pharmaceuticals (13), pesticides (11), drugs/stimulant substances (9), and personal care products (1). Notably, prominent ECs including caffeine, benzoylecgonine, naproxen, imidacloprid and BP4 exhibited reduction percentages exceeding 75% from station A to station D. These findings underscore the efficacy of the semi-natural ponds system improving the water quality and reducing external nutrient and ECs loads reaching the wetland. Nonetheless, nutrients and Chla concentrations indicate that the wetland remains hyper-eutrophic, highlighting the need for innovative management strategies to further enhance wetland water quality.

# The decline of a protected coastal wetland: Albuferas de Adra (Almería, Spain)

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Albuferas de Adra is the most important coastal wetland in south-eastern Spain. Its ecological importance as a permanent coastal wetland in a semiarid region made it worthy of being declared as Natural Reserve in 1989. Albuferas de Adra is also included in the list of Protected Areas of the Ramsar Convention, and constitutes a ZEPA, ZEC and LIC site. Despite all this environmental protection, the wetland is subject to several direct pressures derived from the occupation of its watershed by intensive agriculture. As a result, its lagoons have experienced a dramatic reduction in water quality, reflected in an exponential increase in salinity and in trophic level (hypereutrophic). Consequently, the ecosystem biodiversity is significantly threatened. Among other impacts, common reed beds are being progressively replaced by bare patches and halophilic species, the protected fish *Aphanius iberus* has disappeared in the wetland in the last 5 years, the presence of the frog *Hyla meridionalis* has experienced a drastic reduction, and the population decline of six waterfowl species (*Podiceps cristatus*, *Anas platyrhynchos*, *Gallinula chloropus*, *Aythya fuligula*, *Tachibaptus ruficollis* and *Oxyura leucocephala*) has been recorded, with a particularly significant decline in *Oxyura leucocephala* during the breeding season. The community of marsh birds is also strongly simplified as a consequence of habitat degradation. All these impacts are extremely threatening the ecological integrity and medium-term viability of the wetland, and it is imperative the immediate development of effective protection and conservation measures, which must include the transformation of the agricultural use of the basin.

## Flood-driven riverside litter accumulation: identification of critical areas within river basins

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The escalation of litter accumulation in aquatic environments is recognized as an emerging global concern. Although rivers represent the main conduits for land-based waste into the oceans, the spatial dynamics of litter accumulation in these systems remain poorly investigated, especially after hydro-climatic extreme events. Floods have been identified as major drivers of litter mobilization, including macroplastics, within rivers. However, predicting flood-induced litter accumulation along riverbanks is complex due to the cumulative interplay of multiple environmental and anthropogenic factors. Using empirical data collected from 14 stream reaches in two Northern Atlantic rivers, in Portugal, we evaluated which factors, among geomorphological, riparian and anthropogenic descriptors, best explain riverside litter accumulation after floods. Specifically, we addressed two major questions: i) how do the accumulation and characteristics (type, size) of riverside litter vary across a rural-urban continuum after floods?; and ii) what are the main factors influencing the accumulation and deposition of riverside litter? We found that the combination of human population density and the stream slope at river reach showed the highest explanatory power for the accumulation of riverside litter. Our findings indicate that litter tends to be retained close to the source, even under flood conditions. This work highlights the importance of gathering field data to identify critical areas of riverside litter accumulation within river basins. Our findings can further support environmental managers in designing and implementing effective cleanup campaigns and implementing plastic recovery strategies at specific areas.

## Functional and structural indicators of riparian restoration success: three case studies at the Vitoria-Gasteiz Green Belt

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Riparian restoration (RR) aims to return river ecosystems to their original ecological conditions. However, many RR actions focus on recovering river hydromorphological characteristics only, with little effect on their communities or their functioning. It is thus unusual to consider the recovery of the ecological integrity and its capacity to preserve biodiversity and provide ecosystem services. In the RIBER project, we establish a framework that seeks to alleviate the common deficiencies of RR projects: i) the aim of RR must be the reestablishment of river ecosystem integrity; ii) evaluation of RR success should consider multiple level indicators; and iii) it must taken into account the ecosystem resilience. Considering this framework, we studied three streams flowing from the mountains that surround Vitoria-Gasteiz to the urban center. The three case studies presented common stream gradients, with all three of them having restored reaches within the urban Green Belt. These three case studies allowed us to evaluate RR success using multi-level indicators. We thus examined several communities (benthic algae, microbial decomposers, macroinvertebrate detritivores and the riparian forest) and ecosystem processes (biofilm accrual and organic matter decomposition) at three sites per stream: a pristine headwater site, a downstream site altered by land-use and not restored, and a downstream site where riparian vegetation had been restored. Most of the assessed indicators pointed to a slight recovery of ecosystem integrity in restored sites, suggesting that RR projects are useful but higher efforts are needed in order to truly recover river communities and ecosystems.



# A new habitat model for river macroinvertebrate communities

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River habitat models often overlook macroinvertebrate communities, their functionality, and ecological needs, primarily considering single species as ecological targets.

In this research, we aimed to develop a new habitat model for the whole macroinvertebrate community, based on the MesoHABSIM method. We studied three rivers located in Northern Italy characterized by braiding morphology, gravel riverbeds, and summer low flow. We combined the recently developed Flow-T index with a Random Forest (RF) regression to apply the index at the mesohabitat scale. By integrating field data and 2D hydrodynamic models, we calibrated the model in the Trebbia River (2019 campaign) and validated the model in the Trebbia, Taro, and Enza rivers (2020 campaign).

The RF model identified the most important habitat descriptors for describing the community habitat. The 12 selected descriptors belong to frequency classes of water depth, flow velocity, substrate, and longitudinal connectivity to the main channel. The model calibration yielded an R<sup>2</sup>CV (cross-validation) coefficient of 0.71, whereas the validation on the test dataset provided an R<sup>2</sup>CV<sub>test</sub> coefficient of 0.63.

The results show good agreement between the simulated and the observed data, ensuring sufficient accuracy and reliability of the model. The model has potentials to predict the ecological response of macroinvertebrate communities to flow regime alterations and river morphological changes. In addition, our results extend the MesoHABSIM methodology, widely used for fish habitat assessment, to a different target community. This extension opens possibilities for applying our approach to design ecological flows in both perennial and non-perennial rivers, including those without fish fauna.

## Poor taxonomy, unsuccessful data interpretation.

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The identification of Chironomidae is difficult and time consuming because egg, larval, pupal and imaginal forms may be identified. As a result, most of the studies consider Chironomidae as a single taxonomic unit. Therefore, a lot of information is lost when only family level are considered. In a study made in the Pesquera River (Prat et al 2021), the use of subfamily level or the color and size of the larvae gives more information to understand better the effects of pollution. In a recent study (Fritz et al, 2023) comparing indicator species in permanent (P) vs intermittent(I) streams in 2 Ecoregions of USA, found that when using family level 11 (P) and 12 (I) families are indicators of streamflow condition, but Chironomidae do not appear to be indicative of streamflow. When using genera level, 7 of 23 genera in Permanent rivers and 10 of 21 genera in Intermittent ones are Chironomidae. As most studies of macroinvertebrates use only EPT and OCH taxa at genera level data interpretation may be incorrect.

# Defining macrophyte communities in the national reference network of Spanish rivers

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The application of European Water Framework Directive (WFD), and more specifically the ecological status assessment of rivers and streams, implies the need of define the reference communities that correspond totally or near totally to undisturbed environmental conditions. The composition and abundance of aquatic flora is one of the biological quality elements to be monitored for the classification of ecological status of rivers. Since the year 2019, macrophytes, which include algae, bryophyte and vascular plants are being monitored along with other quality elements to stablish the reference conditions of Spanish rivers. A national network of sites without significant anthropic pressures were defined based on stressors' thresholds. The reference river network used in this study included a total of 315 sites covering the whole Iberian Peninsula and 32 national river types. The study period covered the years 2019 to 2022, with a survey per year. The main objective was to define river type-specific macrophyte communities under reference conditions. Of the total taxa collected (determined at genus and species level), 11% were cyanobacteria, 26% eucaryotic algae, 40% bryophyte and 23% vascular plants. Multivariate analyses were performed to test significant differences in community structure between river types and to define indicator species for them. Multivariate classification of river types based on macrophyte communities showed weak correspondence with the present river typology, suggesting the merging of some river types. Finally, we discuss some aspects about the use of macrophytes as bioindicators to evaluate the ecological quality of river ecosystems.

# The Albufeira Project: Joint program for the assessment of water bodies in the Spanish-Portuguese Hydrographic Basins

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The project “Joint program for the assessment of water bodies in the Spanish-Portuguese Hydrographic Basins” (acronym ALBUFEIRA) was developed between 2018 and 2022 within the scope of the Operational Program INTERREGV (POCTEP). One of its main objectives was to advance in the harmonization of methodologies for the assessment of the ecological status/potential of water bodies shared by Spain and Portugal. For this purpose, 44 water bodies were studied: 32 rivers, 7 reservoirs and 5 transitional (estuarine) bodies. The ecological status/potential of each of them was determined by applying the methodologies of each country. The results obtained in 70% of the water bodies studied were discordant. In rivers, discrepancies were due to the location of sampling points, the assignment of types and the definition of reference conditions, the use of different and non-intercalibrated indicators of benthic invertebrates and ichthyofauna, discrepancies in the taxonomic identification of diatoms, and the use of different hydromorphological indicators. In reservoirs, the main discrepancies were due to differences in reference conditions and the establishment of class boundaries, the number of samplings and the use of different metrics and indicators (physico-chemical and biological). Finally, in estuaries, discrepancies were due to differences in the indicators used (physico-chemical and biological), in the sampling procedures and in the reference conditions and adjustment class limits defined for physico-chemical parameters and some biological indicators. The implementation of the project has allowed to improve the knowledge of the Spanish-Portuguese water bodies and to define proposals for harmonization of methodologies to mitigate the discrepancies.

# Designing a novel biological water quality index: implementing metabarcoding and machine learning techniques using Arcellinida as a model

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Anthropogenic pressures are exerting an increasingly significant impact on freshwater ecosystems, particularly in arid climates. Bioindication is the predominant method for monitoring environmental quality. Various biomonitoring protocols have been developed and incorporated into Spanish legislation, such as IBMWP, which relies on visual observation of macroinvertebrates. Nonetheless, these protocols suffer from complex procedures and a lack of taxonomic precision, resulting in limited sensitivity. Furthermore, the majority of these protocols have been designed specifically for lotic systems and are unsuitable for lentic water bodies.

Exploring novel biological indicators present in all lentic environments is imperative, enabling convenient sampling and providing sensitive outcomes that are easily interpretable. Additionally, metabarcoding-based protocols present appealing features including sensitivity, efficiency, cost-effectiveness, and reproducibility, while eliminating the reliance on taxonomic expertise.

In this project, we developed a biomonitoring protocol to screen Arcellinida diversity based on metabarcoding. Arcellinida are widely distributed in inland water bodies, display significant ecological niche specialization, and demonstrate swift reactions to environmental disruptions. These attributes make Arcellinida outstanding bioindicators for monitoring purposes.

The goal of this project is to set an innovative rapid and reliable metabarcoding-based protocol for monitoring of water quality in lentic systems. We applied this protocol across the three main river basins in Southern Spain to reveal the Arcellinida communities composition. Furthermore, we will utilize machine learning methodologies to formulate environmental quality indices, with the aim of incorporating this approach into Spanish legislation for assessing the environmental health of freshwater lentic ecosystems, which are presently confronted with substantial threats.

# Future generations and river management: unravelling children's mental models of the ecological challenges facing these ecosystems

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There is broad agreement in science education community on the importance of mental models for learning science. In the context of river ecosystems, students' construction of mental representations through science education activities can help them to understand the complexity of these ecosystems and also to predict how climate change and future anthropogenic impacts could affect them. This can lead to foster a sense of environmental stewardship, which lays the groundwork for informed decision-making and responsible behaviour towards these ecosystems. In this study, we aimed to understand how primary and secondary school children develop mental models regarding river ecosystems before and after an outdoor educational activity offered by a research group linked to the Museu del Ter, which consisted of analysing the biological quality of a river. To this end, a simple exercise in which children drew both how they imagined a unimpacted and impacted river was conducted before and after an educational activity, and then again after 15-20 days from this activity. They could also add some words to identify elements or processes that were difficult to draw or interpret. These depictions were analysed using a Qualitative Content Analysis methodology with the aim of identify the most important key elements in their drawings: water, inert substrates, riparian vegetation, fauna from near and inside water, organic matter, ecosystem services, anthropogenic impacts and climate change. Mental models therefore ranged from the simplest, in which a river is seen as just water, to the most complex, in which all key elements were included.

# Assessing trends in bad trophic state in the Albufera Lagoon (Valencia, Spain) from inherent Optical Properties of water

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The optical properties of water provide information about the quality of the aquatic environment. Transparency and Chlorophyll are fundamental variables in the assessment of trophic state. Albufera of Valencia is a shallow hypertrophic oligohaline mediterranean coastal lagoon of 21 km<sup>2</sup>. Over the last five decades, human activity has drastically altered this ecosystem, leading it into a turbid phase characterized by the disappearance of macrophyte meadows and the predominance of phytoplankton, because of agricultural and urban-industrial pollution. Evaluating optical properties in the Albufera Lagoon is an approach for monitoring its trophic state. Using field data collected between 2016 and 2023 and satellite imagery, semi-empirical algorithms were developed to estimate some variables of interest. The most effective algorithm for estimating total suspended solids, based on the red-edge and blue reflectances, had an nRMSE of 8.76%. In the case of transparency, using green and red-edge bands, it obtained a 17.8%, and for chlorophyll-a red and red-edge bands are best with a 14.67%. The importance of understanding the specific characteristics of the studied water bodies and the optical properties of their components in the development of algorithms for the estimation of water quality variables is highlighted. On the other hand, remote sensing, and the development of specific algorithms for estimating water quality variables in inland waters are valuable tools for monitoring. Also, the integration of remote sensing in environmental management can provide essential information for decision-making in water resources management

## Citizen science finds first evidence of microplastics in high mountain lakes of Sierra Nevada

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Microplastics pollution is one of the most concerning global threats to human and ecosystem health, and their presence has been documented in most of aquatic ecosystems worldwide, including remote mountain lakes. The citizen science campaign “74 High Mountain Glacial-Lake Oases,” aims to advance research and conservation efforts on high-mountain lakes of Sierra Nevada (Spain), considered sentinels of global change and unique ecological laboratories, and is enabling the collection of scientific data otherwise inaccessible to research teams. One of the most striking results of this campaign was the detection and quantification for first time of microplastics in 35 high-mountain lakes and the determination of the factors controlling their abundance. Microplastics were present in most of these lakes, with an abundance of >20 particles per liter, like that found in some of the most polluted lakes in the world. Microplastics were found for all size-fractions, but the abundance of particles of size <45 µm was higher. Although atmospheric transport is a main pathway of microplastics input to these ecosystems, their quantity was related to the presence of meadows surrounding the lakes. This finding suggests an anthropic origin caused by mountaineers who visit lakes with meadows much more frequently than rocky lakes. The high abundance of microplastics found in these lakes, which are headwaters of rivers that feed reservoirs for drinking and irrigation water, is a major concern that warrants future investigation to minimize the impacts of this growing type of contamination.



# Phytoplankton bloom formations in La Barca reservoir: causes and implications for ecological quality

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A global decline in water quality has been observed over the last decades. In reservoirs, ecosystem degradation has been mainly linked to eutrophication and climate change driven warming, known to favor phytoplankton blooms. Although these blooms are an inherently natural process of aquatic ecosystems, phytoplankton blooms are increasing in intensity and duration, especially during spring and summer periods. Here we examine phytoplankton blooms in La Barca reservoir (Asturias, Spain) between May and November 2023 at four stations including the tailing areas and in the main body of the reservoir. To characterize the causes and consequences of phytoplankton blooms, we performed physicochemical profiles of the water column along with taxonomic analysis and quantification of phytoplankton, enabling the calculation of the ecological potential index (IPE) of the reservoir, based on the law established in the RD 817/2015.

The general objectives of this work are to describe both the physicochemical characteristics of the reservoir and the spatio-temporal variability recorded in the phytoplankton community, and its possible implications on the ecological potential of the reservoir, given that this potential is focused exclusively on the biological quality element phytoplankton.

# Quantifying restoration success in Mediterranean streams by linking biodiversity, functionality and hydromorphological heterogeneity

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Restoration initiatives aimed at enhancing the hydromorphology of streams are on the rise globally but often fall short of achieving optimal ecological health. We contend that this shortfall arises from a lack of consideration for the spatial scales of stream hydromorphology crucial for biodiversity and ecosystem functionality. Moreover, conventional metrics relying on biological community composition may not fully capture the recovery trajectories of key ecosystem functions. Here, we present preliminary findings from Mediterranean sites within the RESTOLINK project (<https://restolink.weebly.com/>). We conducted comparisons of hydromorphological, biodiversity, and functional variables measured across multiple spatial scales among three types of stream reaches: (i) restored (formerly impacted by small dams that were removed), (ii) impacted (located upstream from small dams), and (iii) reference (maintaining well-preserved conditions). Generally, variables assessed in the restored reaches showed a tendency to deviate from those observed in impacted reaches and align more closely with those measured in reference reaches, suggesting a positive impact of restoration efforts. However, responses of biodiversity and ecosystem functions did not consistently follow the same trend post-restoration. Hydromorphology emerged as a significant factor driving differences observed among reaches. Overall, our findings underscore the importance of integrating hydromorphology, biodiversity, and ecosystem functioning for a comprehensive evaluation of stream restoration success.

**OS5**

**Exotic and Invasive Species**



## Fish invasions: live bait vs intentional introductions which is worse

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It is globally recognized that freshwater fishermen can have a major impact on promoting fish introductions, both through live bait disposal and the intentional introduction of target fish species. The aim of this study was to obtain a proxy of propagule pressure for fish invasions on the Iberian Peninsula associated with angling. We estimated the number of fish introduced per year, either as a disposal of live bait or as an intentional introduction. To achieve this goal, a bilingual survey was implemented online in Portugal and Spain and in person (direct inquiries) in two Iberian regions: the Lower Ebro (older fish introductions) in Spain and the Lower Tagus (recent fish introductions) in Portugal. Results showed lower introduction rates reported on the Ebro River than on the Tagus River. However, the popularity of non-native species, like European catfish, was higher in the Ebro. In both countries, the use of fish as life bait was of low prevalence (approx. 5%), yet it corresponds to large numbers of fish being introduced. Our conservative estimates revealed 273,600 events of bait discharge per year. Regarding the intentional introductions, we estimated a total of 140,000 intentional introduction events per year. These findings show that even a low prevalence of risk actions, corresponding to a small number of people, could have huge impacts. These results show that it is imperative to raise anglers awareness and maximize the supervision actions led by authorities

# Proposal for an index of alteration of the ecological state of river type water bodies due to the presence of invasive exotic species

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Although the effects of invasive alien species (IAS) on the aquatic ecosystems they colonize are well known, their presence is not considered when assigning the ecological (or potential) status of water masses required in European hydrological planning. This work presents the preliminary development of the Index of Alteration of the Ecological Status by Invasive Exotic Species (IAE4 I) and its application to 49 water masses of the Júcar Hydrographic Demarcation, with the presence of 11 different IAS. The IAE4 I is only applicable to “River” type water bodies. It’s calculated for each coded water body and each IAS considered. It’s composed of five blocks of variables related to the ecological and biological characteristics of each IAS, its ecological niche and real or potential effects, the hosting capacity of the receiving aquatic ecosystem and the interaction with other IAS present. In the selection of the variables, priority has been given to the availability of information and the ease of obtaining it, with the aim that the IAE4 can be applied extensively and relatively quickly and easily. The IAE4I varies between 10 and 140 and its result determines whether a change in the ecological status/potential of the body of water evaluated is justified. The results obtained indicate that the IAE4I is sufficiently precise and can be an appropriate tool to assess the incidence of IAS on the ecological status of “River” type water bodies. In the sample of water masses analyzed, 96% could require a downward change in their current ecological status.

# The impacts of non-native species on river bioassessment

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The number of non-native species/taxa (NN) continues to rise in rivers across the world. However, there is still insufficient evidence of whether existing bioassessment tools are sensitive to the effect of NN on the ecosystems. To fill that gap, we reviewed indices used in 17 countries from six continents and analyzed 8 databases and indices from 5 countries/4 continents. Fish have the highest number of NN reported, followed by macrophytes, invertebrates and algae. NN are associated with the general degradation of rivers (Canonical Correspondence Analyses). Native species for all biological groups negatively affected by NN richness and abundance. Yet, index scores are not consistently lower when NN are present and can even increase. Scores of indices accounting all NN were strongly negatively correlated (Spearman  $r=0.7$ ,  $p<0.001$ ) with NN richness and abundance; when NN are only partially considered, the correlations vary from strong to not significant. When indices do not account for NN, some were still weakly correlated with the presence of NN. In view of these results, we have 4 recommendations: Include specific metrics for NN in the indices based on all biological quality elements. Investigate NN species of algae and all invertebrate groups. Eliminate sites with NN from reference databases and remove NN from metric calculations (e.g. total richness) to avoid biased conclusions. Identify all taxa to species in biomonitoring programs to facilitate detection of NN species.

# Effects of *Acacia* invasion on water quality, litterfall, aquatic decomposers and leaf litter decomposition in streams

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Small streams and their riparian vegetation are closely linked ecosystems. Thus, the invasion of native riparian forests with non-native species can impact stream ecosystems. We assessed the effects of the invasion of broadleaf deciduous forests by evergreen, nitrogen (N)-fixing *Acacia* species on seasonal variation of water characteristics, litterfall in the riparian area, aquatic decomposers, and oak leaf litter decomposition, by comparing three streams flowing through native forests (“native streams”) and three streams flowing through invaded forests (“invaded streams”) in central Portugal. Fungal conidia concentration in water was higher in invaded streams in Spring/Summer when litter inputs, water temperature, and nutrients concentrations were higher. The fungal community structure differed and taxa richness was lower in invaded streams that were bordered by (almost) monospecific stands of *Acacia* trees. Macroinvertebrate density in leaf litter did not differ between stream types. Still, macroinvertebrate community structure differed and taxa richness was lower in invaded streams which had lower diversity of leaf litter. Finally, total decomposition rates of oak leaf litter were similar between stream types, despite the differences in decomposer communities. Overall, *Acacia* invasion changed water quality, litterfall seasonality and composition, and decomposer communities (especially fungi). In invaded streams, the effects on fungal communities seemed to be mostly mediated by changes in litter inputs rather than increases in water nutrients concentrations that remain in the oligotrophic range. The protection of non-invaded riparian galleries and restoration of the invaded ones could protect and restore stream ecosystems.

# A new European population of *Sinotaia quadrata* (Benson, 1842) (Gastropoda: Viviparidae) located at the Segura River basin (SE Spain)

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A new population of the Viviparid alien snail *Sinotaia quadrata* is reported for the Segura River basin (SE Iberian Peninsula), after the finding of specimens in two consecutive seasons in the river and adjacent irrigation canals. Attribution to the species was confirmed by the use of morphological and genetic characters. *Sinotaia quadrata* is a snail belonging to the Viviparidae family, native from East Asia (China, Taiwan, Korea), which has colonized multiple countries, including other Asian areas, but also Argentina in South America, North America, South Africa, and is also present in Europe. The population seems established, as juvenile specimens were found. In this paper, we report a new population of *Sinotaia quadrata* in Spain, and confirm its identity by molecular markers, showing also the same species identity with all the other European populations known to date.



# Invasive Species in Alqueva Multi-Purpose Project

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The richness of natural ecosystems lies in their native biodiversity and the way these elements have shaped that natural system over time. However, this balance is fragile and the entry of one or more new species, with invasive characteristics, can affect this balance between species and alter the native ecosystem, sometimes irreversibly.

In addition to the ecological impacts, its rapid colonizing capacity can also cause multiple negative effects, at a social and economic level, directly or indirectly, and can interfere with the main and secondary activities carried out, including the collapse of structures that depend on the affected resources. Repercussions on public health are also possible, as invasive species can carry unknown agents for local immunity.

In this context, EDIA, the manager of the Alqueva Project (EFMA), which main source of water – Alqueva dam – is located at Guadiana river, has been developing, for more than a decade, a set of actions aimed at understanding and controlling aquatic invasive species that pose the greatest risk to the EFMA, namely water hyacinth (*Eichhornia crassipes*) and the zebra mussel (*Dreissena polymorpha*).

This presentation will present EDIA's strategy for the management and control of invasive aquatic species and the work carried out, with emphasis on water hyacinth and zebra mussels.

The development of a medium and long-term strategy for the management of invasive alien species is a fundamental component in the context of biological invasions and the preservation of ecosystems.

# Growth of *Azolla filiculoides* in the lower-Tagus and Torrejón Tiétar sub-basin

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This study aims to understand the factors that promote the growth and spread of *Azolla filiculoides* in reservoirs within the lower Tagus basin, with the goal of predicting future massive growth and proposing suitable prevention and control measures.<sup>(1)</sup>

Initially, a bibliographic review of factors affecting its growth and main control measures was conducted. Subsequently, satellite images from 2016 to 2021 were analyzed to assess *Azolla* presence and coverage. Field studies were then conducted to collect macrophyte samples, accompanying macroinvertebrates, and assess relevant water physicochemical parameters (nutrients, coloration, chlorophyll-a). Additionally, degradation experiments were carried out to assess macrophyte (*Azolla* and *Lemna*) decomposition and changes in BOD5, COD, and water coloration.

Key findings indicate *Azolla*'s sexual growth and reproduction in tributaries under low flow conditions and high phosphorus concentrations. Subsequent dispersal of *Azolla* occurs from these tributaries to the reservoirs via sporophytes (which have accumulated phosphorus). Symbiosis with *Anabaena azollae* provides nitrogen to *Azolla*. Stable water levels along reservoir margins further favor *Azolla*'s growth.

Key factors promoting *Azolla*'s vegetative growth include ambient temperature, humidity, and light conditions, as well as low flow hydrological conditions, channel morphology, and the presence of coves, along with their orientation relative to dominant wind components.

Early detection is crucial for effective control, particularly during conditions conducive to expansion (dry winters), enabling barrier installation before *Azolla* reaches the main channel. Mechanical and manual methods have proven most effective for control, with selection dependent on accessibility and area coverage.

<sup>(1)</sup> Study financed by the river Tajo Water Authority.

# Hidden threat in exotic species: Potential disease-causing agents from freshwater turtles.

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Exotic species can spread new diseases and serve as vectors, hosts, or amplifiers for native diseases. Freshwater turtles have been used by humans for several purposes, as a result, they are widely traded, which explains their invasion success. Still, the impact of non-native turtles on diseases dynamics is poorly studied. We examined the disease-causing agents identified in non-native freshwater turtles to better assess the consequences of exotic freshwater turtles. After analyzing 55 articles, we found that there has been an increase in the number of published articles in the last decade, linked to an increase in the total number of publications of freshwater turtle disease-causing agents ( $p < 0.05$ ). When just non-native turtles were studied, Emydidae was the most recorded family, with *Trachemys scripta* as the most mentioned species. We found 250 disease-causing agents, more than half were Bacteria ( $n = 122$ ), followed by Animalia ( $n = 65$ ) and Virus ( $n = 30$ ), the remaining taxa accounted for 14%. Because 87% of the examined bacteria taxa were zoonotic, public health concerns may be connected to the large quantity of bacteria research. The limited number of research in virus highlights limitations in turtle virology. Only 17 studies also studied native turtles. These studies are critical to understanding the function of exotic turtles in disease dynamics. The most studied disease-causing agents were Animalia (47%) and Bacteria (42%), with Polystomatidae and Enterobacteriaceae being the most shared pathogen families between native and non-native species. This study provides a starting point for addressing public health and wildlife disease impacts caused by non-native turtles.

## Interactive effects of invasive species on stream functioning

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Freshwater ecosystems, particularly fluvial ecosystems, face severe threats worldwide due to a combination of natural and anthropogenic disturbances which have led to a rapid decline of biodiversity, outpacing the biodiversity loss rate of their terrestrial or marine counterparts. Invasive species, in particular, pose a widespread conservation problem and are considered one of the greatest threats to global biodiversity. Freshwater ecosystems are among the most invaded globally and are particularly vulnerable to the impacts of invasive species. These invaders can alter trophic networks by affecting resource availability when the invasive species is a primary producer (bottom-up effects) or engaging in direct herbivory and predation on native species when the invasive is a consumer (top-down effects). Besides, several invasive species are likely to coexist in an ecosystem. In that case, the combined ecological effects of multiple invaders may result in additive effects, or due to their potential interactions, synergistic or antagonistic effects. However, despite growing interest in the study of multiple stressors on ecosystems, there remains limited information on the ecosystem-level effects derived from the interaction of invasive species, particularly in systems controlled by omnivorous consumers. Here, by means of a field-based mesocosm experiment we aimed to evaluate the combined effects of two invasive species, a primary producer (giant reed, *Arundo donax*) and an omnivorous consumer (red crayfish, *Procambarus clarkii*), on the functioning of low order streams and the associated invertebrate community. Our results may contribute to the design and implementation of combined strategies for the control and management of these species.

# The biology and ecology of the invasive crayfish *Procambarus clarkii* in the Abegondo-Cecebre reservoir (Galicia, NW Spain)

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The American crayfish *Procambarus clarkii* (Girard, 1852) is one of the invasive alien freshwater species of greatest global concern. Although its biology and ecology have been extensively studied in natural water bodies and in certain artificial water bodies such as rice fields, there are a limited number of published studies in freshwater reservoirs. In this work we studied the spatial and seasonal use of the Abegondo-Cecebre reservoir by *P. clarkii*, as well as its potential role as biological reservoir for *Batrachochytrium dendrobatidis*, the amphibian chytrid fungus. Results indicate that most of the active burrows were concentrated near the river mouths entering into the reservoir, especially in the Mero river. Contrastingly, most of the captures of individuals occurred downstream, in the area close to the reservoir wall. Statistically significant differences in sex-ratios, age classes and cephalothorax length (independent of sex) were observed between the periods immediately before and after the winter quiescence phase. However, the presence of *B. dendrobatidis* was not detected in a total of 90 individuals of *P. clarkii* analysed. Current knowledge may be useful for implementing long-term effective and sustainable mitigation strategies in the area, as the reservoir provides drinking water to the large city of A Coruña, but it is included also in the Biosphere Reserve “Mariñas Coruñesas e Terras do Mandeo”, a Special Area of Conservation within the Natura 2000 network of EU protected areas.

**OS6**

**Biogeography and  
Evolution of Species**



# The role of palaeoclimatic variation on body size evolution of extant Odonata and Trichoptera (Insecta).

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Ecomorphological responses of organisms to palaeoclimatic changes are still poorly understood in ectothermic animals. In this study, we investigate patterns of body size evolution in different monophyletic groups of aquatic insects (i.e., dragonflies, damselflies, and caddisflies) as a function of relevant palaeoclimatic shifts in Earth's history. Specifically, we examine (1) Cope's rule (the tendency for body size to increase over evolutionary time) and (2) Bergmann's rule (the tendency to develop larger body sizes in cold climates) in two aquatic insect lineages with highly contrasting functional features and eco-evolutionary history. We hypothesized that body size evolution is explained in terms of both Cope's and Bergmann's rules. Our results suggest an impact of palaeoclimatic changes on body size evolution of extant aquatic insects, trends that have been far less studied in freshwater invertebrates compared to their marine and terrestrial counterparts.

# Historical biogeography, current distribution and phylogeography of frogs of the *Blommersia wittei* complex from Madagascar

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Madagascar is a biodiversity hotspot with an incredible diversity of amphibians. Here, we study a species complex formerly identified as *Blommersia wittei*. It includes *B. wittei*, two recently described species from the Comores (*B. transmarina* and *B. nataliae*), a recently described species from west Madagascar (*B. bara*), and one undescribed species (*B. sp. 12*). One mitochondrial and three nuclear loci recover *B. bara* + *B. sp. 12* sister to the Comoroan species + *B. wittei*. There is no evidence of mitochondrial introgression but allele sharing between *B. wittei* and *B. sp. 12*. *B. wittei* Sambava populations are genetically distinct, qualifying as a novel candidate species. Divergence time estimates suggest a Miocene diversification with recent demographic expansions in the last million years. Environmental niche models suggest that the distribution of *B. wittei* is smaller than currently thought, being restricted to the north of the island, while *B. bara* occurs across western-central Madagascar with a suitable climate in the north overlapping with the *B. wittei* predicted range. Paleodistribution hindcasts in the Last Glacial Maximum suggest range contraction of *B. wittei* to the north, and of *B. bara* in its southern edge. Those suggest a recent range expansion of *B. wittei* from the northwest to the south in agreement with the genetic data and demographic reconstruction. There is a clear contact zone between *B. bara* and *B. wittei* from Antsohihy to the Sambirano River, which could be wider. The three species from Madagascar meet in a small area at the Sambirano, which makes this an attractive system for studying contact zone formation and speciation in tropical areas.



# Bats, the cave fertilizers: a metabarcoding study on the case of Arcellinida (Amoebozoa: Tubulinea) in a karstic hydrosystem in Croatia

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The cave fauna has always intrigued scientists due to their high endemism rates and their remarkable adaptations to these particular ecosystems. While the drivers of diversity have been extensively studied in animals, only limited information is available on micro-organisms. Arcellinida (Amoebozoa: Tubulinea) is a group of amoebas characterized by their shelf-constructed test or shell, which are typically characterized as ecological narrow specialists with limited geographical distributions.

Here, we investigated the diversity and distribution patterns of Arcellinida within caves, with a specific emphasis on the influence of bat guano on communities. We sampled two hydrosystems encompassing both surface and subterranean waters located within the Dinaric karst in Croatia, a global hotspot of diversity. We used microscopical observation and a specific metabarcoding approach based on the mitochondrial cytochrome oxidase subunit I (COI) region to observe diversity patterns.

Abundance and biodiversity within caves were lower than outside, except in guano-rich sites which revealed an increased diversity, as well as many unique sequences not found elsewhere. These findings suggest the existence of guano-specific communities, which may possibly be endemic. They contribute to a better knowledge of subterranean biodiversity and its drivers, and underscore the importance of microhabitats in karstic environments diversity.

# Navigating the spatial and temporal aspects of beta diversity

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Beta diversity is often assessed at different spatial and temporal scales and, consequently, it has been defined in different ways. Here, we define four aspects of beta diversity that encompass the diverse applications of this concept in the literature: 1) spatial beta diversity, 2) temporal variation in spatial beta diversity, 3) temporal beta diversity, and 4) spatial variation in temporal beta diversity. Each of these aspects can be analyzed using presence/absence or abundance data, and can be characterized using taxonomic, phylogenetic, or functional community features. Our contribution helps standardize terminology and ecological interpretation, thereby minimizing ambiguities in the study and applications of beta diversity. Beta diversity is fundamental to biodiversity conservation and assessing the impacts of anthropogenic change on ecological assemblages. Therefore, a unified terminology for beta diversity will facilitate enhanced communication not only among researchers but also between researchers and biodiversity conservation practitioners.

# Anostracans (Crustacea, Branchiopoda) from North American lakes show unclear phylogenetic relationships between taxa and distinctive dispersal patterns

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Dispersal is essential for all organisms as it enables colonisation of new areas and survival of species. Numerous aquatic invertebrates disperse passively in a form of dormant fertilised eggs that are resistant to extreme weather and digestion systems of vertebrates. We first study genetic diversity of a *Branchinecta* fairy shrimp inhabiting lakes of the Saskatchewan Plain in Canada. Secondly, we aim to reconstruct the phylogeny and dispersal events in genetic distribution of this anostracan. Finally, we compare genetic diversity and distribution of *Branchinecta* and *Artemia franciscana* (= *Artemia monica*) in the Saskatchewan plain, the two ecologically very different anostracans. To this aim, we sequenced COI and 16S gene fragments of the collected *Branchinecta* individuals from Saskatchewan. We found a relatively high number of haplotypes for both markers (79 in COI and 81 in 16S) in *Branchinecta* from Saskatchewan. Our COI *Branchinecta* sequences showed intraspecific level of resemblance to *Branchinecta mackini* sequences from USA. Three genetic clades at the COI gene region were found in *Branchinecta*, different to *A. franciscana* where all available COI sequences from Saskatchewan belonged to one genetic clade. This suggests that *Branchinecta* sequences might be originating from several distinct Ice Age refugia, contrary to *A. franciscana* which might be originating from one area of refuge. Our list of *Branchinecta* populations in Saskatchewan is far from exhaustive as there are >100,000 lakes in this region. High density of suitable habitats and frequent dispersal events could explain high genetic diversity found in the studied *Branchinecta* anostracans.

# Crossing salinity barriers: metabarcoding of the family Cyphoderiidae to understand diversity patterns

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Salinity is, arguably, the most important barrier that divides diversity on Earth, especially for microbes. Indeed, protist communities differ drastically in composition and between marine and continental ecosystems. The diversity patterns that characterize their diversity result from the evolutionary history of each group, and notably how and when the salinity barrier has been crossed.

Metabarcoding allows the retrieval of massive amounts of diversity data from wide sampling designs. These data can be used to follow spatial diversity patterns and infer evolutionary histories. For these reasons, we have developed a specific protocol for metabarcoding a model group, for which we will follow inter-and intraspecific diversity patterns across the salinity barrier. We based this approach on a variable marker, the mitochondrial cytochrome oxidase (COI) gene, of the testate amoebae family Cyphoderiidae (Rhizaria; Euglyphida). This family is expected to be a good model group to evaluate the differences in diversity patterns since they inhabit both sides of the salinity gradient. This protocol should be as specific as possible to enable retrieving, ideally, all sequences of Cyphoderiidae present in the environment. In addition, environmental sequencing studies will show the widespread existence of members of this family, especially in marine systems, that have never been characterized morphologically or ecologically.

# The conquest of athalassohaline water bodies by freshwater lobose testate amoeba (Arcellinida, Amoebozoa)

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The salinity barrier is considered as one of the most important frontiers dividing biodiversity and influencing the distribution of organisms. Crossing it requires profound physiological adaptations and is therefore supposed to happen rarely in evolutionary histories but also opens the possibility for newcomer organisms to colonize new niches and diversify.

Most of these transitions have been studied between freshwater and marine environments. Here, we explored the biodiversity of Arcellinida, a group of protists that are common and diverse in freshwater sediments and soil but seem absent from the sea. However, we focused on athalassohaline water bodies. These non-marine saline lakes usually experience strong salinity fluctuations, sometimes reaching extreme values, but their island-like nature could mean lower biotic pressures (competition and predation) for newcomer organisms. We combined single-cell barcoding, culturing approaches and environmental metabarcoding to explore the biodiversity of Arcellinida in different water bodies of Spain and Chile, with salinities ranging from freshwater to values higher than seawater. An extensive phylogeny of the group revealed multiple (recent and more ancient) colonizations of the saline environment by Arcellinida from freshwater ecosystems. Some species were halotolerant, living in a wide range of salinities, including freshwater. In spite of the extreme conditions, Arcellinida diversity in athalassohaline water bodies is surprisingly high and remains largely unexplored.

**OS7**

**Ecology of Wetlands,  
Lakes, and Estuaries**



# Limnology of Mongolian Lakes

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Mongolia has around 10,000 lakes, 3,000 of them larger than 10 ha which constitutes an extraordinary lacustrine heritage. Although most of them are of less than 100 ha, Mongolia holds some of the largest lakes in Eurasia such as Khuvsgul nuur and those of the Great Lakes Basin. However, which is more important is that most of the lakes are practically undisturbed. Despite their great interest Mongolian lakes have not been systematically studied. Only the most important ones, particularly Khuvsgul nuur have attracted the attention of the scientists, whereas few of the remaining ones have only been occasionally surveyed with very specific scientific purposes. In 2005 Limnologists from Barcelona University started an ambitious project focused to catalog and characterize Mongolian lakes. So far, after eighteen extensive field surveys carried out until 2019 covering practically the entire country, data about surface, hydrology, mineralization, trophic status, and aquatic fauna of 1288 lacustrine water bodies is available. As Mongolian lakes show a great variability due to changing climatic, topographical and geological conditions across the country, they have been grouped in five categories namely: (1) Large permanent freshwater lakes; (2) Shallow permanent or semi permanent freshwater lakes not turbid by inorganic suspended particles; (3) Large permanent highly mineralized, and even saline (not hypersaline) lakes; (4) Shallow lakes and lagoons both permanent or temporal, with slightly to highly mineralized clayey waters; (5) Hypersaline lakes and lagoons. In each type of lake, its characteristic communities of brachiopod and copepod crustaceans are described.

## A proposal of a tool based on lagoon metabolism to estimated ecological status of coastal wetlands

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We present the case study of the ecological state of the Ter Vell lagoon, a Mediterranean coastal lagoon (L'Estarç, Girona, Spain), using the ecological metabolism as indicator. We use Gross Primary Production (GPP), respiration (R) and logratio of Primary production to Respiration (logPR). The objective is to quantify the changes in the ecological state of the lagoon by comparing the metabolism of five consecutive years (2018-2023), including years of greater and lesser degrees of humidity or aridity, and different nutrient intakes. We use the free-water DO (dissolved oxygen) methodology, taking high frequency data with specific probe and the estimation of metabolic rates based on the adjustment of a Bayesian model BASE (BAYesian Single-station Estimation). The comparative data between cycles demonstrate that logPR is positive ( $>0$ ) under situations of clear water dominated by macrophytes, while logPR is negative ( $<0$ ) under situation of stress as for example an excess nutrient inputs. GPP and R values are not so clear because they are more dependent to variables not related to ecological state, such as water turn over. Our data suggest that logPR could be used as metabolism-based indicator of environmental stress.



# Assessment of the ecological status of the Ave Estuary: beyond the challenge of WFD application

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The Water Framework Directive (WFD) constitutes the European legislation that governs the water policy and management, encompassing the assessment of all water bodies. This evaluation extends to estuaries, crucial aquatic ecosystems highly endangered by human activities, resulting in a decline in ecological quality. This study aimed to evaluate the ecological quality of the Ave Estuary, employing the WFD approach and complementing it with an assessment of the impacts of microplastics (MPs) retained in the sediments. Since 2021, water and sediment samples have been collected at the medium estuary during spring and autumn seasons. Results of physical and chemical parameters showed high concentrations of ammonia ( $> 0.30$  mg N/L) and phosphate ( $> 0.11$  mg P/L), contributing to a moderate ecological status across all sampling periods. Benthic macroinvertebrate communities were evaluated as poor with low richness ( $< 10$ ) and Shannon diversity index ( $H'$ ) varying between 0.08 in summer 22 and 0.67 in autumn 23. The findings from the biological elements consistently indicated a poor ecological status for all the sampling periods. MPs concentration in the sediments averaged 330 items/kg during spring 23 and autumn 23. The overall results reveal that the Ave Estuary fails to meet the stipulated requirements of the WFD, highlighting an additional environmental concern with the assessment of MPs that warrants thorough evaluation. Therefore, additional efforts are imperative to improve the water quality in this area.

# Exploring the value of modern pollen and non-pollen palynomorphs in Ría de Vigo (NW Iberia) for palaeoenvironmental reconstructions

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The Ría de Vigo is the southernmost Galician ria. It is a funnel-shaped valley opening in a SW direction towards the Atlantic, and with the sea penetrating about 30 km from the mouth of the ria, partially blocked by the natural barrier formed by the Cíes Islands, to the Ensenada de San Simón in its innermost part. This configuration turns the estuary into a sedimentation trap, with notable differences in the type of sedimentation and sedimentation rates existing along its main axis. Such differences may also affect the sedimentation patterns of pollen and other non-pollen palynomorphs (NPPs types).

We analysed modern sediment samples distributed all along the estuary and observed that linear increases occur for the concentrations and percentages of most of the different pollen types identified. However, exponential increases were identified for some few taxa. A preliminary analysis of these results suggests that the exponential increases may be related to phenological (flowering period of the taxon with respect to the date of sampling) and taphonomic factors, the last driven by matters as total precipitation, size of the basin/sub-basin and source areas, distance to the source areas, river plume and marine currents transportation, sedimentation particle size, sedimentation velocity, sedimentation rate, etc.

Understanding these depositional processes and the main pollen sources in modern sediment samples can help in palaeoenvironmental reconstructions of different types of coastal ecosystems.

# Anthropogenic pressures induce regime shifts in a small saline lake (Lake Alboraj, SE Spain)

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Lake Alboraj is a permanent small karstic lake located at southeastern Spain, included in a Special Area of Conservation (SAC) of the European Natura 2000 Network. The gypsum-rich substratum provided the lake water with a high sulphate concentration (4000-8000 mg/l) and high conductivity values (7-12 mS/cm). The lake has experienced historical and recent hydromorphological and limnological changes due to the influence of anthropic impacts like water extraction, irrigation agriculture, the lowering of groundwater level, introduction of exotic invasive fish species and the arrival of wastewaters via underground. Monthly physicochemical and biological depth profiles were carried out during the years 2016-2024, including dissolved oxygen, redox potential, water temperature, conductivity, pigments (chlorophyll a, and bacteriochlorophylls from sulfur bacteria), and nitrogen and phosphorous concentrations. Several lake regime shifts took place during the study period: a four-year turbid state (2019-2022) was recorded between two clear-water states. Changes affected stratification dynamics, which included the persistent anoxic hypolimnion for four years, the full-anoxic water column during the summer thermic stratification, and the recording of only three mixing events. Lake regime shifts were driven by anthropogenic pressures and impacts that occurred in this sequence: 1) a massive death of carps favored the raise of charophyte meadows; 2) eutrophication provoked the growing of epiphytic cyanobacteria; 3) burial of charophyte meadows by cyanobacteria and decay; 4) a four-year turbid state enhanced by wastewaters and the increase of carp population; 5) the channeling of wastewaters began a new clear-water state. Finally, some insights about the future lake management are proposed.

## Food web approaches in alpine lakes: Iberian lakes as study case

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Food webs in alpine lakes are particularly sensitive to global change through cascading effects altering key ecosystem processes like, for example, energy fluxes and trophic interactions, but often remaining unknown due to the lack of empirical data. We present preliminary results from two intensive sampling campaigns conducted in 2022 and 2023 in Iberian lakes to study structure and functioning of alpine lake food webs. In a comparative approach between two closely situated lakes in Gredos mountains, non-native *Salvelinus fontinalis* inhabiting lake Cimera showed a broader niche (both isotopic and stomach contents), higher reliance on terrestrial food resources, higher  $\delta^{13}\text{C}$  values and higher trophic position than the native *Salmo trutta* inhabiting lake Grande. Furthermore, we observed for the first time the outstanding feeding of *Squalius carolitertii* on micromammals and fish, indicating that the fish species is primarily omnivore but some individuals can become apex predators. Finally, we will also discuss some explorative analysis about the spatial feeding of *S. trutta* based on stomach contents, showing that populations at Grande and Nava lakes displayed a higher reliance on terrestrial food resources compared to populations inhabiting Trampal, Barco and Duque lakes. The study will continue over the next years, covering spatiotemporal and environmental gradients, including morphometric, productivity, anthropogenic, fish and resource gradients. Long-term research in alpine lakes can help to understand the effects of the global change on food webs and provide valuable insights for stakeholders and contribute to management decisions regarding the ecological status of freshwater ecosystems.

# Richness and diversity of birds in temporary ponds of the Las Salinas de Huentelauquén Ramsar site (Chile).

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Temporary ponds or pools are small wetlands with an intermittent hydro-period, common in Mediterranean areas, where precipitation is concentrated in winter. These habitats are home to a great variety of birds, but in Chile little is known about the birdlife associated with these wetlands. The objective of the work was to document the assemblage of birds in the temporary ponds of the Las Salinas de Huentelauquén Ramsar site (257329E-6498780N). The abundance, richness and diversity of avifauna was estimated in 10 pools and the surrounding semi-arid matrix during May-August 2015. In each pool and its matrix (plains), all birds within a 100 m radius were recorded. The sampling effort was two monthly censuses. Richness was assessed using non-parametric estimators. Diversity was estimated by Shannon-Wiener ( $H'$ ) and equity by Pielou ( $J'$ ) with SDR4. The significance of richness,  $H'$  and  $J'$  were evaluated by paired resampling test. In the ponds, 980 birds belonging to 4 orders, 11 families and 23 species were recorded, where Charadriidae presented the greatest richness. In the semi-arid matrix, 1,246 birds belonging to 5 orders, 14 families and 30 species were counted; where Emberizidae had the greatest richness. The diversity in the pools ( $H'=2.12$ ) was lower than in the plains ( $H'=2.24$ ). On the other hand, the equity of birds in the ponds ( $J'=0.68$ ) was greater than in the matrix ( $J'=0.63$ ). These results are discussed in relation to the conservation strategies of the temporary ponds in the semiarid region of central Chile. Funded by PFUE-RED-21992, MINEDUC, Chile.

**OS8**

**Global Change and  
Aquatic Ecosystems**



# Temporal Dynamics of Climate, Landscape, Hydrology, and Ecology: Implications for the Pitillas Lagoon Ecosystem

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The Pitillas Lagoon, a critical aquatic ecosystem in Navarra internationally renowned for its diversity and significance to migratory birds, is experiencing a notable decline in their presence due to recent transformations. This study employs a holistic approach to investigate the evolution of Pitillas and discern temporal trends in climate, landscape, hydrological, and ecological factors that may be influencing the state of the lagoon. By utilizing satellite imagery, climate records (i.e. precipitation and temperature), biological communities' data (i.e. macrophytes, phytoplankton and benthic invertebrates), and water quality monitoring data (i.e. nutrients and general chemistry), we aim to better understand the factors contributing to the lagoon's degradation. Analyses reveal substantial temporal changes: rising minimum temperatures, decreasing water levels, "browning" of its waters, and changes in physicochemical properties, e.g. increased salinity, nitrates, and organic carbon. These changes have impacted biological communities, leading to an increase in cyanobacteria, the presence of potentially toxic algae like *Prymnesium* sp., loss of hydrophytes, and a simplification of benthic invertebrate communities. Results indicate that Pitillas is undergoing a desiccation process, consistent with historical cycles of drying and rewetting. While such processes have occurred in the past, there is a paucity of information on the ecological changes that occurred during these periods. Current changes result from the combined effects of both anthropogenic and climatic factors. Understanding the temporal dynamics of Pitillas Lagoon is crucial for effective conservation and management strategies.

## Biogeochemical effects of restoration actions on Mediterranean wetlands to enhance its climate change mitigation capacity

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Wetlands are strongly active ecosystems in exchanging carbon and greenhouse gases (GHG) with the atmosphere, and they can play a role in climate change mitigation, though this capacity depends on wetland's type and ecological status. This capacity can be improved through management and restoration actions that maintain or recover their ecological health. We have tested the effect of management and restoration actions related to the hydrology, soils, and vegetation to show how these actions change the rates of GHG exchanges with the atmosphere, as well as the radiative balance of the wetland. This has been tested in both coastal and inland wetlands, the latter including both saline and freshwater sites, all of them located in Spain. Further, we used -omic techniques to describe the community composition and trace gene expression of specific microbial guilds responsible for the main GHG exchange processes, in order to understand the reasons for the observed biogeochemical changes promoted by the assayed actions. This work was supported by the project LIFE Wetlands for Climate (LIFE19 CCM/ES/001235) funded by the European Union, as well as by projects CLIMAWET-CONS (PID2019-104742RB-I00) and WETCARVER (PDC2022-133205-I00), both funded by Agencia Estatal de Investigación and the Ministerio de Ciencia e Innovación (Gobierno de España).



# Combined effects of salinisation and temperature on microbial-mediated leaf decomposition and invertebrate consumption

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Water salinization is a recognized growing threat to freshwaters, whose consequences on streams' function, per se or concomitantly with other stressors, are still far from clear. This microcosm study evaluated the combined effect of salinization (0 and 4 g/l NaCl) and temperature (5, 15, and 20 °C) on microbial-mediated oak leaf litter decomposition, with fungal biomass, sporulation, and microbial respiration as microbial descriptors. Invertebrate consumption was also assessed using the common shredder *Sericostoma vittatum* (Trichoptera, Sericostomatidae). Mass loss was affected by temperature and interaction between salinity and temperature. Under salt conditions, mass loss was higher at 15 °C and reduced (~ 10%) at 20 °C. Microbial activity was lower at 5 °C and higher at 15 and 20 °C, irrespective of salinity. Fungal biomass was affected by both temperature (5 < 20 < 15 °C) and salinity (4 < 0 g/l NaCl), although the interaction between both was not significant. The interaction of both variables affected the production of spores: salt addition strongly reduced sporulation rates at all temperatures despite a significant increase in conidial production with temperature. Invertebrate leaf consumption was significantly reduced only by salinization. Overall, our results seem to indicate that temperature may modulate the effect of salinization (at least at ≥ 4 g/l NaCl) on stream leaf decomposition. While stronger salinization effects may be observed at higher temperatures, a consistent strong inhibition of shredders' feeding behavior promoted by salt, regardless of temperature, may anticipate important repercussions on streams' secondary production throughout the year.

# Assessing GLOBAL river SALinity and associated ions (GlobSalt)

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Freshwater salinization (FS) impact in rivers remains poorly understood compared to other stressors, with some regions (e.g. Asia, Africa) poorly explored. To assess how pervasive this issue is globally and identify salinization hotspots, we compiled global data on river salinity and associated ions. We retrieved information from 62 different datasets, harmonized it and merged it with watersheds from HydroATLAS to summarize information at the sub-catchment scale. Our global data set (GlobSalt database) features 13 parameters: electrical conductivity (EC), pH, major ions ( $\text{Na}^+$ ,  $\text{K}^+$ ,  $\text{Mg}^{2+}$ ,  $\text{Ca}^{2+}$ ,  $\text{Cl}^-$ ,  $\text{SO}_4^{2-}$ ,  $\text{SiO}_3^{2-}$ ), total organic carbon, nitrate, ammonium, and phosphate. GlobSalt contains approximately fifteen million records on a monthly scale for two hundred seventy thousand river stations from 1980 to 2023 from all continents except Antarctica. EC and pH were the most commonly measured parameters. The global median EC was  $506 \pm 205 \mu\text{S cm}^{-1}$ , with 60% of rivers falling in the range of 50 to  $500 \mu\text{S cm}^{-1}$ , which is within the salinity niche of most freshwater organisms. Sodium and chloride showed the strongest correlation ( $r^2 > 0.6$ ) with EC. We found a large spatial variability in EC, with some regions such as the Mediterranean, the Midwest of the US, arid regions of Argentina and Chile and Southwestern Australia having high mean salinity values. Temporally, EC was fairly stable, with no evidence for strong increasing or decreasing trends from 1980 to 2023. Overall, GlobSalt represents a critical resource for improving our understanding of FS dynamics, identifying regions at high risk and informing management strategies.

# Synergistic and long term effects of emergent chemical contaminants alter leaf litter decomposition and the activity of aquatic decomposers

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We will use leaf litter decomposition and decomposer activity in freshwaters as a model system to discuss the effects of emergent chemical contaminants in mixtures. We will mainly focus on the effects of micro/nano-plastics and pharmaceuticals due to their global distribution. Emergent chemical contaminants decreased leaf litter decomposition and the activity of aquatic decomposers. The effects in mixtures were mainly synergistic. Moreover, the effects along generations showed that effects were prolonged over time making the effects of emergent contaminants in freshwaters of great concern. We will discuss measures to mitigate the problem involving water agencies and citizens.

# Unexpected CO<sub>2</sub> influx and efflux daily cycles in bare dry lake sediments

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CO<sub>2</sub> fluxes from dry sediments are key in the carbon cycle of inland waters, but how those fluxes respond to occasional rewetting, and the mechanisms shaping these responses is poorly understood. Here, we conducted a field rewetting experiment to determine rain-related variations in CO<sub>2</sub> fluxes, stable isotopes (13C-CO<sub>2</sub>), and microbial community composition in long-term dry sediments of Lake Gallocanta (Spain). We measured both influx and efflux of CO<sub>2</sub> from the sediments. CO<sub>2</sub> fluxes were only marginally sensitive to rewetting, being time and the interaction timeXrewetting the main explanatory factors. To further assess this pattern, we built a numerical model by using the CO<sub>2</sub> measurements and 13C-CO<sub>2</sub>. We found that at least two mechanisms control the daily CO<sub>2</sub> flux cycles. The first one, biological and related to aerobic respiration, peaking in midday hours and resulting in CO<sub>2</sub> efflux. The second, abiotic, responsible for the net CO<sub>2</sub> exchange during nighttime (CO<sub>2</sub> influx). Among the microbial community, CO<sub>2</sub> fixation could be performed by members of the Cyanobacteria (oxygenic photosynthesis), the Campylobacterota (chemolithotrophic sulfide oxidation) as well as by ammonia-oxidizing Crenarchaeota. However, the positive relationship between Cyanobacteria and CO<sub>2</sub> efflux, and the absence of a differentiated isotopic signature during fractionation, led to the conclusion that their role in the CO<sub>2</sub> influx was likely minor. We suggest that abiotic processes such as weathering are key to properly determine the CO<sub>2</sub> flux of long-term dry sediments, and that net CO<sub>2</sub> emissions might not be an accurate proxy for carbon remobilization from drying lake sediments.

# Cow-pasture impact in Alpine mires as recorded by peat properties and diatoms

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Peatlands are highly vulnerable ecosystems playing a crucial role in carbon sequestration and biodiversity conservation. Cattle grazing and trampling accelerate their degradation and pose a significant threat to habitat integrity and biodiversity, particularly in the southeastern Alps. The main objective of this research was to examine the impact of grazing on physical, chemical, and biological characteristics of peat, with a focus on diatoms, to gain insights for a sustainable conservation of these habitats. Seven 50-cm deep peat cores were sampled from mires located in the Adamello-Brenta Nature Park (Trentino, Italy) along a grazing-induced disturbance gradient. Results indicated that grazing primarily affected the upper 15 cm of the peat, resulting in increased density, reduced water content due to compaction, and lower carbon-to-nitrogen ratios, the latter one caused by cow manure inputs and increased mineralization processes. Over 200 diatom taxa were recorded and several of them are included in threat categories of the Red List for central Europe (e.g., *Cymbopleura valaiseana*, *Eunotia hexaglyphis*, *E. triodon*). Highly-grazed areas exhibited a higher percentage of species that live in eutrophic conditions, linked to increased nutrients originated from cattle manure, and aerial species, which survive in environments with unstable water availability. This research demonstrates that diatoms can be used as efficient indicators to highlight the impacts caused by cows.

# Riparian forests are key regulators of thermal conditions across European rivers

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Understanding the drivers of riverine thermal conditions is critical to preserving river biodiversity and functioning in the face of global change. In particular, riparian forests are a key ecosystem component providing shade and modulating air movement above the river and in exchange with the adjacent environment. To better understand the role of riparian forests as regulators of riverine thermal conditions, we investigated 20 sites in each of six basins from Southern Spain to Finland. Combining remote sensing data and in situ measurements, we studied how forest shade and land cover composition in the riparian zone influence river water and air temperature. Shade over the river was estimated with hemispheric photos taken in the field while land cover was defined using CORINE-Land Cover (Copernicus Land Monitoring Service). Daily water and air temperature were measured in the field from spring to late winter over a year. Forest shade had a significant effect on water and air temperature, yet, the direction and magnitude of this effect was season-dependent. Also, the land cover composition was related to the seasonal regime of air temperature (i.e., the combination of seasonal mean, minimum, maximum, SD, and range), while the dominance of forest/non-forest cover was related to the seasonal regime of water temperature. Our findings will help to predict the implications of warming and land use change for ecosystem functioning and suggest riparian forests as an important tool for climate change adaptation in rivers.

# Incorporating water temperature and discharge into climate change models of freshwater invertebrates across Europe

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Freshwater organisms are expected to be profoundly impacted by the predicted increase in water temperatures and discharge alterations associated with climate change. However, available models focus mostly on changes in air temperature, potentially failing to incorporate these impacts. Given that freshwater biodiversity is declining at an alarming and exponentially increasing rate, there is an urgent need to assess the relation of this biodiversity to water temperature and discharge to plan effective management and conservation actions. Here, we modeled the distribution of freshwater macroinvertebrates across Europe for present and future conditions including recently available data on water temperature and discharge. We also included other environmental variables that might be relevant in understanding the current spatial distribution of invertebrates (e.g. geology, adjacent land use). We used 40 datasets of standardized monitoring protocols of freshwater invertebrates spanning 23 years. The study includes a comparison of the predictions for the future at different taxonomic resolutions, from species to family level, to discuss the high variability of response to water temperature between species of some genera and families. This work is a first step towards the selection of taxa sensitive to climate change, the final aim being to build a multimetric macroinvertebrate-based index that could serve to monitor the effects of climate change on rivers and streams at the European scale.

# Will climate change force extinction of endemic species? Conservation challenges for *U. tumidiformis*

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Climate change and shifts in temperature and precipitation are increasingly recognized as significant contributors to ongoing loss of biodiversity. The Mediterranean region is highly vulnerable as a biodiversity hotspot and being highly susceptible to desertification. Among the affected species, freshwater mussels emerge as one of the most imperiled invertebrate groups globally, especially under stress in this region. Of particular concern is *Unio tumidiformis*, an endemic endangered species found exclusively in the southern Iberian Peninsula, inhabiting temporary Mediterranean-type streams. These freshwater mussels rely on fish hosts, specifically those belonging to the genus *Squalius*, for successful reproduction. This study aimed to assess the vulnerability of *U. tumidiformis* to climate change by determining current habitat suitability, and projected future conditions, as well as those of its hosts. Using an ensemble forecasting approach, we modeled the potential distribution of the species and evaluated future shifts in habitat suitability using climate data projections from CMIP6 (IPCC).

Alarmingly, our results indicate a projected reduction of 99% in climatically suitable areas for *U. tumidiformis* across the Iberian Peninsula by 2040. Suitable habitat for its fish hosts may decrease by up to 42% throughout the century, with favorable habitats persisting in the northern regions. These findings underscore the urgent need for conservation interventions. Potential strategies may include translocating *U. tumidiformis* to the northern extent of their historical range and/or implementing stream engineering to enhance resilience to droughts. Such measures are crucial to mitigate threats posed by climate change and protect this critically endangered species and its associated ecosystems.



# Island lake ecosystems experience abrupt regimes shifts due to Global Warming

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Warming is inducing regime shifts in lake ecosystems worldwide, but there are still uncertainties in quantifying and predicting such changes across various spatial and temporal scales. Here, we advance our understanding by combining multi-lake sediment records from the Azores archipelago (North Atlantic Ocean) encompassing more than a century of anthropogenic and climate change, to test the regional coherence of lake ecosystem changes in response to global warming. We found a threshold response in all five diatom communities records of 0.3 °C above the North Hemispheric temperature anomaly. Furthermore, the regional shift towards an increase in the dominance of small planktonic species agrees with previous reports on the effects of increasing the strength and length of lake stratification in the reorganisation of planktonic communities. This shift also drives homogenisation in the spatial distribution of diatom species due to the increase in dominance, resulting in over 25% net loss of regional diversity. These findings underscore the key role of lake sediment records as sentinels of responses to global environmental changes occurring across varying spatial and temporal scales, to underpin evidence-based decisions on the protection, restoration and conservation of lake island ecosystems, its biodiversity and associated ecosystem services.

# Multiple stressors alter greenhouse gas concentrations in streams through local and distal processes

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Streams are significant contributors of greenhouse gases (GHG) to the atmosphere, and the increasing number of stressors degrading freshwaters may exacerbate this process. However, it is unclear whether the influence of multiple stressors on GHG concentrations in streams is controlled by local processes (e.g., increases of in-situ metabolism) or distal processes (e.g., changes in upstream and terrestrial GHG production). Here, we hypothesize that the mechanisms controlling multiple stressor effects vary between carbon dioxide (CO<sub>2</sub>) and methane (CH<sub>4</sub>), with the former mainly responding to distal processes and the latter being more influenced by changes in local stream metabolism. To test this hypothesis, we measured stream metabolism and the concentrations of CO<sub>2</sub> (pCO<sub>2</sub>) and CH<sub>4</sub> (pCH<sub>4</sub>) in 50 stream sites encompassing marked gradients of nutrient enrichment, oxygen depletion, thermal stress, riparian degradation and discharge. Our results indicate that nutrient enrichment was associated with higher stream heterotrophy and pCO<sub>2</sub>, whereas increased oxygen depletion and water temperature resulted in higher stream pCH<sub>4</sub>. These models indicate that CO<sub>2</sub>-equivalent concentrations can double in streams that experience high nutrient enrichment and oxygen depletion. Structural equation models revealed that the effects of nutrient enrichment and discharge on pCO<sub>2</sub> were related to distal processes rather than local metabolism. In contrast, pCH<sub>4</sub> responses to nutrient enrichment, discharge and temperature were related to both local metabolism and distal processes. Our study illustrates potential climatic feedbacks resulting from freshwater degradation and provides insight into the processes mediating stressor impacts on the production of GHG in streams.

# Impact of Global Change during the Anthropocene on the ecological status of a Mediterranean high mountain lake

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Mediterranean high mountain lakes are highly sensitive ecosystems to global change causing significant changes on their ecological status. A sediment core was taken in Borreguil Lake (2800 m a.s.l., Sierra Nevada National Park) and dated with radioisotopes, spanning the last ~180 years. Our aim was to determine whether the ecological status of the lake has changed during the Anthropocene and, if so, whether these changes were linked to climate change and/or direct anthropogenic impact. The core was analysed for diatoms, chironomids, pigments, and geochemical data. Climate variables were obtained from a climate model developed specifically for our study area. The composition of diatoms, chironomids, and pigments underwent the most significant changes since the 1980s and 1990s. The concentration of  $\beta$ -carotene and chlorophyll-a, which are indicative of algal biomass, increased since 1940 and particularly since 1990. The increase in algal biomass was primarily attributed to the rise of pigments specific to chlorophyceae, cryptophyceae, and cyanobacteria. However, diatom-specific pigments decreased. Chlorophyll-a concentration changes were mainly associated with spring temperature and precipitation. Aphanizophyll concentration, which is specific to nitrogen-fixing cyanobacteria, increased since the 1990s. This suggested a nitrogen limitation in primary production. The changes in diatom and chironomid assemblages, linked to summer temperature, and annual temperature and precipitation, respectively, indicated a trend of increasing aridity and warmer temperatures towards the present. Thus, our results indicated significant changes in primary and secondary producers specially since the second part of the 20th century concurrent with trends in rising regional air temperature and declining precipitation.

# Organic carbon remobilization from the dry bed of the desiccated Aral Sea

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Many lakes in the World, particularly from endorheic basins in arid and semi-arid areas, are shrinking. This implies that the sedimentary carbon sink, sometimes the result of thousands of years of burial, may be prone to remobilization once the accumulated organic matter is exposed to atmospheric oxygen. To test this hypothesis, we have visited the endorheic lake Laguna de Gallocanta in Spain, and the largest vanishing lake on Earth, the Aral Sea in Kazakhstan. Applying a time for space substitution approach, we investigate if the organic carbon content of the sediments follows a chronosequence of drying, whether carbon emissions are actually happening in those sediments, and which organic carbon fractions are being remobilized. We put our results in the greater context of the potential implications of emissions from those emerged sediments for the global carbon cycle, and also discuss the potential role of recovering those lakes as a mitigation action against climate change compared to other customary mitigation alternatives.

# The importance of wetlands to fulfill climate change mitigation targets

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Within the framework of the Paris Agreement, the European Union has set its objective of climate neutrality by 2050. In order to achieve a significant reduction in Greenhouse Gases (GHG) emissions, it will be essential to improve the contribution of carbon sinks like forest, agriculture croplands and wetlands.

The European legislation establishes that, from 2026, wetlands GHG emissions must be accounted to meet the GHG mitigation targets. Nevertheless, at Spanish level, the Miterd's Carbon Footprint Register does not currently include actions in wetlands as absorption project typologies. To amend this, the proposal to update this Register seeks to reinforce the possible actions related to wetlands management and restoration enhancing carbon sequestration and GHG emissions abatement. Equally relevant is the proposal for a European Commission to establish a EU certification framework for carbon removal (CRCF), currently under negotiation which, once approved, will be a key element for promoting carbon sequestration in wetlands.

It is thus essential to develop methodologies that allow the calculation of GHG emissions/absorptions. The Intergovernmental Panel on Climate Change (IPCC) guidelines originally included some wetland types (peatlands, mangroves), now being expanded to other types for which, such as the case of Mediterranean coastal wetlands or inland saltwater wetlands, more research is needed. Some already available studies demonstrate the correlation between good conservation status, good ecological status and positive carbon sequestration balances of Mediterranean wetlands, which could enhance the support to the conservation and restoration of such valuable ecosystems capable of providing a sort of ecosystem services.

# DAMOLAKE: a multi-scale experimental approach to assess greenhouse gases emissions under warming and nutrient-enriched conditions in lakes

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Anthropogenic pollution and global warming can be promoters of disturbances in biogeochemical cycles (e.g., carbon and nitrogen). These changes can lead to an increase in greenhouse gas (GHG) emissions and nutrient input from inland freshwaters, such as lakes. For these reasons, the potential of lakes to be a sink or source of GHG (e.g., methane) and the factors involved must be addressed. Aquatic vegetation and the associated microbial community are crucial in the methane metabolism (considering both production and consumption) of freshwater ecosystems. In a global-change context, submerged meadows will be impacted and their senescence will cause a massive contribution of organic matter to sediment, favoring methane production and emission to the atmosphere. Further, changes in submerged meadows also impact distinct levels of the trophic web, such as plankton communities. Using a microcosm and mesocosm approach, we have established three research goals: (i) to assess the transformation of macrophyte-derived organic matter to methane, (ii) to compare the effect of the vegetation type (vascular plants vs charophytes) under different temperatures on GHG production, and (iii) to assess the impact of charophytes under warming and nutrient-enrichment conditions. In a microcosm experiment, the transformation capacity of macrophyte-derived detritus to methane is species-specific and estimated, on average, as 1.1 kg of methane/kg of macrophyte dry weight year. Moreover, charophytes and vascular plants showed different GHG production rates. In an indoor mesocosm experiment, a 5-degree warming scenario increased three-fold the methane production from charophytes meadows and promoted changes in phytoplankton and zooplankton functional groups.

# Can leaf conditioning status affect the decomposition process in salinized streams?

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Secondary salinization is a global threat to freshwater ecosystems, with important negative consequences on streams' structure and functioning. Here we assessed the effects of salt contamination (0, 1, 4, and 6g NaCl/L) on the decomposition of oak (*Quercus robur*) leaves with distinct conditioning histories. Mass loss, microbial activity, fungal biomass, and sporulation rates were evaluated on leaves previously immersed in the stream for 7 (P) or 21 days (C), and further submitted to a 2-week salinization period in the laboratory. Both leaf types were offered to the shredder *Schizopelex festiva* (Trichoptera, Sericostomatidae) maintained in the corresponding salinized media. Mass loss of C was higher, but strongly inhibited by increasing salinization; no effects were observed on P across salt treatments. A marked depressive effect of 6g/L salt on all microbial descriptors was observed on either leaf types. At 1g/L, C showed higher mass loss, fungal biomass and microbial activity, while P presented higher fungal biomass and sporulation at lower and intermediate values. *S. festiva* consumption was consistently reduced when fed C. Distinct and richer fungal communities associated with P may have favored shredders' consumption across salinities, despite a depressive effect of waterborne and/or food-mediated salt-toxicity. Results suggest that less conditioned leaves (P) are more able of counteracting the deleterious effects of salt contamination on leaf decomposition ( $\leq 4\text{g/L}$ ) through energetic investments on fungal propagation strategies. Higher sporulation rates and biomass accrual of richer salt-tolerant fungal communities may support the stream fungal pool, and stimulate leaf incorporation into secondary production by leaf-consumers

# Effects of agriculture and forestry in Galician headwaters

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Increasing land occupation by agriculture and forestry is leading to the degradation of freshwater ecosystems. These activities can mobilize sediments, nutrients, and pollutants, which can exceed the recovery capacity of streams and rivers compromising water quality and quantity and thus ecological status. Our study aimed to analyze the effects of agroforestry on the ecological status of streams and rivers and to assess the role of a range of riparian buffer zones in preserving biodiversity and maintaining Good Ecological Status. For this, we studied 60 small streams and rivers in Southern Galicia (NW Spain) affected by intensification gradients in agroforestry and applied diverse methodologies (GIS, field protocols) to assess pressures at basin, riparian, and channel spatial scales. Results allowed the assessment of ecological impact under different levels of agroforestry activity, and the evaluation of the most informative and relevant pressure-affected variables. The relationships between the ecological status and the variables measured at various scales (basin, riparian, and channel) under agroforestry pressures were studied, and modeled. Decision trees were used to determine the quantitative criteria and thresholds needed to achieve environmental objectives in rivers and streams under varying levels of agroforestry pressure. This pilot study provides a scientific basis for managing freshwater ecosystems and for setting protection levels at different spatial scales.



# Alteration of the hydrological, thermal and metabolic regimes produced by hydropower dams in Picos de Europa National Park

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Mountain rivers are often perceived as ecosystems of high naturalness, characterized by minimal human pressures, especially when they are located within protected areas (e.g., National Parks). However, reality frequently challenges this perception, as mountain rivers are ideal places for the installation of hydroelectric developments. Many of these infrastructures, despite being relatively small, can cause significant alterations to the hydrological and thermal regime and, consequently, impacts on the biodiversity and ecological processes. In this context, the aim of this work is to evaluate the hydrological, thermal and ecological alteration caused by hydroelectric infrastructures in the river network of the Picos de Europa National Park. In order to meet this objective, a study has been designed that includes 13 study points, 6 of them impacted by small hydroelectric power plants and 7 controls. Continuous monitoring of flow, temperature and river metabolism was carried out from June to October 2023. The results showed that, in general, hydropower plants produce a significant increase of temperature and decrease in flow downstream of the dams; the response of the metabolism has shown greater variability that is probably related to the influence of other factors, such as the biomass of primary producers or the amount of resources, which are currently being analyzed. The results of this study will be very valuable for the management of these infrastructures in order to co-design sustainable hydropower schemes needed to achieve a balance between electricity generation, impacts on ecosystems and benefits on society, supporting the achievement of the Green Deal targets.

# Effects of drying and urban pollution on macroinvertebrates in three permanent Mediterranean rivers

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Rivers receive different anthropogenic stressors, which can act alone or in combination. Their effects depend on the intensity, frequency and order (legacy effects) of the stressors, with consequences for biological communities, ecosystem functioning and human wellbeing. We conducted a field experiment to assess the effects of drying (flow interruption simulation) and urban pollution (Wastewater treatment plant, WWTP) on macroinvertebrates communities in three 3rd order permanent Mediterranean rivers. First, we placed bricks (artificial substrates) for macroinvertebrate colonization. After that, we simulated two intensities of drying (pulse: short and repeated; press: long and continuous) moving bricks in/out the riverbed. Finally, we transferred the bricks downstream to a WWTP effluent, obtaining a total of 7 different treatments. We have analyzed individual and joint effects of these stressors on the alpha and beta diversity and species composition of the macroinvertebrate communities. Drying significantly decreased species richness and total abundance in the three rivers, and changed the composition of communities due to the loss of aquatic taxa, while its effect on Shannon diversity varied among rivers. Urban pollution changed the composition of communities due to a replacement by more tolerant taxa and reduced the macroinvertebrate abundance in one river. This suggested that future water scarcity in permanent rivers due to climate change and water abstraction can cause a severe impact on the structure of macroinvertebrate communities. The risk of effects of WWTP effluents on the community could increase with lower flows and less dilution capacity.

# Late Holocene climate dynamics in the Azores Archipelago

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The location of the Azores Archipelago makes this group of islands an excellent setting for investigating past long-term temperature and precipitation changes in the central North Atlantic region. Here, we present a chironomid-based quantitative temperature reconstruction and a record of oxygen isotope composition of chironomid head capsules for the last ca. 1200 years, based on the Lake Prata core (São Miguel Island, Azores). Chironomids from Lake Prata show that the temperature trend is aligned with the prevailing past global temperature trends, indicating warming during the Medieval Climate Anomaly (MCA; ca. 950-1300 CE) and subsequent cooling during the Little Ice Age (LIA; ca. 1300-1800 CE). Furthermore, changes in lake water oxygen isotope composition from chironomid head capsules were recorded with more positive  $\delta^{18}\text{O}$  values ( $18.2 \pm 0.2\text{‰}$ ) during the MCA and more negative values (varying between  $14.8\text{‰}$  and  $17.6\text{‰}$ ) during the LIA, indicating a positive shift in the precipitation-evaporation balance and/or heavier precipitation in winter relative to summer. Our study revealed that, in subtropical regions with a temperate oceanic climate and low thermal variation, there is a limited correlation between the isotopic composition of precipitation and temperature. Additionally, the temporal sequence of oxygen isotope values obtained from chironomid head capsules closely corresponds to variations in precipitation seasonality inferred from distinct climatic proxies. This consistency underscores the robustness of quantitative reconstructions and indicates that  $\delta^{18}\text{O}$  chironomid records are promising for future investigations, especially in the context of lakes located on oceanic islands characterised by limited thermal variability.

# Assessing the combined impacts of land-use intensification and climate on Iberian stream macroinvertebrates

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Freshwater ecosystems are deteriorating rapidly due to the growing number of impacts. The combined action of land use intensity (LUI) and climate change poses an unprecedented challenge for assessing the ecological status of rivers. Changes in temperature and flow due to climate change could alter the impact of land use intensification, amplifying the effects of stressors, such as nutrient enrichment or hypoxia. However, we know little about the capacity of biomonitoring tools to detect the individual and combined effects of these impacts. Using 35 independent stream reaches, we assessed the combined effect of LUI and climate on aquatic macroinvertebrate communities at local (river) and regional (basin) scales. As response variables, we selected common metrics used in biomonitoring (e.g., IBMWP and IASPT) and indicators of trophic composition of macroinvertebrate communities (percentage of predators, gatherers and shredders). We found that nutrient enrichment and LUI were the main factors explaining changes in aquatic macroinvertebrate communities. In particular, the IASPT metric showed a strong response to LUI irrespectively of the climatic context. Overall, higher levels of aridity and air mean temperatures were associated with lower values of biomonitoring indices related to the composition of mayflies, stoneflies and caddisflies. Interactive effects were only found for trophic composition indicators, suggesting that they may be promising metrics for assessing the combined effect of changes in LUI and climate. These results offer insights into the effects of global change on rivers, and can aid to identify indicators capturing both individual and combined impacts on aquatic ecosystems.

# Assessing the role of vegetation on CO<sub>2</sub> evasion from dry freshwater sediments

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Dry inland water bodies have garnered comparatively little attention as a source of carbon dioxide (CO<sub>2</sub>) towards the atmosphere, although they can emit CO<sub>2</sub> at approximately similar rates to terrestrial land and lotic water bodies. While terrestrial vegetation often colonizes dry freshwater sediments, research on CO<sub>2</sub> fluxes has been focused on unvegetated (bare) sediments. Here, we explore the Dryflux-II project database, a unique global effort compiling standardized CO<sub>2</sub> measurements for more than 120 dry inland water bodies across different biomes. We performed CO<sub>2</sub> chamber flux measurements in vegetated and bare plots in each system under dark and light conditions and characterized the sediments and the main environmental drivers. The findings revealed that in light conditions, the concentration of CO<sub>2</sub> flux was lower in vegetated than in bare sediments under similar conditions.

Conversely, in dark conditions, vegetated plots exhibited positive CO<sub>2</sub> fluxes of similar or higher magnitude than in bare sediments, likely attributed to plant respiration. That pattern was consistent across ecosystem types (lakes, reservoirs, rivers, ponds) and biomes (from arid to arctic). However, when considering the net CO<sub>2</sub> flux all the systems except reservoirs presented a positive flux towards the atmosphere.

Our results indicate that vegetation significantly influences the magnitude of the net CO<sub>2</sub> flux of dry sediments but without changing its direction to the atmosphere in most cases. A proper understanding of terrestrial vegetation dynamics in dry sediments is paramount as it might play a vital role in counterbalancing the carbon losses from dry sediments mediated by microbial respiration.

**OS9**

# **Extreme Aquatic Ecosystems**



# Can hypersaline endorheic saline lakes act as a C sink? preliminary results from the CARINSAL project

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Saline endorheic aquatic systems (SEAS) represent 40% and 20% of total lake volume and surface area, respectively, at the global scale. Our knowledge about the carbon budget of SEAS is very incipient, however recent findings pointed out that these ecosystems can accumulate huge amounts of organic matter. Considering SEAS could be significant carbon sinks, since they store carbon in anoxic sediments, the impact of drying interacting with salinity on CO<sub>2</sub> emission is a key point that requires being investigated. Through an experiment approach in the laboratory, we analyzed how dry benthic sediments from different endorheic shallow lakes along a natural salinity gradient, respond to rewetting in terms of net CO<sub>2</sub> emissions. Our preliminary results show a clear distinctive pattern. Whereas hiposaline and freshwater lakes showed a net CO<sub>2</sub> release after rewetting, hyper and mesosaline lakes, showed a net CO<sub>2</sub> uptake under dark conditions. Results suggest that biotic processes such as microbially driven dark carbon fixation, could be responsible for these net CO<sub>2</sub> uptake in presence of inorganic reduced sulfur compounds. Similarly, algal and bacterial biomass and extracellular enzyme activities showed different responses to rewetting along the salinity gradient. Although these are very preliminar results, the importance that sediments of saline lakes could have acting as a C sink encourages us to advance along this line and unravel which processes could be behind these results, their temporal variability and driving factors. The implementation of isotopic, mineralogical and molecular approaches (metagenomic, q-PCR of specific genes) are necessary to advance in this line.

**OSIO**

**Ecohydrology and Groundwater**





# Groundwater dynamics in Mediterranean environments: A case study of Menorca's aquifers responses to climate variability and human impacts

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Groundwater serves as a critical and substantial freshwater source in regions with Mediterranean climate conditions, facing notable precipitation variability, meteorological droughts, and increased water demand during dry seasons. The reliance on groundwater intensifies in areas with predominant permeable lithologies, contributing to aquifer recharge and non-perennial stream presence. Uncertainties in future water availability together with rising tourism in Mediterranean coastal areas emphasize the urgent need to understand groundwater responses to precipitation variability for sustainable resource use. In this research, we analyze the temporal evolution of 18 wells situated on the island of Menorca (Spain) over the last 36 years (1984 - 2020). Analysis of monthly and annual trends using the Mann-Kendall test reveals diverse patterns: 3 series display an increasing trend, 11 a decreasing trend, while 4 lack significant trends. To further our understanding, these monthly series have been correlated with climatic variables. To do so, the Standardized Precipitation Index (SPEI) was correlated to the Standardized Groundwater Index (SGI) at various time scales. Three primary responses emerge: i) medium-term SPEI (>12 months) influences SGI; ii) non-significant correlation to SPEI; iii) negative correlation (anti-phase relationship). These diverse responses underscore the complex reality of groundwater dynamics, primarily attributed to lithological variations and the percentage of highly permeable rock strata in aquifer recharge areas. Extractions for human consumption have disrupted aquifer-climatic relationships, with 14 out of 18 cases showing insignificant or negative recharge correlations. Understanding the complex relationship between precipitation, extraction, and aquifer response is crucial for effective water resource management amid climate change.

# Effects of forest maturity and naturalness on ecohydrological processes: a global-scale meta-analysis

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Forests play a crucial role regulating hydrological processes because they influence water quantity and quality, regulate thermal regimes at multiple scales, act as carbon sinks, and protect soil from erosion. However, accurate knowledge of the hydrological behavior of forests is still needed, with special concern on how forest characteristics such as composition, growth stage, or naturalness influence their hydrological response. An in-depth comprehension of the relationship between hydrological processes and specific forest characteristics will improve the current strategies for integrated catchment management of both water resources and the associated ecosystems. This is paramount in the actual context of global change and associated management actions taken to mitigate risks for humans and biodiversity.

In this study, we focus on assessing the relationship between forest maturity and naturalness and some of the most relevant hydrological processes at a global scale. For that purpose, we conducted a systematic literature review and a meta-analysis to determine the difference in the responses depending on maturity and forest type. The articles reviewed showed that soil moisture, evapotranspiration, and air temperature were the most studied processes. We found that mature natural forests have more stable hydrological conditions than young planted forests, with higher infiltration and soil moisture rates and lower evapotranspiration and temperature values. These findings provide a good baseline for further research on the relations between hydrological processes and forest maturity and naturalness. Yet, there is a need for further impact-control studies that consider these characteristics to confirm more precisely the results reported here.

**OSII**

**Aquatic Ecotoxicology and  
Environmental Risk Assessment**



# Comparative toxic effects of caffeine and naproxen on rotifer populations from Fuente de Piedra wetland (Málaga, Spain)

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The structure and function of Mediterranean wetlands is being strongly affected by the presence of emerging contaminants (pharmaceuticals, pesticides, industrial chemicals, surfactants, endocrine-disrupting compounds, and personal care products, among others). In Spain, Fuente de Piedra (37°6'N, 4°44'W) is considered as one of the most important wetlands. The discharge of treated wastewater from the nearby Fuente de Piedra town is one of the main problems in this ecosystem. Since 1999, effluents of treated wastewater go through several small semi-natural ponds to try to reduce nutrient concentrations in the water inflows into the main water body. Two of the main emergent contaminants we detected in Fuente de Piedra (FP) are caffeine (stimulant drug) and naproxen (medical drug). The objective of this study is to analyze the acute toxicity of these compounds (from 0 to 450 mg/L) and their interactions on two rotifer species isolated from FP wetland (*Brachionus rubens* and *Keratella tropica*), as planktonic organisms may be used as biological indicators of contaminant impacts on aquatic systems. These rotifer species were chosen because previous results showed that *B. rubens* inhabit those semi-natural ponds in FP, while *K. tropica* is nearly absent from the most contaminated water body in FP. Preliminary experimental results showed differences in the median effective concentration values (EC50) among rotifers. We will discuss further implications of our results on the risk assessment in the FP wetland.

# Planktonic and benthic cyanotoxin-producing cyanobacteria in Spanish National Parks: a risk assessment perspective

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This study aims to characterize cyanobacterial communities from two protected areas (Monfragüe and Guadarrama Mountains National Parks) to evidence the presence of cyanotoxin-producing species and the risk they may pose to users and fauna. For such purpose, planktonic and benthic communities from diverse aquatic ecosystems (reservoirs, ponds and rivers) were studied in from 2021 to 2023. Toxic cyanobacteria were identified in all types of ecosystems explored combining approaches such as microscopy, genetic (PCR-based screening and metabarcoding) and analytical chemistry. In reservoirs, high levels of cyanobacterial biomass were determined (>500 µg/L of chlorophyll a), along with the confirmation of diverse cyanotoxins such as hepatotoxins (microcystins) and neurotoxins (saxitoxins and anatoxins). *Microcystis* sp. was apparently responsible for the persistent microcystins detection (up to >200 µg/L). Microcystins were also detected in ponds and our results suggests connectivity with the surrounding reservoirs since we found overlapping of identical sequences of the genes 16S rRNA and *mcyE* (microcystin-related). In rivers, toxic benthic mats were found, and particularly, *Microcoleus*-dominated mats containing neurotoxins (anatoxins) reached high abundance levels of up to 50% of coverage of the riverbed. Microcystin and saxitoxins-related genes were confirmed in some benthic mats, but these toxins were not detected. Furthermore, a long-term monitoring conducted suggests recurrent proliferation of such mats in some rivers. Our results were utilized to design risk assessment protocols to minimize negative effects. As a whole, these findings reinforced the need to include toxic cyanobacteria in periodic monitoring programs of the ecological state of various aquatic systems.

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# Toxicity of single and mixture of antibiotics: model organism's insights

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Sulfamethoxazole (SMX) and Trimethoprim (TRIM) were considered priority substances to be monitored in inland waters by the Water Framework Directive, due to the risk that pose to the aquatic environment. These antibiotics and their mixture (MIX) are used in human and veterinary medicine, and aquaculture to prevent and treat the most diverse bacterial infections. This study aims to evaluate the potentially toxic effects of environmentally relevant concentrations of SMX (150 µg/L), TRIM (30 µg/L), and MIX (150 µg/L SMX + 30 µg/L TRIM) in different model organisms: *Allivibrio fischeri* (bioluminescence inhibition), *Escherichia coli* ATCC25922 (growth inhibition), *Lemna minor* (growth inhibition and biochemical biomarkers), *Daphnia magna* (immobilization/mortality, reproductive effects, and biochemical biomarkers) and *Danio rerio* (survival, hatchability, abnormalities, and biochemical biomarkers). The bacteria tested showed different sensitivities to the antibiotics, being *E. coli* the most sensitive (~ 50 % of growth inhibition after all the antibiotic treatments). *L. minor* photosynthetic pigments and proline content were significantly decreased by all the antibiotic treatments. To *D. magna*, no significant alterations were observed in biochemical biomarkers after 48 h, however after 10 days all the antibiotic treatments caused oxidative stress and neurotoxicity. Similar biochemical results, as well as abnormalities (e.g., body curvatures), were observed in *D. rerio* embryos for all antibiotic treatments. The environmental concentrations tested showed be marginally or slightly toxic, affecting individually and sub-individually the organisms tested. Thus, society and the scientific community should be vigilant to environmental contamination and ecosystem sustainability concerns, associated with SMX and TRIM levels in surface waters.

## Are freshwater planarians affected by light pollution?

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Artificial light at night (ALAN) is a pervasive stressor in many urbanised aquatic ecosystems, that disrupts the natural light patterns and consequently may lead to dysregulation of biological rhythms. The recent advent of light-emitting diodes (LEDs), that have replaced the traditional street sodium lamps, has increased the presence of blue light spectrum in the environment which can further increase the ecological effects associated with ALAN. There is an alarming lack of information on how ALAN may affect the physiology, behaviour, and fitness of benthic aquatic invertebrates.

Here we present results from different laboratory assays aiming to evaluate the responses of the freshwater planarian *Girardia tigrina* to ALAN. Using LED lights, *G. tigrina* was exposed to three ALAN intensities during the dark phase of the photoperiod (0- control; 1 and 10 lux). Reproduction was observed over 21 days of exposure while biochemical responses and head regeneration after decapitation were evaluated after ten days of exposure. Acute effects on locomotion and respiration rates were assessed overnight (under the different ALAN conditions).

ALAN exposure caused neurotoxicity, alterations in energy metabolism, and oxidative stress in planarians. Also, head regeneration was delayed and reproduction was decreased suggesting significant effects of ALAN on planarians' fitness.

Results show that planarians can be adversely affected by ALAN levels commonly observed in many urban aquatic ecosystems, and call for more research concerning the effects of ALAN on aquatic benthic communities and ecosystems functioning, and also on possible mitigation measures to reduce light pollution.

## Effects of runoff and leaf litter origin on a freshwater shredder

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Urban stream ecosystems are exposed to many perturbations, including pollution of water and food resources. Stormwater runoff transports a high variety of pollutants from urban areas to nearby stream ecosystems, including oils and heavy metals, products derived from tire wear, pet droppings, etc. Leaf litter from urban vegetation is an additional source of pollutants that can affect river food webs. Although the ecotoxicological effects of pollutants typically found in urban stormwater have been well characterized, there is still little information on the effects of bulk stormwater and of its potential interactions with leaf litter pollution. To better understand the effects of different sources and forms of urban pollutants on stream macroinvertebrates, we conducted a 28 day microcosm experiment in which we exposed individuals of *Echinogammarus berilloni* to a crossed test with two factors: water type (River water, Rainwater and urban Runoff) and origin of organic matter (Natural and Urban), totaling six treatments. We measured *Echinogammarus* survival, growth and food consumption rates. Mortality was higher in Urban Runoff and Rainwater than in River water, whereas was not affected by litter types. Water pollution reduced litter consumption, whereas individual growth was lowest in Rainwater treatments. Our results point to a weak detrimental effect of urban stormwater on stream invertebrate performance.



# Effects of the herbicide bentazone on the structure of plankton, benthic diatoms and macroinvertebrate communities representative of Mediterranean coastal wetlands: a mesocosm experiment

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Pesticide pollution poses one of the most important threats for the ecological status of Mediterranean coastal wetland ecosystems. The effects of the herbicide bentazone on aquatic communities representative of Mediterranean wetland ecosystems were investigated using mesocosms representing two ecological scenarios, one with rooted macrophytes and one without macrophytes. The herbicide was applied weekly for four weeks in 24 mesocosms at concentrations of 25, 250, and 2500 µg/L. The impact of bentazone on various aquatic groups, including benthic diatoms, phytoplankton, zooplankton, and aquatic macroinvertebrates, was examined before the first application, after the last application and sixty days after the last application. The results indicate that benthic diatoms were the most affected group in terms of community structure (NOEC 25 µg/L), with some populations being affected by all tested concentrations (NOEC < 25 µg/L). Zooplankton was the other group most affected, with large effects observed on the last sampling day (NOEC 25 µg/L), mainly driven by the replacement of large filter feeders, such as cladocerans and calanoids, by small filter feeders, like rotifera. No effects on the aquatic community structure were observed in the other two groups. The results indicate bottom-up effects caused by the herbicide, altering the structure of primary producers' community, which in turn, indirectly affects zooplankton community. This study suggests that measured bentazone concentrations in Mediterranean coastal wetlands of the Iberian Peninsula could result in direct and indirect effects on the structure of aquatic communities in the long term.

# How Bisphenol A in food impacts zooplankton performance under warming-fluctuating temperature?

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Bisphenol-A (BPA) is a synthetic organic compound commonly used as additive of plastics and recognized as an emerging contaminant of growing environmental concern. Transfer of BPA in aquatic organisms and BPA interaction with climate change drivers, as warming and fluctuating temperature, are still gaps of knowledge. We aim to study the influence of BPA on growth rate and reproduction traits of *Daphnia magna*, using a gradient of BPA concentration similar to that found in aquatic ecosystems, to examine (i) direct vs. trophic transfer effect of BPA, through dissolved BPA vs. BPA assimilated by algal food (*Scenedesmus* sp.), and (ii) the combined effects of BPA in algal food and two temperature regimes, i.e. warming constant and warming fluctuating. BPA, mainly that transferred to *D. magna* through food, exerted significant adverse effects on growth rate, moulting ability, and reproduction of *D. magna*, even at low BPA-concentrations, although the magnitude of effects augmented at high BPA-concentrations. The interaction BPA×temperature showed a slight positive effect of BPA on *D. magna* growth rate under warming constant temperature but inhibitory under warming fluctuating temperature, at lower BPA-concentrations. Notably, we found a BPA threshold below which *D. magna* growth rate became temperature-dependent, with significantly higher growth under warming constant and warming fluctuating temperatures. Our study underlines the key role of temperature to predict how toxins (such as BPA) may affect *Daphnia* performance via trophic transfer in aquatic food webs, with broader implications in ecosystem functioning and structure in a changing world.

# New data on the characterization of trace elements in the main freshwater inflows to the Vigo estuary (NW Spain) - Assessment of the environmental status and identification of chemical signatures

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The rivers and streams flowing into the Vigo Ria comprise both quasi-pristine and anthropogenically-impacted water bodies. During a total of 22 sampling campaigns, here we have monitored the freshwater inputs to this coastal bay over a two-year period, covering different precipitations and flow conditions. A complete chemical dataset (Al, V, Cr, Mn, Fe, Co, Ni, Cu, Zn, Ga, As, Rb, Sr, Cd, In, Sb, Cs, Ba, Pb, Ti, Zr, Nb, Ta, Mo, Hf, W, Re, Th, U, and the REEs) were analyzed both in the dissolved (<0.2 µm) and particulate phase following trace-metal clean procedures during sampling, handling and analysis of samples. This study includes new data for several elements with typical ultra-trace concentrations and represents one of the first studies reporting their values in freshwaters of the Iberian Peninsula. The main objectives of this study are to (i) assess the environmental conditions of these water systems (e.g. Cu, Cd, Pb, Ni, Ag) including several 'technology-critical elements' (e.g. Pt, Nb, Ta, Ga, In); and to (ii) define aqueous processes influencing hydrologic basins and the chemical components of terrestrial fluxes to the ocean (e.g. REEs, Nb/Ta, Zr/Hf, Th/U). Results will be discussed in terms of identifying contamination point sources and chemical fluxes of trace elements to the Vigo estuary, and the characterization of the chemical signatures of the different catchment areas.

# Integrating Novel Technologies for Early Warning of cyanobacterial blooms

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Freshwater scarcity and pollution pose significant challenges to water resource management, especially considering the evolving legislative standards for drinking water quality. Cyanobacterial blooms, driven by eutrophication, emerge as a critical public health and ecological concern due to the production of cyanotoxins. These toxins can entail a risk to numerous species, including humans, restricting water usage (recreational and/or consumption purposes).

A multidisciplinary research group has been created to address this problem by developing collaborative projects to transform water quality management through the integration of innovative technologies. Methodologies for early detection and mitigation of cyanobacterial blooms are being implemented and tested to design more reliable and precise alternatives to the traditional management, focusing on the development of an Unmanned Surface Vehicle (USV) and a Decision Support System (DSS) under the modelling capabilities by Digital Twins. Different waterbodies in Spain, with varying cyanobacterial abundance, were analyzed combining: technical expertise for developing and fine-tuning of multiparametric probes for in situ monitoring and physical-chemical and biological analysis conducted in laboratory. Calibration efforts have been conducted to obtain a reliable algorithm relating phycocyanin probes signals with phycocyanin concentrations, extracted in the laboratory, and thus cyanobacterial biomass. Our results prove the high robustness and coherence between these methodologies in different waterbodies and cyanobacteria assemblages for the generation of an early warning system. This is a reliable, low cost and fast system that would allow a future scenario of forecasting cyanobacterial occurrence at any waterbody.

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Keywords: cyanotoxins, water management, Unmanned surface vehicles, metabarcoding

## Is caffeine a toxic compound to standard aquatic species?

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Caffeine (CAF) is an emerging organic contaminant already detected in diverse environmental aquatic matrices, exhibiting potential bioactivity towards non-target organisms. This study aims to evaluate the effects of CAF in standard aquatic species from different trophic levels, namely: *Raphidocelis subcapitata* (growth inhibition), *Lemna minor* (growth inhibition, pigment content, and biochemical biomarkers), *Chironomus riparius* (lethal effects, emergence, and biochemical biomarkers) and *Daphnia magna* (feeding rate, life history parameters and biochemical biomarkers). Acute toxicity was observed for *R. subcapitata* (EC<sub>50</sub> < 100 mg/L), *L. minor* (EC<sub>50</sub> = 520.7 mg/L), and for feeding rate of *D. magna* (EC<sub>50</sub> = 323 mg/L). Biochemical responses showed that CAF induces oxidative stress in *L. minor*. Additionally, a decrease in carotenoids and an increase in chlorophyll contents were observed at higher concentrations tested (1000 mg/L). *D. magna* feeding assay showed that CAF induces oxidative stress (up to 498 mg/L) and neurotoxicity. In *D. magna* sub-chronic assay, CAF also caused oxidative stress, with an increase in antioxidant and biotransformation enzyme activities (up to 7.5 mg/L), and neurotoxicity effects up to 60 mg/L. *C. riparius* emergence assay showed an increase in mortality, a decrease in growth rate, and biochemical disturbances with the increase of CAF concentrations, up to 61.5 mg/L. The presented results showed that CAF residues induce significant impacts in standard aquatic species. This study underscores the importance of prioritizing the monitoring of CAF, as it has been demonstrated to pose a significant hazard to various biota organisms, alerting both the scientific community and society at large.

# Effects of Artificial Light at Night (ALAN) on the freshwater invertebrate *Daphnia magna*

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Artificial light at night (ALAN) is an anthropogenic emergent pollutant that disrupts circadian rhythm patterns and behavior in many organisms. Melatonin, which conveys information about light/dark periods also plays a crucial role in mediating immune and antioxidant responses, can be suppressed by ALAN and especially blue light spectrum emissions. Our work aimed to overcome some of the knowledge gaps concerning physiological responses to ALAN in the cladoceran *Daphnia magna*. In laboratory settings, *D. magna* was exposed to two ecologically relevant levels of ALAN (1 and 10 lux) during nighttime emitted from white LED lamps. To experimentally validate the mediatory role of melatonin, experiments were conducted with extra treatments, where exogenous melatonin was added to the medium to counteract the expected ALAN-induced reduction in endogenous melatonin. Effects of ALAN on *D. magna* biological fitness were evaluated using fitness-related traits and biochemical endpoints related with physiological condition. Results show that ALAN reduces the reproductive output and *D. magna* longevity. It also induces higher respiration rates and feeding inhibition. In treatments with exogenous melatonin, these effects were ameliorated further suggesting its key physiological role linking ALAN to biological fitness. Biochemical responses related to oxidative stress, energy metabolism and general stress responses are being analyzed and will be discussed to complement the effects observed at the organismal level. This work adds data and a physiological perspective to the growing body of research done on the ecological effects of light pollution in urban aquatic ecosystems.

# OSI2

## Ecosystem Services



# Linking river hydrology to coastal fisheries: quantifying ecological and commercial benefits of Ebro River

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Over the last years, the Ebro River faces scrutiny and debate whenever heavy rainfall events lead to significant water discharge into the sea. These events are often perceived by the public as a missed opportunity to capture water for human benefit, oversimplifying the intricate relationship among flow dynamics, ecosystem functioning, ecosystem services, and societal demands. However, river discharge pulses represent a crucial yet often overlooked component of river-coastal coupled ecosystem dynamics. The aim of this study is to determine the relationship between meteorological forcing (e.g. rainfall, storms, temperature) and fluctuations in Ebro River nutrients concentration and flow on fish landings -- a critical ecosystem service in coastal waters surrounding the Ebro Delta. We gathered data from the five main ports differentiating by fishing method (small-scale, towed and encircling nets), over a span of 22 years that encompassed seasonal and interannual fluctuations in river flows. Our analysis revealed clear evidence of the influence of riverine nutrient enrichment and meteorological processes on total fish landings, independently of the fishing gear considered. This study illustrates that river outflow benefits to fish landings can be significant in oligotrophic seas such as the Mediterranean, while meteorological phenomena like wind and storms enhance the productivity of surface layers through mixing and upwelling. We contend that it is crucial to include river discharge in coastal management plans, recognizing its key role not only in the sustainability and resilience of coastal ecosystems but also in the provisioning of key ecosystem services.



# Agro-forestry ecosystem services in integrated water resource management

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Integrated landscape management should consider a complex interplay of processes and biodiversity patterns across hydrological units to warrant water security. This approach should secure the provision of goods and services for human welfare, while reducing vulnerabilities to global change risks. In this context, the design of an efficient configuration of land uses and best practices is paramount. The objective should be to maximize biodiversity and regulating ecosystem services (ES) without a large detriment to the economic income of farmers or forestry operations.

The main objective of this study is to assess the interactions between different ecosystem services in agro-forestry systems at the catchment scale, focusing on the region of Cantabria as a case study. In some cases, these ES show synergistic interactions, while in others they negatively affect the performance of one another (antagonistic interaction). These interactions will require appropriate analytical tools to guide integrated landscape management to resolve conflicts of interest. For that purpose, we first test and quantify the relations between ES pairs by performing Spearman's rank correlations. Then, we delineate landscape hotspots combining the intensity of each ES with the overlap of other ES generated by the same landscape unit. Finally, we set the criteria to select the priority land use(s) in each case.

The final outcome will be a proposal for a multi-criteria decision-making framework to select the areas of the basin with the greatest multifunctionality in order to promote more sustainable land management, favouring synergies between the ecosystem services considered, and maximising their delivery.

# Biodiversity and the supply of multiple ecosystem services in river basins of northwest Portugal

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Aquatic biodiversity has a high social, economic, cultural and aesthetic value and is crucial for maintaining multiple ecosystem services. Assessing the status and trends of aquatic biodiversity and ecosystem services is key for the development of strategies for a sustainable use of natural resources and to implement measures to deal with major threats. We conducted a comprehensive survey of freshwater biodiversity and ecosystem services in three watersheds in NW Portugal (Cávado, Minho and Lima River basins). This allowed us to map biodiversity, identify vulnerable species and habitats, and diagnose environmental factors shaping current species distribution. In parallel, we assessed six key ecosystem services, namely carbon storage, habitat quality, crop production, nutrient retention, water yield, and sediment retention, within the river basins in northwest Portugal. We employed the InVEST model to map the state of these ecosystem services in 2018, along with three projected land cover scenarios for 2050 indicative of i) business-as-usual, ii) farmland return, and iii) afforestation. Our findings indicated that the business-as-usual scenario could lead to detrimental impacts on carbon storage, habitat quality, and sediment retention. In contrast, the farmland return scenario showed less drastic decreases in habitat quality and sediment retention compared to the business-as-usual scenario. The afforestation emerged as the most favorable scenario, with a 13.6% increase in carbon storage and a 1.3% improvement in habitat quality. Our findings show that assessing the status and trends of aquatic biodiversity and of considering multiple ecosystem services is critical for developing strategies for adequate watershed management.

# Climate change effects on water security across European river networks

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Climate change increases the risk of droughts and floods, and threatens water provisioning and water uses, what produces important consequences for human societies and nature. Managing multiple climate change risks for enhancing water security needs a strong conceptual framework and a comprehensive assessment of main drivers and indicators for specific water security dimensions. In this sense, this study proposes an integrated catchment modelling approach in which key hydrological processes and the provision of specific Ecosystem Services are modelled. The modelled ES account for 3 dimensions of water security: water provisioning, flood regulation and drought regulation using present and future scenarios in six river networks across a large environmental gradient in Europe. Specific approaches were adopted in alignment with the most suitable spatial and functional scales (i.e., landscape units: river channels, hillslopes and floodplains) in which ES may be provided. As expected, the spatial and temporal distribution of the provision of these ES highly depended on the hydrological response of the catchment, mainly driven by spatial (topography, soil type, land use, hydrogeology...) and climate variables (precipitation, temperature, evapotranspiration...). The methodology applied in this study allows quantifying water availability provided by rivers and the potential of the catchment to store water over time and to reduce the impact of hydrological extremes. Undoubtedly, these analytical tools will contribute to evaluate catchment resilience to climate change, supporting the decision-making process, and the design of adequate adaptation actions to climate change.

**OSI3**

**Urban Aquatic Ecosystems**



# Functional and structural responses of freshwater primary producers submitted to urban runoff contaminants

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Urban runoff transports water from roofs, roads, and soils into streams, carrying a variety of contaminants which can impact ecosystem biodiversity and functioning. These effects might be particularly important during rainfall episodes and following prolonged dry periods. To test the effects of these contaminants on freshwater primary producers, we collected urban first flush samples during a rainfall event and analysed the functional and structural responses on algal cultures and freshwater biofilms. We therefore conducted two acute exposure experiments using different concentrations of six typical contaminants found in urban runoff: Diuron, Benzyl butyl phthalate (BBP), Bisphenol A (BPA), Diclofenac, a mixture of 16 Polycyclic aromatic hydrocarbons (16PAHs) and copper. We assessed the impact of acute concentrations of these contaminants, both individually and in combination, on the planktonic alga (*Raphidocelis subcapitata*) and freshwater biofilm communities. We observed that planktonic alga showed higher sensibility compared to biofilm communities, which rapidly recovered after the exposure of contaminants. While some contaminants such as Diuron, 16PAHs and Copper had a strong effect, either individually or in combination, others did not produce substantial effects. *R. subcapitata* exhibited the highest sensitivity to Diuron, with the lowest observed EC50 value at 24.22 µg/L. Our results suggest that rainfall episodes may produce acute but transient impacts on the planktonic organisms, with less significant impacts on freshwater biofilms.

## Nature Based Solutions to protect aquatic ecosystems in urban areas

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Water stress in cities is expected to increase in the following years due to climate and global changes, jeopardising natural aquatic ecosystems due to rising water abstraction, pollution, droughts and floods. In this context, cities need to deploy solutions to increase water availability, and decentralised systems to on-site treating non-conventional sources (e.g., rainwater, run-off, greywater) for safe reuse or groundwater infiltration emerge as promising alternatives.

The NINFA EU-project (<https://ninfa-project.eu/>) is developing and testing diverse Nature-Based Solutions (NBS) to increase water availability while protecting groundwater from pollutant infiltration, including microplastics, heavy metals, and hydrocarbons (present in run-off), and contaminants of emerging concern and antibiotic resistance genes (present in domestic wastewater). Among them, a modular green tile acting as a sustainable drainage system, is being optimised in terms of substrate materials and plant species to increase pollutants retention ability, also evaluating its potential connection to advanced oxidation processes to reduce up to 95% the infiltration of hydrocarbons and microplastics in groundwater. In parallel, a Constructed Wetland coupled to a bio-electrochemical system, has been implemented at pilot scale in Valladolid (Spain) in the framework of the Urban Green Up EU-project (<https://www.urbangreenup.eu/>). This technology has been demonstrated for 2 years, efficiently removing organic matter from urban wastewater, captured from the sewer. Coupled to a proper disinfection system, this technology can operate as a decentralised urban system that allows safe water reuse for plant irrigation, street cleaning or groundwater infiltration.

## Naturalized Urban Ponds biodiversity drivers

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Urban parks offer shelter from the hostility of the city to both citizens and organisms, and those that have a pond can further be a climatic refuge in summer. The overall objective of the project “Contribution of Naturalized Urban Ponds to the environmental sustainability of cities (NATURBPOND)” is to assess how naturalised urban ponds (NUP) contribute to the achievement of environmental goals associated with the transition towards more sustainable cities. One goal is to contribute to the protection and restoration of biodiversity and ecosystems. We hypothesise that NUPs are valuable ecosystems that support biodiversity at different spatial scales and contribute significantly to human well-being. To assess this, we measured biodiversity in a variety of NUPs in Barcelona, following an environmental gradient from north to south, taking into account the proximity of other natural ecosystems and their isolation or aggregation in the urban landscape. We focused on algae, macrophytes, macroinvertebrates and fish.

The richness of organisms is uneven: macroinvertebrates are poor, but algae - especially diatoms - are important. Main factor determining biodiversity and community structure is the presence or absence of fish. It determines the condition and appearance of the ponds: when fish are present, the ponds tend to have abundant phytoplankton, without macroalgae and macroinvertebrates. In the absence of these, there is a greater presence of macroalgae and macrophytes - often Characeae - and a clear appearance of the water. Fish management can make a significant contribution to having NUPs that host more biodiversity and more complex communities.

# Dependence of Fish Kill Events on the Thermal Mixing Events in Deep Eutrophic Subtropical Urban Sandpit Lakes (Uruguay)

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In the urban landscape of Ciudad de la Costa, Uruguay, lie over twenty small artificial lakes, remnants of sand extraction initiated several decades ago. Much like other urban lakes globally, they have faced significant anthropogenic pressure and eutrophication is a common condition. Some previous publications referred to them as shallow-polymictic. Massive fish kills have occurred recurrently, which generates strong local socio-environmental crises. We intensified monitoring since the winter of 2016, identifying a monomictic regime in some lakes. Meanwhile, a multi-lake synchronized anoxia event in 2017 gave us clues that this pattern could be shared. This work aimed to understand relevant processes beyond intra and inter-lake water quality variability and fish kill causation. We used complementary monitoring strategies including high-frequency meteorological data and three custom-designed automated monitoring buoys with sensors at different depths (temperature, dissolved oxygen, pH). After almost four years the monitored lakes have shown monomictic regime with a prolonged stratification period or only a few annual turnover events. Vertical temperature differences vanish in April (autumn) each year. Hypolimnetic anoxic conditions prevailed since late winter, and the frequency of epilimnetic anoxia and fish kills was highest in late summer following the seasonal thermocline extinction. All the registered fish kills were linked to vertical turnover processes. The combination of conditions (such as water column Schmidt stability and wind intensity) for which mixes occur was analyzed. The new knowledge together with real-time monitoring allowed us to anticipate the most extreme fish kill event, granting social validation of the data-driven environmental management.



## Results of a nature-based solution system to reduce urban runoff pollution

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In urban areas, the washout of structures and surfaces during precipitation events results in the runoff transport of soil particles and pollutants that can reach freshwater ecosystems, impacting aquatic biodiversity and ecosystem functions. Moreover, in the context of climate change, the frequency of extreme events such as intense storms are anticipated to increase. Some initiatives, such as the establishment of separate sewer systems for wastewater and rainfall runoff water, or the planification of nature-based solutions have been implemented to treat these waters. Our study assesses a nature-based solution for rainfall runoff treatment in Viladecans (Barcelona, Spain), formed by three ponds where water from individual rainfall-runoff sewer system in an urbanized area with roads and industries arrives. Two of these ponds have green filters with diverse vegetation to treat runoff water before it arrives at the pond. In contrast, the third pond lacks a green filter. The aim of our work is studying the quality of rainfall runoff water and the effectiveness of these filters to reduce urban runoff-derived contaminants, and the potential effects on some ecological processes. We are sampling across the three ponds before, during, and after rainfall events, and analysing nutrients, organic contaminants, metals, dissolved organic matter characteristics, organic matter decomposition, and CO<sub>2</sub> emissions. Preliminary results show relevant concentrations of certain metals, nutrients, and dissolved organic carbon in the urban runoff, with the green filter effectively reducing the concentration of most contaminants (e. g. Al and Mn). This research offers insights urban runoff treatment and sustainable water management.

# Can naturalization of urban ponds support the mitigation of greenhouse gas emissions from cities?

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Naturalization of urban areas is considered as a strategy to address the ecological transition to achieve more sustainable cities and approach the objective of net-zero emissions stated in international agreements such as the Agenda 2030 or the European Green Deal. As part of this strategy, the naturalization of urban ponds has the potential to be used as an action to reduce the carbon footprint of cities and mitigate greenhouse gas (GHG) emissions. Here, we evaluated how naturalization could affect the concentrations and fluxes of GHG in urban ponds of the city of Barcelona. With this aim, we measured surface water GHG concentrations in 41 artificial urban ponds spread throughout the city in summer 2023, of which 28 were naturalized (NA) ponds and 13 non-naturalized (NN) ponds. Results revealed that most of the urban ponds were CO<sub>2</sub> and CH<sub>4</sub> supersaturated relative to the atmosphere. NA ponds showed higher CH<sub>4</sub> concentration than NN ponds, while no significant differences in CO<sub>2</sub> were found between pond types. In contrast, N<sub>2</sub>O concentration was significantly higher in NN ponds. Therefore, while naturalization could increase CH<sub>4</sub> emissions from urban ponds, these emissions could be at least partially offset by the reduction in N<sub>2</sub>O emissions, considering that N<sub>2</sub>O has a higher (~ 10 times) global warming potential than CH<sub>4</sub>. Consequently, naturalization of urban ponds can be considered as a potential mitigation measure to achieve the target of net-zero emissions in cities.

# Pharmaceuticals in urban streams of Coimbra, Portugal: a concern for the health of aquatic communities

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The consumption of pharmaceuticals all over the world have been increasing in the last decades and is commonly related to the longer life expectancy. Meanwhile, their presence in the freshwaters, mainly in urban areas, can cause negative effects on its ecological quality, being a concern to the health of aquatic communities. This study evaluated the concentration of pharmaceuticals in urban streams of Coimbra, Portugal, as well as its possible correlation with their ecological quality. Water samples of 16 urban streams (US) were collected in Autumn 2022, as well as samples of benthic macroinvertebrates. Solid phase extraction (SPE) followed by UHPLC-ToF-MS were used to the quali-quantitative analysis of pharmaceuticals in the water. The most recurrent pharmaceuticals in the streams were carbamazepine (anticonvulsant), irbesartan and losartan (antihypertensive), and caffeine (stimulant). The highest concentrations were detected for caffeine (10431 ng/L – US Arregaça), irbesartan (1690 ng/L – US Escola Agrária) and ampicillin (206 ng/L – US Coimbra-B). A Principal Component Analysis (PC1 explaining 62.5%) highlights that one stream in the centre of the city, US Arregaça, is particularly affected by pharmaceuticals. The concentration of carbamazepine was significantly correlated with the ecological quality ratios of the official Portuguese Index for Invertebrates (IPtI) ( $R^2 = 0.24$ ;  $p$ -value = 0.05), indicating that pharmaceuticals can affect the aquatic communities and the integrity of freshwater ecosystems which can also result in additional adverse effects over human health. This study shows that should be defined preventive and mitigation measures against their spread in aquatic ecosystems in highly urbanized areas.

# Impacts of urbanization on microbial community structure and functioning in stream sediments

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Urbanization has become a major driver of change in natural ecosystems worldwide. In stream ecosystems, urbanization often leads to a degraded physical habitat with frequent influx of organic and inorganic pollutants. The receiving biological communities, such as the sediment microbiome, are often shaped by urbanization with potential implications for ecosystem functioning. Notably, studying the impacts of urbanization on stream ecosystems poses significant challenges due to regional variations in climate and urban areas. Our study analyses the impacts of urbanization on microbial community structure and stream metabolism across 47 streams in seven European countries. For each sampling site, climate, land-use, and population density were characterized, as well as water and sediment properties (e.g., pH, conductivity, temperature, nutrients). In addition, sediment cores were collected, metabolic rates (i.e., gross primary productivity and ecosystem respiration) were measured, and the microbial community of these sediments analysed by metabarcoding (i.e., bacteria, fungi, algae, and protists). Our findings reveal that urbanization negatively affects both primary productivity and respiration, with urban rivers exhibiting sediments rich in heavy metals and poor in total carbon. Sediments displaying high primary productivity also showed increased green algae abundance and reduced diatom and predator abundance, such as ciliates and amoebae. These results underscore the impacts of urbanization on river ecosystems and emphasise the need for responsible management of urban rivers, particularly in the face of increasing urban migration. Our study contributes critical insights into how urbanization alters ecosystem function and highlights the importance of preserving streams in rapidly urbanizing landscapes.

# Artificial Light at Night and aquatic insects: life-history responses under multistressor scenarios

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Light pollution, resulting from artificial light at night (ALAN), affects the physiology, behaviour and ecology of organisms living in urban environments and has been suggested as a major driver of insect decline. Besides ALAN, urban aquatic ecosystems are also exposed to several other stressors namely pesticide exposure, increased temperatures, and salinisation. Effects of ALAN on aquatic invertebrates are seldomly evaluated and mostly in isolation, i.e., without considering multi-stressor scenarios that are relevant for urban ecosystems. Here we report data from laboratory assays aiming to evaluate physiological and life-history responses of *Chironomus riparius* to environmentally relevant levels of ALAN (1 and 10 lux during nighttime). Exposure to ALAN significantly induced oxidative stress and damage in larvae after a 10-day exposure. Moreover, exposure to ALAN also altered haemoglobin levels and reduced the time to emergence of female imagoes with possible consequences for reproduction. Exposure to ALAN also resulted in a decrease in the upper thermal tolerance of *C. riparius* larvae and additive effects on fitness-related traits were observed in combined exposures to ALAN and salinity. Our results indicate that ALAN can mediate the responses of aquatic insects to other stressors that are relevant for urban aquatic ecosystems. Ongoing work is also evaluating whether exposure to ALAN alters *C. riparius* sensitivity to pesticide exposure. These findings add important data on physiological and life-history responses of insects to ALAN and are discussed considering the pervasiveness of this stressor and its potential fitness impacts and ecological consequences for aquatic invertebrates in urban environments.

OSI4

**Biodiversity and Conservation of  
Aquatic Ecosystems**



# Exploring the threatened aquatic insect biodiversity of the Canary Islands

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The Canary Islands are a biodiversity hotspot, including particularly high levels of endemism related to a combination of geological, geographical, and climatological factors that promote speciation. However, most conservation and research efforts have been directed towards marine and terrestrial biodiversity, neglecting freshwater habitats. This lack of knowledge is worrying because freshwater biodiversity is globally disappearing at alarming rates and the streams in the Canary Islands are subjected to very strong hydrological stress due to climate change and water abstraction. The only studies on the aquatic insects that inhabit the small streams of the islands date back to the 1980's and 1990's. To help filling this gap of knowledge and guide future conservation efforts, the BIOACUANA and CONACAN projects explore the communities of aquatic insects in the streams of Tenerife, La Palma and La Gomera. In this communication we will show basic and updated information on the taxonomic and genetic diversity of aquatic insects in the Canarian archipelago. From 2022 (autumn) and 2023 (spring and autumn), we sampled aquatic insects (larvae and adults) seasonally from 38 undisturbed stream reaches, both outside and inside of protected areas. Preliminary results suggest no significant differences between sampling seasons but strong differences between islands, with certain taxa being significantly associated with each of the islands. In this talk we will discuss our findings within the context of the global freshwater biodiversity crisis, in which small islands could play a vital role as a refugia for endemic and undiscovered species.

# Promoting integrative research on Mediterranean climate regions through a comparative assessment with temperate ecosystems

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Mediterranean climate regions (Med-regions) are globally uncommon, yet they represent biodiversity hotspots, and contribute vital ecosystem resources to human society. Med-regions worldwide face potential threats due to global change factors outlined in the World Scientists' Warning to Humanity manifesto. However, their adaptive response to natural climate variability can buffer human-mediated impacts through a high resilience capacity. In this talk, we first show some preliminary results of a comparative assessment between Mediterranean and temperate ecosystems in the Mediterranean Basin and Europe to explore the resilience capacity of Med-regions in front of major global changes factors: eutrophication, land use change, biological invasions, and climate warming. We also explore three main types of integrative studies at different spatial scales, from local to global, which would enable and facilitate conservation of Mediterranean ecosystems worldwide: (i) integrative research across ecosystems within Med-regions; (ii) integrative research across Med-regions; and (iii) integrative research across political boundaries. Despite conservation efforts in terrestrial, freshwater and marine ecosystems, there is limited research promoting integrative research in Med-region ecosystems. Recognizing the similar resilience capacity and adaptive mechanisms of organisms in this climate, we advocate for integrative research in Med-regions that allow to promote an interdisciplinary approach with common concepts and metrics, enhancing our understanding of the general mechanisms underlying Med-region ecosystems. We believe this talk can foster future research synergies to identify the tipping points across ecosystems and aid in determining desired future conditions in Med-regions worldwide.



# Zooplankton from littoral wetlands of the Valencian region: how do these ecosystems have changed in the last decades?

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Along the coastline of the Valencian region there are important littoral wetlands, which are the remains of old wetlands that were almost continuously distributed covering the main alluvial plains. Since ancient times, men have drained many wetlands, but in the Mediterranean coast, this drainage has been even more accentuated caused by urban pressures. Some of these wetlands, due to their size or location, have persisted, and nowadays, most of them are under protection. They include a great diversity of aquatic microhabitats (marshes, fens, springs, canals, lagoons...), which harbour a great diversity of organisms, including zooplankton, an important constituent of aquatic trophic webs. We aimed to characterise zooplankton community in several coastal wetlands and compare recent data with those obtained 30 years ago. Zooplankton and water samples were taken at different sites of six selected wetlands (Cabanès, Marjal dels Moros, Albufera, Marjal Pego-Oliva, Santa Pola, and El Hondo). Zooplankton individuals were identified and quantified, together with physicochemical parameters, nutrients and pigments. Zooplankton diversity at each wetland was estimated, and the relationship between physicochemical parameters and zooplankton was explored by multivariate analyses. Cladocerans were more frequently found in freshwaters, while copepods were more common in brackish waters. Conductivity, dissolved oxygen and nutrients were the main factors explaining zooplankton community structure. Compared with data of the 1990's, an increase in species richness has been observed, including several exotic species such as *Apocyclops panamensis*, *Kurzia* sp. or *Brachionus havanaensis*.

# Successful reintroduction of the European pond turtle (*Emys orbicularis*) with a small number of founders: results from a 20-years small-scale experiment

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Mediterranean climate regions (Med-regions) are globally uncommon, yet they represent biodiversity hotspots, and contribute vital ecosystem resources to human society. Med-regions worldwide face potential threats due to global change factors outlined in the World Scientists' Warning to Humanity manifesto. However, their adaptive response to natural climate variability can buffer human-mediated impacts through a high resilience capacity. In this talk, we first show some preliminary results of a comparative assessment between Mediterranean and temperate ecosystems in the Mediterranean Basin and Europe to explore the resilience capacity of Med-regions in front of major global changes factors: eutrophication, land use change, biological invasions, and climate warming. We also explore three main types of integrative studies at different spatial scales, from local to global, which would enable and facilitate conservation of Mediterranean ecosystems worldwide: (i) integrative research across ecosystems within Med-regions; (ii) integrative research across Med-regions; and (iii) integrative research across political boundaries. Despite conservation efforts in terrestrial, freshwater and marine ecosystems, there is limited research promoting integrative research in Med-region ecosystems. Recognizing the similar resilience capacity and adaptive mechanisms of organisms in this climate, we advocate for integrative research in Med-regions that allow to promote an interdisciplinary approach with common concepts and metrics, enhancing our understanding of the general mechanisms underlying Med-region ecosystems. We believe this talk can foster future research synergies to identify the tipping points across ecosystems and aid in determining desired future conditions in Med-regions worldwide.

# Integrating obligatory species interactions and artificial barriers into conservation planning under climate change: the case of freshwater mussels and their fish hosts

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Species will likely shift their distributions to adapt to climate change, potentially leading to extirpations from currently suitable habitats and occupation of new ones. Freshwater ecosystems are highly fragmented by anthropogenic obstacles (e.g., dams and other in-stream obstacles), limiting the adaptive capacity of freshwater biodiversity. Hence, effective conservation planning must account for the dynamic impacts of climate change and species ability to cope with it. To address this need, we developed a framework that incorporates climate change and physical barriers into conservation strategies for species with obligatory interactions. Given that freshwater mussels (Bivalvia: Unionida) are dependent on fish hosts for larval development and dispersal, we used Marxan to prioritise areas for their joint conservation in the Iberian Peninsula as a case study. We tested two connectivity scenarios: i) assuming unlimited dispersal ability and ii) dispersal constrained by artificial barriers. Additionally, we identified key translocation sites for species unable to occupy future suitable habitat due to the impact of barriers. Accounting for the effects of climate change on species distributions allowed the identification of long-term conservation areas and integrating the location of barriers allowed the identification of priority areas that are more likely to be colonised in the future following climatic shifts. This resulted in an additional loss of conservation features (~5-7%) compared to solutions without dispersal constraints. Between 173 and 357 artificial barriers (~1.6-3.3%) will potentially block species dispersal to irreplaceable areas. Where removal of artificial barriers is unfeasible, translocations emerge as a viable strategy to compensate for dispersal limitations.

# Challenges ahead for freshwater ecosystems in the Colombian Guiana Shield

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The Guiana Shield contains multiple freshwater ecosystems which support a high species diversity. These are hosted by a stable landscape resulting from infrequent large-scale natural disturbances. The Colombian part of the Guiana Shield is one of the best preserved of this ecoregion, because of the yet low human density and reduced impacts. However, the existence of aggressive activities in nearby areas is a warning for the future of the Colombian Guiana Shield. Although still relatively pristine, this region is experiencing high growth in farming and plantation of palm trees, which contributes to forest loss. Other threats include infrastructure development, small-scale mining and water contamination. Growing environmental impacts and transformations ahead require preserving these systems as the necessary step to expand our knowledge on their diversity and functions. Conservation of the biological diversity brings an opportunity for initiatives leading to a respectful development, which could also be of application for other areas in the Guiana Shield.

# Exploring the Role of Artificial Ponds in Sustaining Odonata Biodiversity in the Pre-Pyrenees

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Mountain ponds play a crucial ecological role, housing cold stenotherm species highly vulnerable to global change impacts observed in mountainous regions. Stressors such as rising temperatures, altered precipitation patterns, and pollution threaten these habitats and the communities. To mitigate natural water body loss, artificial ponds are proposed as viable options. These artificial sites could augment regional water networks and support regional aquatic biodiversity. In our study conducted in the Pre-Pyrenees of NE Catalonia, we assessed the macroinvertebrate biodiversity supported by temporary (n=6) and permanent (n=6) natural and artificial (n=10) systems, which were also all permanent water bodies. Generally, artificial ponds showed significantly lower chlorophyll-a content compared to permanent and temporary natural ponds, indicating lower algal biomass. Odonata, a flagship macroinvertebrate group, were absent in temporary ponds, probably due to increasingly short hydroperiods caused by prolonged drought conditions in Catalonia during the studied period. However, permanent ponds (n=5) harbored 31 specimens, and artificial ponds (n=5) housed 16 specimens, across 7 genera: *Aeshna* (4), *Libellula* (24), *Anax* (3), *Pyrrhosoma* (4), *Sympetrum* (9), *Cordulegaster* (1) and *Enallagma* (1). Specimens of *Aeshna cyanea*, *Anax imperator*, *Libellula depressa* and *Pyrrhosoma nymphula* were found in both natural and artificial ponds, highlighting the importance of artificial ponds as additional aquatic habitats in mountainous regions. With our work, we aim to contribute to pond management recommendations, especially in understudied mountainous areas, proposing artificial ponds as a potential mitigation strategy for climate change induced biodiversity losses in those aquatic ecosystems.

# Evaluating the sampling effort in mediterranean rivers: When to stop sampling to get an accurate picture of aquatic macroinvertebrate communities?

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Studies on community and conservation ecology are based mainly in species inventories. However, in the inventory of biological diversity it is usually impossible to register all the species present in the studied area and the results obtained are conditioned by the sampling effort carried out. This work focuses on assessing the changes in knowledge about the aquatic macroinvertebrate community as the sampling effort increases. The study was carried out in 4 rivers from the same typology at the Segura basin, following a commonly used standardized protocol, applied to evaluate the ecological condition in Spanish water bodies. Using taxa accumulation curves and the theoretical adjustment of the Clench function, we found that 25 samples were sufficient to obtain a good representation of the macroinvertebrate families present at each sampling site (78-88% of the estimated total), but it was not enough when we considered communities at species level (completeness values below 70%). The minimum number of samples required to get a completeness value over 70% of the macroinvertebrate community was clearly dependent on the environmental heterogeneity of the water bodies. In water bodies with greater environmental heterogeneity a strong sampling effort is required to obtain inventories with similar levels of completeness and which, therefore, would be comparable. These results can be used to improve the definition of the established protocols destined to compare the taxa richness in aquatic ecosystems, and to be more efficient when assessing the ecological condition of water bodies following the guidelines of the Water Framework Directive.

# Additive effects arise from multiple stressor impacts on biodiversity facets of Atlantic stream macroinvertebrates

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Freshwaters are among the most threatened ecosystems globally due to multiple stressors, that can coincide in time and space, resulting from land-use intensification or hydroclimatic changes. There is a need to assess multiple stressor effects in real-world ecosystems, because current knowledge is still limited to experimental approaches that are prone to decrease biological and abiotic variability. Using a survey of 50 streams across the North of Portugal, we assessed biodiversity responses of macroinvertebrate communities to multiple stressors that include gradients of nutrient enrichment, flow reduction, riparian vegetation structure, thermal stress and depletion of dissolved oxygen. We analyzed these effects on two taxonomic metrics (richness and Hill Shannon), and two trait-based diversity metric (functional richness and dispersion). Our results showed that multiple stressors had additive effects on all diversity metrics. Nutrient enrichment was the most important stressor, followed by depletion of dissolved oxygen and thermal stress. Taxonomic richness, Hill Shannon and functional richness responded similarly. However, functional dispersion was mainly driven by changes in flow velocity and thermal stress. Functional trait composition shifted along a major gradient of stress associated with nutrient enrichment and depletion of dissolved oxygen. Overall, our findings reinforce the need to consider complementary facets of biodiversity to disclosure assembly processes in response to multiple stressors. Our study also suggests that stressors' interactions may be less frequent in real-world streams. Overall, our results provide new insights that can be useful to better assess and manage river ecosystems in a context of global change.

# Unraveling the effects of human impacts and natural variability in food webs from temporary rivers' disconnected pools

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Temporary rivers are common in Mediterranean climate regions due to high seasonality of rainfall patterns. Their scientific interest has increased recently, but there are still knowledge gaps to be addressed. The disconnected pool phase, for example, has received less interest than the flowing or the dry phases, although these pools are key for biodiversity conservation and provide refugia for aquatic taxa during dry periods. Moreover, there is little understanding of how human impacts influence their ecology and impose significant challenges for managers because communities can be naturally variable, as a result of intrinsic pool characteristics (e.g. size, oxygen) and the increase of trophic interactions therein. The aim of this study was to describe food webs in disconnected pools of temporary rivers, and to analyse how they change along human impacts. Fifteen pools were sampled for macroinvertebrates and environmental variables, covering a broad impact gradient in the Spanish Mediterranean region. The Mediterranean Reference Criteria (MRC) was computed to obtain a gradient of human impacts for each pool. Macroinvertebrates were counted and identified at genus level, and their trophic information was obtained from published trait databases (e.g., Freshwater Information Platform). Our results showed a decrease in the number of predators (i.e., lower mean trophic level) and simplified trophic interactions (e.g., lower number of interactions) with increasing human impact. These changes became more relevant after certain thresholds. Identifying tipping points can be critical to ensure that these pools can effectively contribute to biodiversity recovery with flow resumption.



# Revision of blackflies (Diptera: Simuliidae) from Cidacos river through La Rioja (Central-Northern Spain)

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This communication, presents an in-depth review of the sampling efforts carried out by University of Salamanca researchers in 13 places of Cidacos river (Central-Northern Spain) between April and August of 1997.

This thorough review has revealed the presence of 12 species of the *Simulium* genus, which belongs to 4 subgenera (*Boophthora*, *Eusimulium*, *Simulium*, and *Wilhelmia*). In addition, *Simulium* (*Boophthora*) *erythrocephalum* (De Geer, 1776), *Simulium* (*Eusimulium*) *petricolum* (Rivosecchi, 1963), and *Simulium* (*Simulium*) *trifasciatum* Curtis, 1839 arise as first records from the single-provincial autonomous region of La Rioja, and *Simulium* (*Simulium*) *variegatum* Meigen, 1818 from the province of Soria, part of the autonomous region of Castilla y León. New data regarding the elevation and water temperature are provided for 10 species, as well as the range of these two factors have been updated for 3 other species too. It has also been detected that 9 of the species require blood intake from birds and mammals, being important not only due to the discomfort caused by their bites, but also because of the symptoms to which they give rise in wild and domestic animals, and humans, and the pathogenic agents that they are potentially capable of transmitting.

Consequently, the findings from this study shed light on the diversity and bioecology of simuliids in Cidacos river and contribute significantly to increase the knowledge of blackfly species present in the surveyed region. This includes insights into their geographical distribution, altitude and temperature ranges, and an assessment of their potential veterinary and health significance.

# Land-use, protected areas, and the fate of recently described aquatic insects in Europe

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Occupying only 3% of the Earth's surface, freshwater ecosystems contain almost 10% of the world's animal species. Despite their importance for biodiversity, freshwater ecosystems show the highest rates of biodiversity loss and habitat fragmentation due, in part, to the rise of anthropogenic land-uses in the last century. Freshwater biodiversity is mainly made up of insects, a class in which many species still remain to be described (80-95%). Their high biodiversity and key functional roles in freshwater ecosystems urge us to be aware of their conservation status, and find ways to protect both the new species and the areas in which they are being described. Our main goal was to assess the potential fate of recently described aquatic insects in Europe based on the overlapping with protected areas and estimates of land-use changes. We used a database containing recently described species of aquatic insects in Europe between 2000-2020; each one was assigned to a sub-basin according to the HydroBASINS boundaries. Land-use changes were obtained from the SSP scenarios up to 2050, and sub-basin hotspots of recently described species were identified. Around 70% of the recently described species of aquatic insects were found outside of protected areas, most of them in sub-basins that will experience significant changes in land-use. Our results emphasise the importance of finding ways to reverse land-use trends in particular areas outside of protected areas to ensure the conservation of recently described species and those that still remain unknown.

**SSI**

**Advances in eDNA for the study  
of freshwater ecosystems: from  
technical to ecological aspects**



# Transition from morphology to molecular biomonitoring in rivers: what is missing?

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Biological assessment of European rivers, using phytobenthos is presently based on morphology. The diatom community structure is determined through light microscopy study. During the last decade, the metabarcoding approach based on environmental DNA (eDNA) has confirmed its potential for biomonitoring. The transition from morphology to eDNA biomonitoring would allow faster, cheaper, and more accurate results, especially if taxonomy free. Nevertheless, according to the Water Framework Directive (WFD), the ecological status of a site is calculated as the deviation of the site to the reference condition which is type specific. The aim of this study was to validate typologies previously defined by the morphological approach, adapt, or even establish new typologies for rivers after analysis of the Exact Sequence Variants (ESVs) for each river type. Four river types in northern, central Portugal were evaluated using fifty-four reference sites. Both ESVs assigned to taxonomy (130 ESVs) and not assigned (3648 ESVs) were able to differentiate between the types as the morphological data (260 taxa). The ESVs assigned to taxonomy showed the best separation among the four types, thus validating the use of the presently established typology for biomonitoring with eDNA. The free taxonomy approach using ESVs would free biomonitoring from taxonomy. Yet, it could have the drawbacks of preventing the establishment of links between ecological and taxonomical knowledge as well as with ecosystem functioning, which should be avoided by integrative approaches.

# eDNA metabarcoding for assessing fish biodiversity in coastal wetlands from the Ebro delta

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biomonitoring methods. Despite the number of studies in eDNA has increased during the past decade, only a few have been conducted in transitional waters such as coastal wetlands. Coastal wetlands are very dynamic ecosystems that typically hold a high biodiversity and conservation value, but they are also highly threatened. The Ebro delta represents one of the largest and most vulnerable coastal wetlands in the Mediterranean. A high number of endangered fish species are present in many of these habitats, including lagoons and salt marshes. Fish communities are monitored every year since 2008 for conservation purposes using traditional methods. Here we present the first study on fish communities from the Ebro delta using eDNA. In 2022, we sampled 11 coastal wetlands combining conventional fish sampling methods with eDNA. For eDNA we filtered water using a Smith-Root sampler with self-preserving filters and processed using an Illumina MiSeq V3 kit at 10.5 pM. Fish were captured using fyke nets, identified and measured. A total of 22 taxa were detected with eDNA, while only 13 with fyke nets. For endangered species, *Anguilla anguilla* was detected with eDNA at 8 sites, while only at 3 using nets, while *Aphanius iberus* was only detected with nets. Invasive species detection was maximized using eDNA. This suggests that despite the limitations of eDNA, it can be a promising and complementary tool for assessing fish biodiversity and the impact of invasive species in coastal lagoons.

# DNA metabarcoding revealed that local drying and spatiotemporal connectivity affect metapopulations and metacommunities differently depending on their dispersal

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Fragmentation by droughts disrupts the hydrological connectivity of rivers shaping biodiversity patterns of different dispersal groups. Isolated river reaches present a limited gene flow between populations that might lead to local extinctions of weak dispersers. Contrastingly, isolation can foster genetic differentiation. Temporary rivers (TRs) are exceptional model systems as they experience strong changes in spatiotemporal connectivity and harbor species that cover a wide range of dispersal modes. In this study, we used metabarcoding to assess the importance of local hydrological conditions and network connectivity on metapopulation and metacommunity assembly in TRs. We sampled seven mediterranean TRs in the Sant Llorenç del Munt i l'Obac natural park (Barcelona), during spring and summer. We identify macroinvertebrates using traditional methods, which are later used for bulk sample metabarcoding to obtain Exact Sequence Variants (ESVs; calculated by counting the unique genetic sequences detected in a sample). We calculated ESVs richness for all taxa, aerial dispersers and aquatic dispersers and tested the effect of local and river network drying using Generalized Linear Mixed Models and Generalized Dissimilarity Modeling. Our preliminary results show that local drying and spatiotemporal connectivity affect metapopulations and metacommunities differently depending on their dispersal. ESVs richness increased with spatiotemporal connectivity for both aquatic and aerial dispersers, but the response was much stronger in the former. Also, we found significant differentiation of populations between locations for strictly aquatic species (e.g. *Physella acuta*), whereas strong fliers (e.g. *Anax imperator*) presented no discernable spatial pattern of intraspecific variability.

# Implementation of DNA Metabarcoding for the Biomonitoring of Spanish Iberian Rivers Using Macroinvertebrates as Bioindicators: Advantages, Limitations and Roadmap.

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Molecular techniques, such as DNA metabarcoding, are presented as alternatives potentially able to overcome certain limitations of current river biomonitoring systems based on morphological identification, such as long processing times and low taxonomic resolution. In contrast, DNA metabarcoding offers a promising solution, allowing species-level identification from mixed DNA samples, advantages in scalability, automation potential and the ability to use different sample types, including bulk samples and environmental DNA (eDNA). This study applies DNA metabarcoding techniques to the biomonitoring of wadeable rivers in the northwestern Iberian Peninsula, using benthic macroinvertebrates as bioindicators. The research aims to evaluate the detection capacity and performance of these techniques in identifying macroinvertebrates from two different sample types: homogenised bulk samples and water eDNA samples. The study compares the results of these techniques with established official methods based on morphological identification. The results show a significant relationship between macroinvertebrate composition at the IBMWP taxa level (mainly family level) and the method used, with less dissimilarity between bulk and morphological samples than between eDNA samples and either of the other two methods. The study identified several sources of false positives and negatives, as well as other possible causes explaining the differences between the results obtained by molecular techniques and morphological identification. Finally, the following steps were proposed for an effective implementation of the use of DNA metabarcoding in the biomonitoring of rivers in the Iberian Peninsula.

# The drawdown of the Enobieta Reservoir had contrasting effects on procaryotes and eukaryote microbes

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The life expectancy of dams is limited, and when they become obsolete or useless, they must be dismantled to restore the natural environment and recover river connectivity. In the case of large dams, there is still little information on short-term environmental effects of decommissioning. We analysed the influence of the drawdown of the Enobieta dam on riverine microbial communities in the Artikutza valley (Navarre). We performed a BACI study in which we sampled 4 impact sites downstream from the dam, as well as 4 control sites not affected by the dam, before and after dam decommissioning. We took biofilm samples from the riverbed, extracted DNA, and sequenced it using Next Generation Sequencing techniques. On previous analysis, different physicochemical variables were sampled. Before drawdown, the bacterial communities downstream from the dam were very different from control sites, and those differences decreased downstream. Below the dam there was a lack of cyanobacteria, whereas bacteria from Chloroflexi, and Nitrospirota phyla and a few Proteobacteria groups were specially abundant, probably as a consequence of high metal and ammonium concentrations. Those phyla include ammonium oxidizers, anoxic photosynthetic bacteria, and iron oxidizers. After drawdown, communities from the impact sites beneath became more similar to control ones. On the other hand, fungal showed a similar trend before drawdown, but differences between control and impact sites increased after. Our results, thus, point towards contrasting response of procaryotes and eukaryote microbes to dam decommissioning.



# Microbial composition and richness in Atlantic rivers: The role of climate and landscape

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Freshwater biodiversity is under significant threat from the combined effects of human-induced climate and land-use changes. Microbes, occupying almost all habitats in riverine ecosystems, regulate energy fluxes and are prominently positioned at the nexus of global sustainability and climate change. However, considerable uncertainty persists regarding how different taxonomic groups respond to large-scale factors in fluvial systems. Most studies on freshwater ecology focus on a limited number of clades, and few simultaneously include both prokaryotic and eukaryotic communities on a large spatial scale. Here, we analyse River Microbial Communities' (RMC) richness and composition patterns on a latitudinal gradient across six European Atlantic catchments. We investigated RMC response to climatic, hydrological, geological, and land use patterns based on high-throughput environmental DNA sequencing. We have observed a strong regional footprint that determines both the composition and diversity of RMC. Biogeographical patterns can be explained by: i) variables operating at the regional scale (climate and geology), ii) variables operating at the catchment scale (topographical) and iii) land use variables. These results delineate distinctions between communities in warmer catchments with marked dry seasons (Portuguese and French catchments) compared to those with cooler temperatures and more consistent year-round rainfall (Northern Spain and Ireland-UK). The results of our study highlight varying sensitivities among bacteria, fungi, protists, and algae, developing potential indicators of global change, such as taxa resistant to temperature increase and water scarcity, as well as potential indicators of land use changes across the taxonomical groups analysed.

# Metabarcoding mediterranean aquatic macroinvertebrates of the iberian peninsula. Implications for biodiversity and biomonitoring assessments

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Within the last twenty years DNA metabarcoding became a commonly adopted molecular approach to assess biodiversity of freshwater ecosystems and identify the taxonomic composition of biological samples for biomonitoring purposes, among other utilities. DNA based methods are proposed as an alternative of morpho-based identification methods although important challenges must previously be tackled. In this study, benthic macroinvertebrates were collected at 15 sites across the Ebro Basin (Spain) as part of an official national stream monitoring program (Directive 2000/60/EC). The mitochondrial gene for cytochrome c oxidase subunit I (COI) was used as DNA Barcode. Operational Taxonomic Units (OTUs) were taxonomically assigned to major groups of Iberian freshwater macroinvertebrates combining two public reference libraries of COI barcodes (BOLD and NCBI). Taxonomic coverage, taxonomic composition metrics (IBMWP), Ecological Quality Rates (EQR), Ecological Status, and alfa and beta diversity metrics obtained from traditional and molecular approaches were compared. Although with some limitations regarding the families recovered by the molecular approach, we proved that the macroinvertebrate community structure as well as the ecological assessment obtained from metabarcoding on Mediterranean freshwater macroinvertebrates of the Iberian Peninsula is to some extent comparable to that obtained using the standard morphotaxonomic approach. Similarly to other studies, a significant 46,3% of sampling sites were equally assigned by both approaches. We also observed similar beta diversity metrics and significantly correlated alfa metrics. However, further research is needed to improve its use as a routine biomonitoring and biodiversity assessments tool, mainly regarding the incompleteness of Mediterranean reference libraries for the Iberian case.

# DNA metabarcoding and morphological species identification reveal comparable diversity patterns for diatom communities in the high mountain mires

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Diatoms are commonly used as bioindicators because they are sensitive to environmental changes, such as pH gradients, conductivity, or the eutrophication of the habitat. The identification of diatom species has been conducted through morphological features which demands very specialized knowledge and training. To circumvent this obstacle, DNA metabarcoding offers an alternative method for the generation of high-throughput data. However, current literature shows discrepancies between metabarcoding gene markers and morphological identification. We tested whether morphological methods and metabarcoding approaches provide similar results concerning diatom community structure and composition. We studied the diatom community of 28 high mountain mires in the Pyrenees by identifying morphological species and sequencing the Cytochrome c oxidase I (COI) and 18S markers in the samples. We assessed whether morphological and molecular approaches agree in describing diatom alpha and beta diversity patterns. Our results show a significant correlation in the beta diversity patterns of diatom communities between morphological and molecular marker genes whereas our findings indicate that molecular data poorly represent alpha diversity patterns (richness and community structure) compared to morphological identification. The diatom community was predominantly influenced by pH, a pattern observed in both morphological and molecular data. Our results also show that precipitation and temperature are important factors in structuring diatom communities. In conclusion, COI and 18S are good markers to represent the beta diversity of the diatom community and the environmental variables that shape it in high mountain mires.

# Fish metacommunities in a Mediterranean fragmented river network: insights from molecular techniques

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Metacommunity ecology offers valuable insights into the spatiotemporal dynamics of species assemblages, especially in fragmented river networks subject to the dynamics of drying and flowing periods. Molecular techniques, such as eDNA, are powerful tools for studying fish metacommunities in these environments. However, few studies have integrated both traditional and molecular approaches for studying fish metacommunities. We combined conventional fish sampling methods with eDNA metabarcoding to assess the fish metacommunity dynamics in a Mediterranean river network affected by fragmentation. We sampled 18 sites with different hydrological and anthropogenic impacts, including small barriers and dams. For eDNA metabarcoding we filtered water on site using a Smith-Root sampler with self-preserving filters. The fish were captured by electrofishing, identified, and measured. To assess the relative importance of local versus regional processes on fish metacommunities, we used river network and topography distances, fragmentation distances, together with environmental factors.

Our findings indicate a clear effect of fragmentation on fish metacommunities in the basin, using both eDNA and electrofishing. Furthermore, eDNA metabarcoding detected fish at sampling sites where electrofishing was not possible owing to lack of accessibility, providing a more comprehensive view of metacommunity composition. Despite the limitations of eDNA, this study emphasizes the value of eDNA metabarcoding for understanding the drivers of fish metacommunity dynamics. It also provides valuable insights into the use of non-invasive methods to study fish communities in vulnerable habitats in drying river networks and its application as a complementary method for fish conservation management.

# Taxonomy and biogeography of neotropical chironomids using DNA barcoding

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Chironomidae is a globally distributed family of aquatic insects and one of the most abundant and diverse group in freshwater ecosystems. This family has been understudied, particularly in the neotropics, due to taxonomic difficulties arising from their complex morphology, and the challenge of linking their four life stages together. Molecular tools, such as barcoding, can help overcome these difficulties by supporting morphological analyses. Our main aim was to contribute to the knowledge of this family in the Neotropics by combining morphological and molecular tools. We analyzed the DNA barcodes of two genera within the subfamily Diamesinae found in Colombia, Ecuador and Peru. We focused on this subfamily because of its specific environmental requirements. It inhabits waters with low temperatures, typically in mountainous areas, and has a wide distribution (including the Andes), which is interesting from a biogeographical perspective. Species delimitation was performed using three different methods: ABGD, ASAP, bPTP and a maximum likelihood tree (RAXML) and a morphological matrix to distinguish operational taxonomic units (OTUs). Two OTUs were found for *Limaya* using all methods, one in Ecuador and one in Peru. *Paraheptagyia* had approximately 7 OTUs, with a clear separation between those found in Peru and those found in Ecuador and Colombia. The Huancabamba depression, located between Peru and Ecuador, creates geographic isolation for low-temperature waters, which likely limits gene flow. The combination of both tools provided a better understanding of diversity and biogeography studies and are also the foundation for future metabarcoding, community, and bioindication projects.

# Integrative Assessment of River Biodiversity and Water Quality Through Aquatic Vegetation and eDNA Analysis

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This research employs an integrated approach to explore riverine biodiversity and water quality in the Mero and Barcés rivers, focusing on the crucial role of aquatic vegetation as ecological sentinels. By integrating data from physico-chemical sampling, hydromorphological characterization, environmental DNA (eDNA) analysis, and taxonomic assessments of aquatic life forms, the study provides a comprehensive perspective on aquatic ecosystem health. Findings emphasize the significant ecological indicator role of aquatic vegetation and micro-phytobenthic communities, which further highlight differences in water quality along the rivers, underscoring the importance of tailored management strategies to protect and enhance these aquatic ecosystems. We have integrated the eDNA derived biodiversity data with physical and chemical parameters to develop a holistic view of the ecological health of the studied rivers. By correlating the presence of specific taxa identified through eDNA with water quality indicators (like nutrient levels or contaminants detected through chemical analysis), we can derive insights into the impacts of environmental conditions on biodiversity. Implementing these steps will help in effectively synthesizing the diverse information sources to form a cohesive understanding of the ecosystem being studied, enhancing the quality and reliability of the conclusions drawn. Challenges related to the application of advanced techniques such as eDNA analysis are acknowledged, especially the need for specialized equipment and expertise, which impacts the accessibility and implementation of these methods in local contexts. This integrative study advances our understanding of how combined datasets can inform ecological health assessments and supports the development of informed management practices based on robust scientific data.

# Flying between mountains: The effect of wind on high mountain caddisflies diversity and vulnerability

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High Mountain Streams (HMS) are highly vulnerable ecosystems that are significantly affected by the loss of biodiversity due to climate change (CC). Trichoptera, a diverse and vulnerable taxon within these ecosystems, includes numerous endemic species at a high risk of extinction. The vulnerability of Trichoptera depends on biological and ecological traits, as well as external factors that influence dispersal across landscapes. Although wind dispersal could potentially contribute to population resilience through mass effects, this remains speculative. Our aim was to analyse the spatial and seasonal variability of Trichoptera diversity in 17 HMS along the southern Pyrenees, covering a wide altitude (1128 - 2405 msl) and longitude (2°19'3.6'E - 0°8'19.4W), and relate it to species vulnerability and wind patterns. NOAA wind data for three months before each sampling date provided wind speed and direction. We sampled Trichoptera larvae in spring and summer at three sites per stream, categorized by proximity to the source, after the first tributary, and below the tree line. We assessed genetic, taxonomic and functional diversity using traditional taxonomy, metabarcoding and trait data collection. Species vulnerability to CC was determined by integrating biogeographic, ecological, and biological traits with species haplotype richness. Results showed correlations between west-to-east sampling distribution and wind patterns, site vulnerability, and genetic, taxonomic, and functional diversity in spring, with a less pronounced impact in summer. Overall, the results suggest that the effects of CC on HMS in the Pyrenees depend on species vulnerability and the potential of wind to disperse individuals and prevent local extinctions.

# The use of DNA to estimate fish biomass in streams: Universal vs species-specific primers

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Environmental DNA (eDNA) has the potential to become an effective, cost-efficient, and non-invasive method to estimate fish biomass or density. Still, the quantitative aspect of this approach needs a solid ground to verify the relationship between fish DNA concentration and biomass in the field. Here, we investigated this relationship collecting eDNA from mountain streams located in Picos de Europa National Park and compared it to electrofishing survey data. The effectiveness of eDNA to infer fish biomass or density was assessed using different markers: 18S rRNA and the cytochrome c oxidase I (COI) by metabarcoding, as well as a specific *Salmo trutta* within the locus COI via quantitative PCR (qPCR). Salmonidae reads and DNA copies per litter significantly correlated to fish biomass and density, suggesting that eDNA metabarcoding and qPCR produce useful estimations. Using universal markers (18S region) and a specific *S. trutta* showed similar results and relationships remained significant. Furthermore, while the COI marker offered a greater taxonomic specificity, it exhibited lower fish detectability than a universal eukaryotic locus (18S). Further analysis exploring the relationship between eDNA reads and fish age classes (fry, juvenile and adult) has revealed that juvenile biomass is the group that more accurately explains this biomass-eDNA relationship in these mountain streams. Our results suggest that metabarcoding reads can reveal biomass information and provide accurate quantitative information on fish communities in small mountain streams. The application of this methodology is of interest for conserving and managing Salmonidae fish populations in mountain rivers.



# Assessing stream diatom communities in the Canary islands: insights and preliminary results

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The Macaronesian region is characterized by a high proportion of watercourses in volcanic substrates with fluctuating flow regimes, including both temporary and perennial streams with a great diversity of habitats. Very little is known about the diversity and distribution of freshwater diatoms from inland watercourses of Canary Islands, in spite of the relevance of the barrancos (=ravines) in these islands. In the context of the Parques Nacionales-funded project CONACAN about freshwater biodiversity and conservation (<https://conacuana.es>), diatom communities are being investigated by means of microscopy and DNA metabarcoding. Here we will present our preliminary results of the benthic diatom assemblages from the three Canary Islands sampled (La Gomera, Tenerife and La Palma, these are the highest altitude islands and where the most important ravines of the archipelago are found). A high diatom species richness was found in permanent streams such as Arure waterfall (La Gomera) and in Barranco Del Río (Tenerife). Yet no clear pattern was observed in temporary streams. The widespread *Amphora pediculus* and *Planothidium frequentissimum* were rather common in almost all sampling points. Endemisms such as *Nitzschia tenerifa* and *Nitzschia macaronesica* were also found albeit restricted to only few localities of the three islands. Several species isolations have been performed, including some of the endemisms, to obtain their reference DNA sequence. Our preliminary results shown background information on the taxonomic and genetic diversity of the diatom communities, holding potential for the application and posterior adaptation of the WFD within the unique hydrogeological and environmental context of the Canary Islands.

# Sampling methods matter, even with eDNA: biomonitoring freshwater macroinvertebrates using metabarcoding

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Biomonitoring macroinvertebrates is essential for establishing the integrity of freshwater ecosystems. In recent years, environmental DNA (eDNA) emerged as an effective biodiversity assessment method. However, the influence of eDNA sampling methods on the detection of freshwater macroinvertebrates has not been extensively explored. Few studies have compared the results of molecular techniques across different environmental matrices (e.g., biofilm, water) with traditional methods when characterizing aquatic macroinvertebrate communities. Here, we collected eDNA (water & biofilm) and kicknet samples, across thirteen different streams in Picos de Europa National Park. Macroinvertebrate taxa were identified using a COI barcode on water, biofilm and the ethanol used for the kicknet sample preservation and compared to the taxonomical identification of the traditional sorting of kicknet samples. The resulting morphological and molecular inventories were compared by assessing richness, composition, and biological indices. Further, we explored how taxonomic resolution, taxon abundance, or sclerotized levels of different taxa affect taxa detection between sample types. Fewer taxa per site (family richness) were obtained from water and biofilm samples compared to morphotaxonomy. Though, finer taxonomic resolution was registered with eDNA metabarcoding for some taxa. We observed low overlap in community composition between types of samples, especially for common bioindicator groups such as Trichoptera and Diptera. Preliminary findings indicate that eDNA sampling matrices strongly influence the detection of macroinvertebrate taxa and should be carefully considered to reduce detection probability biases. Finally, we discuss the knowledge gaps required to use metabarcoding as a reliable assessment tool in freshwater ecosystems.

# Diatom metabarcoding for WFD assessment and beyond

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Here I will present our experiences in using diatom metabarcoding (based on partial *rbcl* marker) to fulfil the requirements of the Water Framework Directive (WFD) to assess the ecological status of rivers but also going beyond this to see what insights the current metabarcoding technology can give into the ecology and geographical distributions of benthic diatoms.

Our work on Mediterranean rivers in Catalonia (Pérez-Burillo et al. 2020, STOTEN 727: 138445) commissioned by the Catalan Water Agency showed very good agreement between morphology- (LM) and DNA-based (metabarcoding) assessments of ecological status. Where there were important disagreements affecting the WFD class assignment, in several cases we were able to identify the cause, including that the discrepancies can be due to an unacknowledged bias in the LM approach (e.g. not detecting *Fistulifera* species).

In addition, *rbcl*-based metabarcoding is revealing ecological and geographical differentiation among some genetic variants of some morphospecies such as *Achnantheidium minutissimum* and *Fistulifera saprophila* (Pérez-Burillo et al. 2021, STOTEN 798: 149029) as well as global distribution patterns that could be relevant for a better understanding of diatom evolution and biogeography.

# Molecular based methods using benthic diatoms for enhanced river biomonitoring efficiency. Applicability in southern European rivers

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The adoption of molecular techniques in river biomonitoring, especially the use of benthic diatoms, has emerged as a reliable and cost-effective alternative for traditional morpho-taxonomic methods. The exceptional attributes of diatoms, such as high sensitivity, easy eDNA retrieval from periphyton, and availability of DNA barcode databases, make them excellent organisms for the application of molecular methods. Thus, the biomonitoring of river benthic diatoms using a molecular approach has yielded promising results along European rivers, promoting the use of molecular techniques. Our work has focused on the assessment of rivers spanning different biogeographic regions in the southern part of Europe, including Iberian Peninsula, Azores islands and Cyprus. While the results obtained show a good ecological status correlation with traditional approaches, several discrepancies still exist. Some of the discrepancies are related to the lack of DNA barcodes in the databases (especially those that are difficult to isolate and cultivate) and incorrect taxonomic assignments due to taxonomic disparities among cryptic species. However, one of the main problems is related to the calculation of ecological indices and the values or molecular relative abundance. These values are biased by the *rbcl* gene copy number, which is also related to the number of chloroplasts. As a solution, a biovolume based correction factor was applied and validated with samples from Basque water agency (URA) to achieve a better correlation. Although this approach improves the correlation between methods, the establishment of molecular reference values and taxonomy-free approaches may lead to better estimations of ecological status in rivers

# SS2

## Exploring ways of Integrated land and water resource management to achieve temporary pond conservation



## Small is beautiful. The importance of disseminating the role of temporary ponds in biodiversity conservation

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Temporary shallow lakes and ponds are among the most sensitive and threatened continental aquatic ecosystems by global change (i.e. land use changes, climate change, introduction of invasive alien species). The fact that they have a small volume of water along with a cryptic period during their drying phase makes them highly sensitive and vulnerable to these disturbances. Although it has been demonstrated that these ponds harbor a high diversity, conservation measures are urgently needed due to the rapid degradation that these systems have suffered in recent decades worldwide. Therefore, there is a need to raise awareness about the important ecological role they play as biodiversity reservoirs and as supporting elements of landscape heterogeneity and, as providers of regulation ecological services (e.g. water purification, water supply, erosion and flooding control, etc.). From the I-WET research group and the ClimaRiskinPond project, we have developed various communication and dissemination activities at national/international events such as the European Researchers' Night, Science and Innovation Week, and the International Day of Women and Girls in Science, and although aimed at all audiences, we have paid special attention to youngest students, those in Primary Education, but also bringing them to our scientific laboratories at University with the support of the UAM Scientific Cultural Unit (UCCUAM). In addition, we have organized two editions of a citizen science-based pond photography contest, across the construction of an interactive web including the citizen metadata; we have designed stickers and different games, as well as friendly macroinvertebrate identification guides, and videos.

# Facing threats and conservation challenges in temporary ponds: insights from peninsular Spain

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Temporary ponds of the Mediterranean region are often designated as priority habitats for conservation though they face threats such as anthropogenic perturbations, climate change, and biological invasions. Despite their conservation value, these habitats have usually been overlooked in conservation programs in peninsular Spain, partly because their knowledge remains incomplete and scattered. We compiled a comprehensive database (ca. 6500 ponds) from bibliographic research and expert contacts, and used photointerpretation to assess pond locations and physical impacts. Ploughing impacted over 50% of pond edges and more than 20% of pond basins, suggesting that agricultural intensification was a prevalent impact. We also sampled 104 temporary ponds along a latitudinal gradient (2021/22) examining physicochemical, biotic, and abiotic variables to explore the conservation status of temporary ponds in Mediterranean Spain. Anthropogenic pollution affected 24% of the sampled ponds and was likely originated by the abovementioned agricultural activities. Surprisingly, few invasive non-native species (INNS) were found, except for *Physella acuta*. Conversely, the INNS composition in the highly connected pondscape of Doñana National Park (2021), revealed a much higher prevalence of such species, suggesting that isolation may determine the presence/absence of INNS in temporary ponds. Hence, control/eradication programs of INNS would be particularly effective at pondsca- pes where INNS are confined to isolated ponds. Meanwhile, other threats such as anthropogenic pollution and habitat degradation in agricultural areas should be targeted and counterbalanced by pond restoration projects. Our research highlights the urgent need for specific management approaches to mitigate threats and preserve temporary ponds as priority habitats.

# Long-term conservation assessment: temporary ponds of the Doñana National Park after 14 years of monitoring

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Mediterranean temporary ponds are priority for conservation under the European Union Habitats Directive. Most of the temporary ponds of the Doñana National Park (DNP) are groundwater-dependent ecosystems. They harboured a high biodiversity mainly due to the high connectivity of aquatic habitats within the pondscape. However, temporary ponds have been experiencing trends of desiccation in the last decades, mainly associated with groundwater abstractions. We have analysed the variation of the water quality, community composition of macroinvertebrates and the presence of non-native aquatic species in the DNP pondscape from 2007 to 2021. Out of the 87 sampled ponds in 2007, we detected 27 desiccated short hydroperiod ponds, while only 66 ponds retained water in 2021 (a total of 79 sampled ponds). In 2021, we detected a general increase in the electrical conductivity, in external input of nutrient concentrations, and in the number of ponds in which the aquatic invasive non-native species *Procambarus clarkii* and *Physella acuta* occurred, while *Gambusia holbrooki* and *Stenopelmus rufinasus* showed a substantial reduction due to the desiccation or shortening of the hydroperiod across the pondscape. In general, the pond network has reduced its range of hydroperiod, affecting the presence of large branchiopods typically occurring in ephemeral ponds, and has restricted the presence of those species requiring long hydroperiod aquatic habitats to artificially deepened ponds, which were built to supply water for the fauna in summer. This possible species loss, together with the arrival of non-native species, could explain a homogenization of the macroinvertebrate assemblages across the pondscape.



## **Arundo donax management manual: the Segura river basin experiences**

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The proliferation of the giant reed (*Arundo donax*), invasive alien species in the Iberian Peninsula, presents a significant and costly management challenge in rivers within semiarid climatic areas. Over the past 15 years, the Segura River Basin Authority (CHS) has been working to control this invasive species by restoring the riparian vegetation. Since 2009, following the National River Restoration Strategy created by the Water Management Administration to achieve the European requirements, different methodologies have been implemented in several areas of the Segura river basin, from temporal to permanent fluvial systems. The extended experience of more than 50 actions adapted to the particular characteristics of each area, allows the optimization of some techniques and better use of the projects resources.

Based on the CHS experience, we conclude that the current strategy to control the invasion of the giant reed has to be based on weakening its vital reserve organ (rhizome) and fostering competition for natural resources through the restoration of native ecosystems. As a result, the most effective and widely applicable techniques implemented in the Segura river basin, have been compiled in a set of guidelines to be applied in different regions, as well as the instructions to restore and maintenance of the riverbank vegetation. These guidelines also include general considerations and recommendations to maximize the efficient use of resources: selection of priority areas of intervention, the most appropriate method to implement, as well as a global costs analysis.

# Sampling temporary ponds in peninsular Spain: Insights for improved management and conservation

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Temporary ponds play a crucial role in biodiversity conservation, acting as stepping stones in connected pondscapes and harbouring singular species. However, these ponds have been degraded in the last decades facing the current threats of biological invasions, climate change and anthropogenic perturbations (e.g. water pollution, ploughing and groundwater abstractions). The ClimaRiskinPond project has analysed over 200 ponds across peninsular Spain between 2021-2024, and employ a comprehensive methodology. First, we explored the most suitable ponds for sampling and selected them in a latitudinal array across peninsular Spain after assessing their hydroperiod using orthophotos and both existing and inedited databases, it also allowed us to identify the different pressures of the ponds. We filtered the water column to sample macroinvertebrates and amphibians (1-1 l dipnetting, 1 mm mesh size), and zooplankton (50 µm mesh size), while ostracods were sampled through integrated benthic samples (140 µm mesh size) and aquatic vegetation was characterized (*in situ* or pressed) once per dipnetting. All these samples were taken according with the pond size and heterogeneity of microhabitats. We also filtered the water column to analyse the water quality, including chlorophyll-a concentration, dissolved inorganic nutrients (nitrate, nitrite, ammonium, orthophosphate) and total phosphorous concentrations. In addition, we measured pH, electrical conductivity, turbidity, dissolved oxygen concentration, temperature *in situ*, and alkalinity. These results might be applied to the Water Framework Directive through the ECLECTIC index to assess the pond ecological status. We suggest insights based on our experience to complement and simplify a multi-taxa integrated monitoring of temporary ponds.

# Long-term conservation assessment: temporary ponds of the Doñana National Park after 14 years of monitoring

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Biodiversity loss and declining ecosystem functioning is spreading at alarming rates worldwide. In this context, pondscape, including temporary ponds, have emerged as potential Nature-based Solutions that offer valuable ecosystem services and connect freshwater habitats as stepping-stone ecosystems. However, agricultural intensification and hydrological modifications threaten these ecosystems, leading to the degradation and disappearance of temporary ponds. Our study assessed ecological functioning associated to large branchiopods, considered flagship species, through a field mesocosm experiment. We used 90L tanks inoculated with a mix of zooplankton and sediment sourced from six different temporary ponds of the Doñana National Park. To assess the differential role of large branchiopods we used the species *Triops baeticus*, *Streptocephalus torvicornis* and *Cyzicus grubei*. The effect was measured on water quality, i.e. electrical conductivity, pH, and concentration of dissolved oxygen and inorganic nutrients (nitrate, nitrite, ammonium, and orthophosphate), but also on mosquito oviposition, primary production, decomposition rates, and macroinvertebrates and zooplankton assemblages. Preliminary results showed that *T. baeticus* increased turbidity, probably due to bioturbation, and concentration of chlorophyll in vivo, likely indicating an increase in the phytoplankton production through direct zooplankton predation. Conversely, *S. torvicornis* and *C. grubei* treatments showed reduced turbidity and concentration of chlorophyll, maintaining a clearwater state. Hence, water quality changed accordingly with these findings, probably associated with the role of large branchiopods in the nutrient recycling of temporary ponds. This study highlights the notable contributions of large branchiopods to ecological functioning of temporary ponds, underscoring the urgent need for conservation of temporary ponds as unique habitats.

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**Water management of  
reservoirs: the challenges  
of the future generations**



# Algorithm retrieval for the ecological state study of Albufera lagoon through the satellite PerúSAT-I

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Anthropogenic pressures and impacts can alter the functions of coastal wetlands, compromising the uses to which they can be subjected. Albufera of Valencia is a representative case of pressure on water quality with a hypertrophic state since 1970s, but with a great heterogeneity. Remote sensing as a tool for monitoring water quality has great potential as a complementary strategy, remotely, frequently, and continuously over time. PerúSAT-I stands as Peru's inaugural remote sensing satellite, delivering composite images comprised of 4 multispectral bands boasting a spatial resolution of 2.8 m, complemented by 1 panchromatic band with a spatial resolution of 0.7 m., which makes it an ideal sensor for capturing highly detailed products useful for different applications, especially in very heterogeneous waters. This study pretends to assess the potential of PerúSAT-I's multispectral images to generate maps of different variables to evaluate the ecological state of Albufera lagoon. The main objective is to develop specific algorithms to estimate water quality, using PerúSAT-I images synchronic with a Albufera in situ database. During 5 field campaigns, samples were taken, and measurements of ecological indicators were made. All possible combinations of bands were obtained, which were then correlated by fitting a linear regression with the biophysical variables from 22 water samples. The best algorithms have an R of 0.76 for chl-a, 0.75 for 2 SDD, 0.84 for TSM, 0.76 for inorganic fraction, and 0.87 for organic fraction of these suspended solids. A temporal and spatial analysis was obtained by applying these equations to Albufera lagoon images.

# Comparative water quality Assessment in Reservoir Ecosystems by means of Phytoplankton and Zooplankton Indices: A Case Study in Northwest Algeria

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In recent years, there has been significant interest and remarkable advancement in studies focusing on the water quality of aquatic ecosystems. The introduction of plankton as biological indices has enhanced our understanding of the trophic state, water quality, and functioning of limnic ecosystems. Phytoplankton and Zooplankton, for a long time, has been an overlooked yet crucial component in assessing reservoir water quality. Present study aims to compare the effectiveness of phytoplankton and zooplankton indices in assessing the trophic status and ecological potential of two reservoirs, thereby enhancing the reliability of zooplankton as a biological indicator of water quality. Plankton and water samples were collected monthly over two-year period (from November 2020 to October 2022) at the Cheliff diversion dam and the Kerrada storage reservoir. These are part of a large-scale project involving the transfer of dam water to provide drinking water for the M.A.O. corridor, including Mostaganem, Arzew, and Oran, in the northwest region of Algeria. It is important to note that the Cheliff Dam, constructed on the Cheliff River, supplies water to Kerrada Reservoir. According to the phytoplankton abundance and biomass, the Cheliff dam is classified as hyper-eutrophic, while Kerrada ranges from mesotrophic to eutrophic. This classification is proven by the zooplankton species that are related with low and moderate water quality. Moreover, zooplankton abundance exhibits significant correlations with phytoplankton abundance and ecological water quality. These findings suggest that zooplankton species can be a valuable indicator for evaluating water quality status.

# Estimation of carbon content in water bodies from hyperspectral images

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Remote sensing allows observation of water quality, in inland, coastal and oceanic waters. However, operational satellite missions are currently limited by the number of bands, which hinders the detailed detection of certain peaks in reflectance spectra that could provide better information about water components. PRISMA sensor (PRekursore IperSpettrale della Missione Applicativa) has been in orbit since 2019, providing hyperspectral images with 234 bands and 30 m spatial resolution. The aim of this study is to estimate carbon content in water bodies through three variables: DOC (dissolved organic carbon), TOC (total organic carbon), and CDOM (colored dissolved organic matter, which is part of the DOC). For this purpose, different indices and combinations of satellite bands have been studied, based on the in-situ measurement of these variables in three inland water bodies in the Valencian region (Spain) coinciding with the acquisition of PRISMA images. The results show that DOC and CDOM can be estimated with a linear relationship between the ratio of 807 nm and 439 nm bands, with a coefficient of determination  $R^2$  of 0.88 and 0.81 respectively. Meanwhile, the TOC can be estimated with a linear relationship with the ratio between 817 and 674 nm bands, with  $R^2$  of 0.94. This opens new perspectives in water quality studies, especially with the use of future hyperspectral missions being planned by the Copernicus program of the European Space Agency.

# The LIFE CYANOBLOOM project: an integrative approach to the early detection of cyanobacteria blooms

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Cyanobacteria represent a management problem that seriously concerns water stakeholders. The competitive success of cyanobacteria, together with their ability to produce cyanotoxins, may result, under certain conditions, in cyanobacterial harmful algae blooms (cyanoHABs), which can seriously compromise human health, primarily through toxin ingestion but also through dermal contact or inhalation of aerosols. Traditional monitoring methods for cyanobacteria detection, such as in-situ sampling and laboratory analysis, often fail to provide timely warnings. Alternative tools, like satellite remote sensing, genetic analysis, or probe-based pigments monitoring, require further enhancements towards early detection of cyanoHABs and toxicity assessment. CYANOBLOOM is an innovative LIFE project aimed at the early detection and management of CyanoHABs in public water supply reservoirs. CYANOBLOOM proposes a comprehensive solution, combining remote monitoring via satellite data and on-site hyperspectral field measurements (WISPstations), with genetic analysis. The project will be piloted in four water reservoirs across Spain, Sweden, and the Netherlands. The objective of the project is to improve early detection rates through specific decision-making dashboards, in close contact with local stakeholders. By integrating advanced technological and analytical methods, CYANOBLOOM seeks to enhance water management strategies, ensuring safer water supplies and mitigating the adverse effects of toxic blooms on human health and the environment. This initiative is supported by a multidisciplinary consortium of SMEs and environmental clusters from Spain, the Netherlands, Germany, and Sweden, combining expertise in water quality monitoring, genetic sequencing, and environmental management to address the complex challenges posed by CyanoHABs.



# Water resources and agriculture in southwest Portugal - Lessons to learn

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When we look at the last decade, the hydrologic cycle has presented quite a few extraordinary peaks, both of heavy rains and droughts, compared to older time series. In the same time frame, Portuguese reservoirs have presented a similar pattern of variation, from above 90% of their capacity to much lower values. Nevertheless, during the same time frame, in the southwest region of the country, the reservoirs have almost always been close to critical values, regardless of the type of hydrologic year.

In this work we will try to explain this situation looking at the agriculture developments over the last years, and the importance of water use for agriculture compared to the importance of water use to other purposes. The reservoirs we are analysing here, are almost solely used to agricultural purposes, with a few situations of urban water supply, that are not significant enough to have a significant effect on the water consumption patterns.

What is happening now in this region, threatens to expand to the rest of the southern region of Portugal, with water access problems spreading through Algarve, or the problems with water uses in Alentejo region, mainly concerning Alqueva reservoir. From this analysis and trends, we draw some conclusions that could be used as lessons learned, to help design a territorial planning more consistent with expected climate changes in the long run.

# Leveraging Remote Sensing to assess the impact of drought on water quality in the Alqueva reservoir

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Drought stems from natural cycles, while water scarcity arises from human demands. To meet the growing population's needs, water demand in Mediterranean regions for agriculture, industry, and potable water is escalating. This, combined with climate change, worsens droughts and water scarcity, making them more frequent and severe. Remote sensing (RS) technology is emerging as a practical alternative for tracking drought severity and assessing water quality, driven by its affordability and extensive spatial and temporal coverage. This study assesses the impact of drought severity on water quality at Alqueva reservoir, using RS with bio-optical algorithms. These tools allow examining how drought influences water quality indicators (WQIs): Chlorophyll-a, Total Suspended Solids (TSS), and Secchi Depth (SD), by integrating in situ data with Sentinel-2 imagery over the period 2015-2022. We apply atmospheric corrections with neural networks (C2RCC, C2X, C2X-COMPLEX) and several algorithms targeting Chlorophyll-a, TSS, and SD to satellite images for accurate WQI evaluation. In situ results indicate spatial and temporal fluctuations in WQIs - mostly notably between the sampling points Lucefécit and Montante ( $p < 0.05$ ). Results from in situ and automatic products reveal WQIs respond better to different neural networks: Chlorophyll-a has less absolute error with C2X (NRMSE= 15.03%) and SD with COMPLEX (NRMSE= 13.41%). Identifying the optimal processing technique is vital for accurately mapping and analyzing spatial-temporal water quality changes in the Alqueva reservoir, especially during droughts. By doing so, we enable stakeholders to respond quickly to crises, providing them with the necessary resources for quick and informed decision-making.

# Evolution of Surrounding Area Practices: Past, Present, and Future Impacts on Reservoir Water Quality

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Inadequate land use, compounded by the climate change effects contribute to freshwater degradation and disrupt the balances within the ecosystem. The present work aimed to evaluate temporally and spatially the effect of surrounding area, and climate change on two lentic ecosystems (Rabagão and Aguieira reservoirs). For each reservoir, were analysed the land use and soil occupation evolution, as well as the pressures reported along the last 10 years. In addition, official records of water quality parameters were collected alongside the determination of water quality following the Water Framework Directive. Rabagão shown a decrease in agricultural and forested areas, replaced by pasturelands, resulting in considerable pressure on the aquaculture sector, which holds paramount significance in this area. An increase in temperature and pH of water body were observed, however still being within the reference values for good ecological quality, according the WFD metrics. Aguieira exhibits a decrease in agriculture and weed-covered areas, coupled with an expansion of artificialized territories and forested areas. This hydrographic basin faces a wide variety of pressures, namely urban discharge and a substantial volume of water allocated for agriculture purposes. Aguieira displays an upward trend in chlorophyll a levels, temperature, and pH, indicative of eutrophication status and low water confirmed by the WFD assessment. The long-term data showed that the usage of surrounding area of the hydrographic basin is extremely important for the water quality of reservoirs, should be included in the WFD metrics, as well as in the measures to be adopted to improve water quality.

# Suspended matter retrieval from satellite hyperspectral radiometric sensor measurements

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The RESSBIO (Remote Sensing Spectroscopy for Wetlands Biodiversity) project aims to investigate and use the capabilities of hyperspectral sensors for assessing and monitoring the ecological status of lentic aquatic systems. The work presented here uses the PRISMA (PRecursore IperSpettrale della Missione Applicativa) mission to study the total suspended matter (TSM), distinguishing between particulate organic matter (POM) and inorganic matter (PIM). Seven atmospheric corrected PRISMA images (L2D) were used, and nine sampling campaigns were carried out in four Mediterranean inland water bodies, always within a  $\pm 3$ -day time window from PRISMA image acquisitions. A total of 36 integrated water samples collected between surface and Secchi disk depth were taken and georeferenced. The samples were analysed using the gravimetric method, covering a wide range of TSM (1.4-192.5 mg/L), POM (0.8-84.4 mg/L) and PIM (0.6-108.1 mg/L). For images, the remote sensing reflectance average of 3x3 pixel window centred in the sampled points were obtained. The algorithms to estimate TSM and PIM were developed using the ARTMO program, employing the leave-one-out validation method. POM was estimated as the difference between TSM and PIM values, and its accuracy was validated using all in-situ data. The algorithms achieved a normalized root mean square error value (NRMSE) of 10% for TSM and 12% for PIM, demonstrating their robustness when the NRMSE calculated with measured and estimated POM values was also 12%. These findings signify progress towards one of the secondary objectives of the project, i.e. estimating the organic matter balance in Bellús reservoir as a case study.

# Colors of “L’Albufera de València”; new environmental conditions cause the dominance of new species in the phytoplankton

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Seasonal succession of lake Albufera colors, olive green in winter when chlorophytes and diatoms dominate matching cyanobacteria, blue-green in spring with cyanobacteria dominance, that turns yellowish green in summer. At fall 2023, an accentuated, unusual and extended change in environmental conditions of the lake, high water temperatures and strong winds, caused a progressive change of its color arriving to reddish brown (magenta), consistent with the appearance and dominance of a new algae with a very thin and long acicular morphology ( 0.6 x 40 µm), that had not been previously recorded.

This new magenta color triggers a great interest and excitement for people related with the lake, although its origin is explained by a new phytoplankton composition.

The study of these new species was carried out by optical, electron and high-resolution confocal microscopy, analysis of their pigments and spectrofluorometry. These observations confirm that the new blooming morphotype represents more than 50% of the biovolume.

Metagenomics for cyanobacteria using V3-V4 regions of RNAI6S marker shows that the majority of sequences corresponded to Leptolyngbyaceae, which agrees with the microscopic observations, but do not correspond to the new morphotype.

Metagenomics for eukaryotic algae is also necessary, analyzing V8-V9 regions of RNAI8S marker, where appear sequences corresponding to the class Trebouxiophyceae, confirming its relation with the new species, as far as databases allow us.

Phytoplankton size fractioning by sequential filtration would allow us to obtain enriched suspensions of the new algae, to advance its phenotypic and molecular taxonomy.

**SS4**

**Research and management  
challenges on non-perennial  
rivers**



# Effects of flow intermittence on ecosystem metabolism in drying river networks

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Virtually all river networks include drying sections, hence, drying river networks (DRNs) could play an important role in the global carbon cycle as bioreactors of terrestrial and aquatic carbon resources, governed by River Ecosystem Metabolism (REM). However, due to the complexity of the processes driving REM, it is difficult to accurately upscale riverine carbon fluxes from in situ measurements to entire river networks, limiting large-scale estimations of total carbon processed. In this study we aimed to: 1) determine the main drivers of daily REM, measured as gross primary production (GPP) and ecosystem respiration (ER), in perennial and non-perennial streams; 2) upscale daily REM from reach-scale to entire DRNs and 3) assess the effect of drying in REM dynamics at the reach and network scale. Within the DRYvER project, we estimated REM in 120 reaches distributed in six DRNs spanning a large environmental gradient across Europe during three hydrological seasons (pre-dry, dry and post-rewetting). REM was generally low and skewed towards ER, making most sites heterotrophic. Daily REM rates were regulated by catchment size and agricultural land uses, which affected local factors such as light, nitrates and epilithic carbon, while drying had no or little effect. However, large differences appeared between perennial and non-perennial reaches when REM rates were converted into carbon processed by wet area. Our results suggest REM rates present similar patterns and drivers in perennial and non-perennial streams, but the amount of processed carbon is reduced by drying due to wet channel contraction and desiccation.

## Key bacterial groups maintain stream multifunctionality in response to episodic drying

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Microbial biodiversity is fundamental to maintain ecosystem functioning in seasonally variable ecosystems. However, it remains unclear how alterations in water availability caused by episodic drying compromise the ability of stream microbes to maintain multiple functions simultaneously (e.g. primary production, carbon cycling). Using data from 32 streams, we investigated how the phenology of annual drying influences stream sediment microbial biodiversity and their capacity to sustain multifunctionality. Our results showed that stream multifunctionality and most bacteria did not respond to changes in drying phenology. Only two bacterial groups, the drying-resistant Sphingobacteriia and the drying-sensitive Acidobacteria\_Gp7, exhibited positive associations with multifunctionality, whereas bacterial diversity showed a negative correlation with functions. Among these biodiversity aspects, Sphingobacteriia showed the strongest capacity to maintain multifunctionality at low and moderate performance levels. Our findings will help to better understand the mechanisms through which biodiversity sustains the functioning of seasonally variable streams and their responses to global change.



# Challenges and opportunities for the assessment of the ecological status in temporary rivers

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Temporary rivers (TRs) are stream ecosystems in which the effects of anthropogenic disturbances are mixed with the effects of the natural disturbance imposed by flow intermittence. Despite the advances in TRs research, many gaps persist that limit the development of appropriate methodologies for the assessment of the ecological status. In this review, we identify the current challenges for the assessment of the ecological status of TRs and analyze the existing opportunities to address these challenges. These challenges focus on: the differentiation between natural and hydrologically impacted TRs, the differentiation between natural and anthropogenic disturbances, the development of biological indices for disconnected pools and dry riverbeds, the adaptation of hydrogeomorphological indices, and the application of the metacommunity theory in TRs. The opportunities are related to: the use of molecular tools, the existence of alternative indices to the traditional ones, the availability of data to be able to do modeling, and the social implication in the assessment of the hydrological and ecological status. The review focuses mainly on the scientific and management knowledge accumulated since the implementation of the Water Framework Directive in Spain but gathers experiences from TRs around the world to guide conservation and management actions in these unique ecosystems highly threatened by global change.

# Unsupervised classification of hydrological conditions in non-perennial rivers

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Anthropogenic pressures, such as land use change, water withdrawals and climate change, are leading to significant shifts in hydrological cycles, increasing the spatial extension of non-perennial rivers (NPRs). Knowledge about the frequency and duration of NPRs' intermittencies is severely constrained by the small number of streamflow gauges and limited reliability of hydrological models in predicting surface water presence. In this context, Sentinel-2 images can provide useful information for studying hydrological processes, offering effective ways to observe and monitor water surface dynamics with spatially explicit and temporally frequent data. The combination of multispectral satellite imagery and Convolutional Neural Networks (CNNs) can be used to automatically distinguish the three NPRs hydrological conditions: "flowing" (F), "ponding" (P) and "dry" (D). Working with multispectral Sentinel-2 images, the best band triplet for differentiating water from vegetation and sediment is B11-B8-B4, representing SWIR, NIR and Red bands. Learning on these False-Color Images, a CNN was trained to distinguish and classify the three hydrological conditions (F, P and D) in two reaches of the Palancia River (Spain). In addition, the CNN was trained to disregard a fourth class of images, in which cloud coverage was considered too high to proceed with the hydrological classification. In terms of accuracy, the performance of the CNN ranged in 0.79-0.88. Despite the small size of the dataset (1250 images) and the unbalanced data, the obtained result is promising and deserves further refinement, with the purpose of having a most powerful tool for automatic classification of hydrological conditions in NPRs.

# Exploring the diversity of benthic diatoms in Mediterranean temporary streams of Menorca (Balearic Islands)

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This study made it possible to compare benthic diatom communities which inhabit the temporary island streams on Menorca Island over a period of 18 years. In this work, we have compared databases of benthic diatoms from 10 different sites on Menorca Island and during 14 sampling campaigns, covering different seasons from three periods: 2005-2008, 2017, and 2022-2023 (n=100). In the laboratory, samples were processed according to standard methodology and 400 valves were identified per sample. In addition to diatoms, physicochemical and hydro-morphological parameters were measured in each study site and sampling campaign. More than 225 diatom species have been identified at these sites over the years, with the most abundant species being: *Nitzschia inconspicua*, *Amphora pediculus*, *Planothidium frequentissimum*, *N. frustulum*, *Achnanthydium minutissimum*, *Navicula veneta*, *Cocconeis euglypta* and *Navicula gregaria*. From the community level perspective, diatom communities were compared using MDS (Nonmetric Multidimensional Scaling) spatial ordination. Over these years, diversity showed very large variation among the localities studied over the year, with values ranging from 0.77 to 4.22 in 2005 to 2008 and from 1.44 to 4.41 between 2022 to 2023. Diatom indices such as IPS have also varied widely from 4.1 and 18.1 in 2005-2008, to 3.4 and 15.8 in the latter period. A detailed analysis of the localities studied over these 18 years helped to understand the physicochemical parameters variations affecting to diatom communities on Menorca Island.

# A metacommunity framework to anticipate the combined effects of drying and pollution on temporary rivers

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Incorporating the metacommunity framework into effective ecosystem assessment and conservation is urgently needed in temporary rivers, where drying-induced fragmentation alters spatiotemporal connectivity and impacts local species composition. However, current biomonitoring methods focus on the community response to local-scale processes (e.g., habitat and water quality conditions) and neglect regional-scale processes, like isolation and dispersal limitation. Here, we develop a conceptual framework to assess the dispersal impact on taxon richness and biological quality indices by quantifying spatiotemporal connectivity patterns generated by different drying intensities and pollution extents (i.e., polluted vs unpolluted). Using coalescent metacommunity models, we simulated macroinvertebrate communities following drying scenarios based on duration, spatial extent, and location. We characterized communities according to their pollution tolerance scores from the IBMWP index, and used information on dispersal groups (flying, swimming and drifting) from existing databases. We calculated a z-score of the community metrics for each scenario compared to reference scenarios with no pollution. Our preliminary results showed polluted and non-polluted reaches diverged from the reference scenarios when drying and pollution events increased in the river network. However, the magnitude of this decrease depended on the spatial distribution and the pattern of drying, as well as the level of pollution extent. Thus, our results suggest that drying and pollution's interactive effect on aquatic metacommunities makes biological quality indicators fail. We call for incorporating a metacommunity perspective into bioassessment tools, especially in ecosystems with high spatiotemporal variability.

# Survival at stake in Iberian streams: forecasting mussels and their fish hosts under climate change

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Climate change is increasingly impacting biodiversity in the Mediterranean region, notably in non-perennial watercourses of the Iberian Peninsula. For freshwater ecosystems, unprecedented biodiversity declines are being reported, particularly for the highly threatened freshwater mussels, fully dependent on fish hosts to complete their life cycle. Species Distribution Models (SDMs) are commonly used for predicting species distributions, but typical approaches overlook biotic interactions. Our approach assessed the potential impact of future climate scenarios on the distribution of six mussel species in the Iberian Peninsula, considering their obligatory interaction with fish hosts. Ensemble models incorporated environmental conditions and fish host distributions as predictors and were used to forecast current and future distributions. Results indicate severe projected impacts of climate change on Iberian mussel distribution, with *Margaritifera margaritifera* and *Unio tumidiformis* facing sharp habitat losses, potentially leading to regional and global extinctions, respectively. Remnant species *Anodonta anatina*, *Potomida littoralis*, *Unio delphinus*, and *Unio mancus* are expected to experience distributional losses but may also gain new suitable habitats, contingent on fish host dispersal ability. We further show that incorporating fish host distribution into mussel's model predictions resulted in additional areas with species loss under climate change. Our study underscores the imminent risk to the six studied mussels in Iberian Peninsula, emphasizing the urgent need for management actions to reverse trends in non-perennial watercourses and mitigate irreversible damage to species and ecosystems.

# On the connection (relationships) between aquifers and non-perennial streams: some examples from Menorca

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In karst terrains, underlain by carbonate rocks, the streams are mainly non-perennial due to high infiltration rates. In those areas, non-perennial streams play a key role recharging aquifers (groundwater inflows) and, at the same time, aquifers play an important role feeding the streams (groundwater outflows), especially during the dry season. Outflows from groundwater can be caused by a rise on the water table or through a spring. Although the influence of groundwater levels on stream flows and on the maintenance of connected or disconnected pools is well recognized, the empirical studies that analyze stream-aquifer relationships are still very limited. Here, we present several case studies located in the island of Menorca, where non-perennial streams are groundwater-dominated. Besides, groundwater is the main water resource in the island, thus groundwater withdrawals may limit groundwater outflows affecting the flow regime of the dependent non-perennial streams. These streams have not available historical record (gauging stations) and there is no information about flow-drying-rewetting patterns. On the other side, there is a long record of groundwater level measurements through wells. This contribution presents our research advances on the relationships between non-perennial streams and the groundwater levels. The study of this dynamic exchange is crucial for environmental flow assessment and for evaluating the impacts of groundwater withdrawals or the effluents from sewage treatment plants on these non-perennial streams

# Towards a protocol to evaluate the ecological status of temporary rivers: analysis of their associated terrestrial vegetation

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Temporary rivers are highly dynamic ecosystems in space and time that shift between aquatic and terrestrial habitats (flowing habitats, disconnected pools, dry riverbeds). They constitute the dominant fluvial ecosystems in arid and semiarid regions, such as the Mediterranean, and may represent over 50% of the world's river network length. However, their ecological relevance is not completely recognized and as result their protection status in the European and Spanish legislations is not explicitly considered. This work is part of the DRY-Guadmed project that aims to develop advanced tools for the assessment of the ecological status of temporary Mediterranean rivers during the dry phase, and to address existing management challenges and needs. We present a proposal and first results of a sampling protocol that will enable the assessment of the ecological status of rivers during the dry phase by the use of terrestrial vegetation. This protocol was applied to different dry channels from the Guadiana river basin along hydrological and anthropogenic pressure gradients. During the dry phase of 2023, different geomorphological units and their coverage were identified within each study. Then, terrestrial vegetation was sampled separately in each unit, estimating species cover in a semiquantitative scale. The results showed different plant communities associated to different geomorphological units along the study gradient, as well as indicator taxa which characterized each unit, with the potential to become indicators of the ecological status of temporary rivers during the dry phase.

# Diatom responses to human and natural disturbances in Mediterranean temporary rivers during the disconnected pools phase

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Temporary rivers shift among flowing and dry phases. They represent approximately 60% of the world's river network and their occurrence is expected to increase in the future due to global change. Despite their ecological relevance and their biodiversity value, there are still important unresolved questions regarding their ecology, management, and conservation. Diatoms are excellent bioindicators of water quality due to their sensitivity to environmental changes and pollution. However, there are very few studies that assess the effectiveness of current diatom indices in temporary rivers, and none in the disconnected pools during their dry phase. Diatoms in disconnected pools are characterised by biological communities that can be depauperate in species given the natural effects of pool size, time since disconnection, low oxygen levels, density of grazers, and spatial isolation. Our objective was to assess whether diatom communities respond to human impacts, based on the Mediterranean Reference Criteria (MRC) index, and the effect of natural flow disconnection across 65 disconnected pools in the Spanish Mediterranean region. We found that the effects of human impacts mainly explained diatom community variability, whereas natural characteristics of disconnected pools had a secondary role. Standardised diatom indices had a weaker response to human impacts. Our preliminary results suggest that diatoms can potentially be used to assess the biological quality of temporary rivers during the disconnected pool phase, but adaptation of diatom indices is needed.



# Diatom and aquatic macroinvertebrate metacommunity dynamics in temporary rivers

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Metacommunity dynamics can be affected by a variety of factors such as the spatial extent of the study, the spatial arrangement of the focal ecosystem, or the dispersal ability of the target organism. Commonly, environmental factors have been shown to be more important than dispersal dynamics for the organization of metacommunities at small spatial scales. However, in temporary rivers, flow intermittence could amplify the importance of dispersal dynamics even at small spatial scales due to spatiotemporal fragmentation of the river network. In this study, we tested this hypothesis by analyzing metacommunity dynamics in temporary rivers considering two groups of organisms with varying dispersal abilities: diatoms and macroinvertebrates. In addition, we tested the hypothesis that the relative importance of environmental and dispersal factors on metacommunity assembly would change across seasons as a response to drying dynamics. We sampled seven temporary rivers of Sant Llorenç del Munt i l'Obac Natural Park (Catalonia, Spain) covering a wide hydrological gradient during four seasons. We identified diatoms and macroinvertebrates across seasons for seven streams. We found that species sorting significantly explained macroinvertebrate metacommunity structure, while diatoms were mostly structured by dispersal limitation. Furthermore, local drying (at each point) and regional drying (between points) were determining factors for both organisms and their contribution to metacommunity structure varied across seasons. Understanding how metacommunities change in space and time in temporary rivers is essential for their conservation, especially in the Mediterranean Basin which is considered a biodiversity hotspot heavily threatened by human impacts.

# River imaginary: collective social imaginary around the non-perennial rivers of Menorca Biosphere Reserve

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The conservation of non-perennial rivers cannot be achieved without considering their human dimensions. Although it has been discussed that a generalized negative social perception towards non-perennial rivers is one of the main barriers to their conservation, empirical studies focusing on the human dimensions of these socio-ecological systems are still scarce. Here, we aim to study how non-perennial rivers of Menorca island are perceived by the island residents and, specifically, if there is a collective social imaginary around them. To do so, we carried out an individual face-to-face questionnaire to Menorca residents (n=144) and combined all responses to test for the existence of a collective vision. The questionnaire asked respondents: i) to express their level of concern regarding water related issues in the future (water availability, water quality, stream conservation, floods, droughts, and climate change), and ii) to indicate in a physical map which stream reaches are related to several aspects (best ecologically conserved, most impacted by human activities, most prone to flood risk, inhabited by fish, linked to their personal identity, and those with surface water during more days per year). Results showed that stream conservation is not a concerning topic in the social imaginary of residents, while water availability and droughts are main issues. Maps generated combining all individual responses revealed for which aspects there is a collective perception and how it contrasts with the available scientific and technical information. This study exemplifies how the social imaginary around non-perennial rivers may hinder the effective protection of these socio-ecological systems.

# Effects of drying on river greenhouse gas emissions and metabolism. Results from a meta-analysis

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We assess the detected effects of river drying on ecological functions such as gross primary production (GPP), and respiration (R), as well as on the emissions of greenhouse gases (CO<sub>2</sub>, CH<sub>4</sub>, and N<sub>2</sub>O) in rivers and streams. We performed a meta-analysis to determine whether the magnitude and direction of the effects changed across study systems, or because of types of drying (natural or human-induced), the severity of drying, or the climate. Drying induced some positive effects on GPP in two thirds of the studies, though drying magnitude greatly influenced the response. Low flow mostly caused moderate positive effects on GPP, though streambed desiccation caused its decrease. Drying caused a slightly negative effect on R during low flow or desiccation, and mostly in Mediterranean rivers. Drying caused a slightly negative effect on CO<sub>2</sub> emissions in Mediterranean and arid or semi-arid rivers, though enhanced emissions in rivers from oceanic and alpine climates. Drying magnitude was also relevant; low flows caused the decrease of CO<sub>2</sub> emissions, and streambed desiccation enhanced emissions. Both low flows and streambed desiccation caused moderate increases in CH<sub>4</sub> emissions, mostly in tropical or subtropical systems. Finally, drying decreased N<sub>2</sub>O emissions in arid/semi-arid and tropical/subtropical systems, though the wide range of effect sizes regarding the drying magnitude (low flow vs desiccation) or the type of drying (natural or simulated climatic) made this pattern inconclusive.

# Establishing physico-chemical indicators of ecological health in temporary rivers during the dry phase

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Temporary rivers, characterized by the presence of a dry phase, are subject to high levels of environmental variation, alternating between aquatic and terrestrial states. These ecosystems are widespread, abundant and increasing in extent, but a proper biomonitoring program to determine their ecological quality is still lacking and challenging to develop, including the physico-chemical state. When rivers are dry, traditional indicators of river health cannot be measured, however indicators to determine their ecological quality during the dry phase are barely starting to be developed. In this work, we examined extractable nitrate, phosphate, sulphate, pH, conductivity in dry riverbed sediments and co-occurring riparian soils to analyse their sensitivity to anthropogenic pressures, and therefore their potential as quality elements during the dry phase. For this end, we monitored 48 temporary rivers in different catchments in Spain (Segura, Guadalquivir, Guadiana, Tajo and Catalan internal basins) along a multiple-stressor gradient during the dry phase in 2023. In both sediment and soil habitats, nitrogen compounds (nitrate and ammonium) were significantly sensitive to anthropogenic pressures, showing higher values along increasing impact gradient. Our findings support the incorporation of the dry phase of temporary rivers into ecosystem monitoring and assessment works.

# Dry-to-flow transition and sediment transport: Implications for carbon and nitrogen fluxes upon flow resumption

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Intermittent rivers and ephemeral streams constitute more than half of the global fluvial network. While previous studies have explored the impact of drying attributes on river sediment biogeochemistry upon flow resumption, the influence of the dry-to-flow transition and flow characteristics on biogeochemical dynamics remains poorly understood. Flow resumption can occur instantly or gradually, and distinct flow velocities may lead to different sediment transport rates. To investigate how attributes of the dry-to-flow transition and sediment transport regulate carbon and nitrogen fluxes over 24h upon flow resumption, we used gas-tight cylindrical flumes (50 L) in a crossed experimental design. Dry river sediment experienced either i) an instant flow resumption event or ii) 6 hours of slow upward flow (precondition), followed by one of six different sediment transport rates ( $n = 3$ ). Our results suggest that regardless of the attributes of the dry-to-flow transition, sediment transport reduces  $\text{CH}_4$  and  $\text{N}_2\text{O}$  fluxes under a non-linear decreasing trend. On the contrary,  $\text{CO}_2$  exhibited higher rates from 0h to 12h under slow flow resumption, but instant flow resumption rates were higher from 12h to 24h. Sediment transport rates had a poor effect on  $\text{CO}_2$  rates. However, when we considered the 6-hour precondition as the starting point in the slow flow resumption, instant and slow treatments shared similar  $\text{CO}_2$  flux dynamics, peaking after 15h. Our results highlight sediment transport as a modulator of  $\text{CH}_4$  and  $\text{N}_2\text{O}$  emissions upon flow resumption, while  $\text{CO}_2$  flux seems to primarily depend on the presence of water, irrespective of the flow characteristics.

# Satellite images and web IT tools to identify and classify non-perennial rivers

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Quantifying the intensity of flow intermittency and the spatio-temporal extension of dry riverbeds remains one of the most important knowledge gap for non-perennial rivers (NPRs). Accurate mapping and classification of NPRs is currently not available in EU Member states, even if it would provide crucial baseline information for water management and biodiversity conservation.

In this context, satellite images and remote sensed data can be used to identify and classify flow intermittency in NPRs, detecting flow occurrence along river reaches. Satellite data have a short revisit time (about one week), and the recent availability of free of charge, high spatial resolution data (e.g., ESA Sentinel-2 mission) already opened up the possibility of innovative applications for NPRs.

Based on the results of the RIVERTEMP project [Erasmus+ 2022-I-IT02-KA220-HED-000086223], we present a new web IT tool based on the analysis of Sentinel-2 multispectral images. The false colour composition of SWIR, NIR and RED bands is automatically generated by the web tool for selected river reaches, and can be used to distinguish water surfaces from other components of the river corridor. Detecting the presence of surface water allows to identify the hydrological conditions of the river reach, that can be: “flowing” (F), “ponding” (P) or “dry” (D) conditions. The web tool generates imagery time series of each river hydrological condition, allowing the automatic classification of NPRs using the temporal duration of F, P and D classes. A user login web page, manuals and training materials will be freely available from the web-page of the project.

# A comprehensive spatial analysis of invertebrate diversity within an intermittent stream network: responses to drying and land use

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Freshwater ecosystems, highly vulnerable to the impacts of climate change, also face additional threats from human activities. Changes in land use, leading to water pollution and habitat degradation, adversely affect both biodiversity and ecosystem functioning. Intermittent streams, which constitute nearly half of all fluvial systems, support a rich diversity of species adapted to cope with natural drying. This study examines the impact of drying and different land uses on the taxonomic and functional diversity of aquatic invertebrates in a small Mediterranean intermittent stream network. By seasonally sampling 16 reaches, we hypothesised that extended dry-phase durations and agricultural activities would both diminish  $\alpha$ -diversity, with drying having a more pronounced impact on  $\beta$ -diversity than agricultural practices. We also anticipated that drying and agricultural activities would modify species and trait compositions, favouring taxa tolerant to desiccation and generalist species. Drying negatively impacted the taxonomic and functional  $\alpha$ -diversity of aquatic invertebrates but positively influenced  $\beta$ -diversity. Land use was found to affect only  $\alpha$ -diversity. In particular, habitat heterogeneity and increased nutrient levels in the water within the stream network correlated positively with invertebrate diversity. However, the negative effects of drying were less pronounced in the upstream forested regions, which had higher habitat heterogeneity, compared to the downstream areas affected by agriculture. With an expected spatiotemporal increase in flow interruption due to global change, our research underscores the importance of preserving natural and forested streams in intermittent networks, especially in headwater regions, to facilitate recolonization when flow resumes in the stream network.

**SS5**

**Justice, Equity, Diversity, and  
Inclusion (JEDI) in Limnology**





## Reflecting on the impacts of climate change on freshwaters and the role of women through the lenses of art

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Climate change is threatening rivers, lakes and wetlands across the globe, generating inequalities towards and discrimination against women. From the disconformity, women raise their voices and lead the study, defense and protection of freshwater ecosystems. Yet, there is still a general lack of awareness of these gender biases when we talk about climate change and water crisis. The exhibition “Augmented Ecofeminisms: Climate, Water, Women (MUAC)”, created by the Gender & Science Group of the Iberian Association of Limnology, invites reflection on the effects of climate change on freshwaters and the dual role of women as both a vulnerable group and agents of change. Through eight art pieces that combine augmented reality with scientific, artistic and audiovisual material, the exhibition proposes a journey that questions the current management of environmental problems related to water and explores potential futures based on alternative views of contemporary politics. MUAC exhibition, inaugurated in October 2023, has been hosted by several national and international universities and is currently freely available online ([genderlimno.org/muac.html](http://genderlimno.org/muac.html)). Further, the content of the exhibition has been adapted for children through an illustrated book that tells eight stories related to the global water crisis from an ecofeminist perspective (project AUMENTA, FECYT). This open-access book is complemented with didactic materials designed for being used in primary schools. Overall, these science in arts projects aim to contribute to our understanding of the social-environmental impacts of the climate emergency from an ecofeminist perspective.

# Pocas “luces” y muchas “sombras” en el cuidado de mayores desde una perspectiva de género: Un análisis desde la Academia

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Esta comunicación forma parte de un estudio más amplio cuyo objetivo principal es explorar cómo el cuidado de mayores afecta la vida profesional y personal del profesorado y personal investigador, desde una perspectiva de género, para lo cual se entrevistaron 24 mujeres y 12 hombres investigadores de España que son o han sido cuidadores de mayores. Los resultados apuntan a que el cuidado de mayores es realizado básicamente por las mujeres. La normalización del cuidado, por parte de las mujeres, es denunciado por el 33,3% de las entrevistadas. Los hombres lo perciben más como una obligación impuesta. Además, en ambos géneros, el cuidado de mayores despierta sentimientos contradictorios, pero sobre todo de culpabilidad por no ser capaces de hacerlo mejor o por tener sentimientos de rebeldía ante los elevados costes profesionales, físicos, psicológicos, e incluso económicos que supone, y que muchas veces son muy difíciles de gestionar. Sin embargo, algunos entrevistados sostienen que, el cuidado de mayores también les ha servido para aprender habilidades nuevas o para vivir experiencias y enseñanzas positivas útiles para afrontar la vida. La Academia es poco permeable a la situación que vive el personal docente e investigador. De hecho, a nivel institucional, el cuidado de mayores es un problema “invisible”, para el que no hay respuestas o ayuda a las personas cuidadoras. Finalmente, el personal entrevistado aporta algunas ideas e iniciativas que podrían facilitar la tarea del cuidador de mayores.

**SS6**

**Exploring the effects of global  
change on freshwaters: The  
crucial role of LTER studies**



# Temporal variation of river macroinvertebrate communities in Picos de Europa during the last decade

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Mountain rivers are sensitive to global change alterations (climate, land use, etc.). However, due to the small size of their basins, certain segments of mountain river networks are not included in the typological classification defined in the National Hydrological Plan. As a result, they are excluded from the monitoring programs established by Demarcations. To assess the long-term evolution of river invertebrate communities in Picos de Europa, a network of 13 monitoring sites (3 in springs and 10 in rivers) was designed. This network distinguishes Control stretches (7), stretches affected by punctual effluents (4) and catchment runoff processes (2). This monitoring network has been continuously operated since 2012. Data collected on river invertebrate communities during this period have been integrated with other long-term European studies to determine the temporal trend of their biodiversity in Europe. These results, recently published in the journal *Nature*, reveal a stagnation in the recovery of the biodiversity of European rivers in the last 10-20 years. This study highlights the trend of these communities in Picos de Europa, which exhibit one of the most negative trends among all the European rivers analyzed. Furthermore, at local level, we analyzed environmental variables (e.g. climate or water quality) that may be driving these temporal patterns over the last decade, looking if the taxonomic composition of these communities reflects a taxa replacement during this period.

# Looking beyond the surface: Understanding the role of multiple stressors on Albufera lake's chlorophyll-A using Generalized Additive Models (GAMs)

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Human-induced stressors are significantly impacting aquatic ecosystems, requiring a comprehensive understanding of their interactions and effects to guide effective management and conservation efforts. Generalized Additive Models (GAMs) stand out in statistical modelling for their capability to model non-linear relationships in ecology but their use in studying multiple stressors effects is still limited. The Albufera Lake in Valencia, Spain, presents a complex scenario of interacting stressors, from agricultural impacts to climate change and pollution. Here, we developed a database tracking stressors and water quality parameters for the Albufera lake since the 1970s, used GAMs to examine stressor dynamics, their impacts on Albufera Lake's chlorophyll-A concentration, and to identify variable combinations to meet WFD's targets, thus guiding management efforts. The study highlights the impact of reduced water inflow due to more efficient irrigation, the influence of rice cultivation on the Albufera Lake's hydrology and the multi-dimensional effects of climate change. Time-series analysis also revealed a recent summer increase in Albufera Lake's salinity, in addition to persistent eutrophication issues. Pollutant toxicity showed a notable rise during August-September linked to agricultural pesticide applications. Chlorophyll-A concentrations, which reached their maximum in 1985, have been mostly consistent since 1994, mainly due to the improvement in wastewater treatment plants improvement. Modeling results also highlight that enhancing water transparency in the Albufera Lake involves increasing water inflow along with extreme conductivity values.

# Changes in lake macrophyte communities over three decades: Tracing the impacts of global change in shallow Alpine lakes of the Pyrenees

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Submerged macrophyte communities constitute an essential component of lake biodiversity, generating structure, regulating biogeochemistry at lake interphases, and providing habitat and refuge to different groups of organisms.

This study investigates changes in macrophyte communities for 30 Pyrenean lakes over the past 35 years. Employing semiquantitative inventories on transects repeated over time, we assess changes in species presence and abundances, and analyze changes in community structure in relation to measured shifts in lake water chemical characteristics. Specifically, we estimate coverage trends of 18 species, for which we evaluate depth distribution ranges and overall changes at community level by comparing the current biodiversity state with that from late 1980s using mixed models for species responses, temporal beta-diversity index, and multivariate analysis for analyzing and displaying patterns of change in environmental and community data.

Although community shifts appear heterogenous among studied systems, suggesting the interplay of multiple factors, some general trends emerged. The most important trend was the expansion of a few natopotamids, while only two taxa declining, one being an indicator of extreme oligotrophic conditions (*Sphagnum* spp.), and the other a species with emergent leaves inhabiting superficial waters (*Sparganium angustifolium*).

Exploration of water chemistry data series (years 1987, 2011 and 2023) revealed consistent shifts along to directions characterized by reduced concentrations of nitrogen species, alongside with increases in TP, conductivity, and alkalinity. Those changes were coherent with the observed expansions of species that thrive in more alkaline waters in the regional context.

# From long term monitoring to multilevel gap-free socioecological databases

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Freshwater ecosystems integrate multitude of pressures throughout the path that water follows across landscapes. Decades of investment on water treatments, ecological restoration and conservation measures have not changed the general condition of freshwater ecosystems as being among the most threaten on earth. The causal effect relationship between restoration and conservation actions and the recovery of freshwater ecosystems is still far from clear. This daunting scenario gets even more worrisome if we account for the current and future effects of climate change. The generation and exploitation of long-term datasets dealing with freshwater biological communities and multiple drivers (e.g., discharge, temperature, water characteristics etc..) is paramount to gain a deeper understanding on how freshwater ecosystems, communities and populations respond to natural and anthropic changes. This is the only way that conservation measures and restoration activities could be prioritized to produce a systematic change to the current status of freshwater ecosystems. However, even long-term data schemes usually suffer from many spatial and temporal gaps, or even lack of relevant factors that could be essential pieces of the puzzle when trying to understand cause-effect relationships. In this talk, we expose the need to generate data across multiple socio-ecosystem hierarchical levels to assist on a better understanding of processes across scales and we show some approaches that have already attempted to achieve this goal. We also draw attention on new endeavors in this direction that are been developed across LTER nodes and LTSER platforms in the EU and beyond.

## Impacts of reduced precipitation on lake ecosystems: regime shift through enhanced nutrient recycling

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Eutrophication is a main threat to continental aquatic ecosystems. Prevention and amelioration actions have been taken under the assumption of a stable climate, which needs reconsideration. Here, we show that reduced precipitation can bring a lake ecosystem to a more productive regime even with a decline in nutrient external load. By analyzing time series of several decades in the largest lake of the Iberian Peninsula, Lake Sanabria, we found autocorrelated changes in the variance of state variables (i.e., chlorophyll and oxygen) indicative of a transient situation towards a new ecosystem regime. Indeed, exceptional planktonic diatom blooms have occurred during the last few years, and the sediment record shows a shift in phytoplankton composition and an increase in nutrient retention. Reduced precipitation almost doubled the water residence time in the lake, enhancing the relevance of internal processes. This study demonstrates that ecological quality targets for aquatic ecosystems must be tailored to the changing climatic conditions for appropriate stewardship.



# Temporal variation in populations of brown trout in the Picos de Europa National Park

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The brown trout (*Salmo trutta* Linnaeus, 1758) is a species of great importance from an ecological and socio-economic point of view. It is a keystone species and a top-predator, as well as one of the most popular and important sport-fishing species in the Iberian Peninsula. Despite its importance, trout populations have decreased in recent years in most of its natural range, being classified as vulnerable in some countries, such as Spain. The spatial-temporal patterns of the populations of this species can be affected by density-dependent and density-independent factors, depending on the life stage and environmental conditions. Moreover, population-process such as natality-mortality or emigration-immigration can be influenced by fluvial connectivity and the degree of isolation of the populations. In order to determine the main factors controlling the temporal variation patterns of trout populations in the Picos de Europa National Park, an ecological monitoring network of 13 sampling points has been used. In this monitoring network, systematic sampling of fish fauna has been carried out during the summer season from 2012 to present. To analyze the influence of fluvial connectivity and dispersal in local density, populations have been characterized by their isolation degree. Moreover, the most important environmental variables that could affect fish populations have been incorporated to examine their influence on population dynamics. This study highlights the importance of fluvial connectivity and environmental factors in determining the spatio-temporal variation patterns of trout populations.

# Use of biomonitoring data as LTER networks: challenges and opportunities

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Freshwater ecosystems are being continuously monitored in many parts of the world, which make them one of the most interesting systems to study global change effects. Initiatives to exploit the relatively long time series available are gaining momentum and, consequently, papers that set the general picture of the changes of freshwater diversity in the last decades are being published. The Basque Country runs a biomonitoring network with one of the highest spatial and temporal resolution in the world. This network offers infinite opportunities to investigate biodiversity changes, responses of biota to anthropic stressors, changes in seasonality or the stability of freshwater communities. In this talk we will showcase the latest discoveries based on long term data of biomonitoring networks at the continental scale and in the Basque Country. We will show the challenges and limitations that this kind of datasets have. We will also show their potential to address relevant issues regarding to basic and applied ecology.

# Long-term spatio-temporal dynamics of fluvial metabolism in rivers of the Cantabrian Cordillera

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River metabolism includes carbon fixation (primary production, PP) and mineralisation fluxes (ecosystem respiration, ER) by the organisms of the river ecosystem and integrates the response to a wide range of natural and human factors. For instance, metabolism in mountain rivers and streams is highly sensitive to changes in hydrology or temperature and potentially, a good indicator of climate change. Moreover, these ecosystems represent a large fraction of the total river networks, making them key elements for understanding the impacts of climate change. In this context, the main objectives of this work are 1) to analyse the spatio-temporal dynamics of metabolism in rivers of the Cantabrian Cordillera during the last decade and 2) to improve our understanding of which and how environmental factors govern this variability. To achieve these objectives, summer metabolism was estimated in 15 river reaches from 2014 to 2022, using time series analyses. The role of environmental factors was also analysed using mixed effects models. The results show a significant increase in ER since the beginning of the series, i.e. ecosystems are tending towards greater heterotrophy, and higher greenhouse gas emissions. Furthermore, it is shown that PP and ER are mainly conditioned by flow, water temperature and solar radiation. These results are of particular relevance as few studies consider this positive feedback effect of rivers on global warming. In addition, this shows the importance of long-term monitoring of ecological indicators that will allow us to understand the evolution of areas of conservation interest, such as the Cantabrian Cordillera.

**SS7**

**Adaptive approaches for water  
resources management and  
wetland restoration**



# Integrated approaches to decision-making in wetland restoration planning

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In the planning process for wetland restoration, scientific, social, and governmental interests and understanding converge. Hence, these three sectors must cooperate throughout the process and information must flow efficiently (and equitably) among the three. Decision-making tools for restoration planning may be a key element to understand, evaluate and synthesize the results of preparatory assessment on economic effects and ecosystem-service impacts of the proposed policies and interventions. We present the result of a systematic review of decision-making tools for wetland restoration, focusing on a key step of the decision-making process: the definition of objectives. We focus on the definition of objectives because it is often an underestimated step whose inadequate formulation, leading to agreed objectives that are too broad, unclear, or undefined (in terms of action time, expected effects and degree of fulfillment) compromise the implementation, evaluation and refinement of the whole restoration process. In the review, we compile and compare the methodologies used for decision-making during the definition of objectives in wetland restoration, and categorize the objectives identified through their application. We further exemplify how an adequate definition of objectives influences decision-making in subsequent steps in the restoration plan, including the evaluation of restoration success, particularly when facing the various sources of uncertainty inherent to the restoration process. For this purpose, we use the literature reviewed to inquire into the links between the decision-making processes and the socio-natural dynamism of the restoration process, using a theoretical framework of adaptive co-restoration.

# Blanket bog restoration in NW Spain. A case of collaborative approach to habitat conservation management

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EU and UN regulations state 30% of degraded areas should be restored by 2030, and wetlands are among the first in the list. Effective restoration in natural areas stem from participatory approaches that includes all actors in the design and implementation of the actions. Here we report on a restoration action on a blanket bog in the SAC “Serra do Xistral”, in the north of Lugo province, NW Spain. This demonstrative action was part of the Life in Common Land project, aimed to improve the conservation status of bogs and wet heathlands in the area. Blanket bogs are inland wetlands of high importance for climate and water regulation because they are composed largely by peat that serve as carbon sink, capture atmospheric water, and are priority habitats according to the EU Habitats directive, therefore in need of urgent conservation. Blanket bogs are distributed exclusively in the northern Atlantic region, with only one relevant area in southern European countries, in Xistral. The Xistral mountains were afforested with Scottish pine (*Pinus sylvestris*) during the 1950s, now identified as a main environmental issue for the preservation of the bogs. We describe the workflow starting with the identification of the commoners needs and conservation priorities to decide over candidate areas. The design of the action was developed with inputs from local practitioners and owners to guarantee the feasibility and replicability. This experience may aid in planning new restoration actions to improve the conservation status of the blanket bogs in their southern distribution limit.

# Estuarine ecosystem services, societal benefits, and human interactions: Mondego's wetland case study

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Transitional ecosystems represent essential natural habitats, providing important ecosystem services (ES), societal goods, and benefits crucial for human well-being. The provided ES are essential for coastal protection and water quality regulation, while socioeconomic benefits include fisheries and recreation/tourism. Although industrialisation and urbanisation have been imposing major pressures to transitional ecosystems, climate change (CC), such as sea-level rise (SLR), is expected to intensify them during the 21<sup>st</sup> century. However, socioeconomic dynamics (implementation of adequate adaptive responses) will assist in managing those estuarine/coastal ecosystem's pressures. The DPSI(W)R(M) conceptual framework tool requires its elements to be linked in a logic articulation of compartments inside a defined area and is pertinent for problem-structuring in coastal/estuarine ecosystems. Therefore, Drivers of basic human needs require Activities to be fulfilled, leading to Pressures, which, will lead to natural system's State changes, that Impact human Welfare. Here, policymakers will develop management Responses (as Measures) to address risks associated with human activities. DPSI(W)R(M) allows the assessment of environmental and socioeconomic impacts of all the activities related to the ecosystem, both inside and outside the area impacted by CC. The Mondego estuary (Portugal) has suffered from human development by intervention, altered natural environment to accommodate commercial vessels, margin regularization and, agricultural exploitation. Therefore, this work synthesises knowledge of the Mondego for impact management of risks associated with the ecosystem's exploitation, quantifying the DPS framework's first stages. This enables the analysis, with local stakeholders, of future scenarios for the estuary and population, while working towards a sustainable future under SLR threat.

## Exploring the human dimension of climate change: emotional attachment to a coastal environment

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There is a growing trend to include public participation in research projects, not only to bridge the gap between science and society, but also to consider the public perception as part of the decision-making process in environmental management. Numerous studies focus on the impacts of climate change and human-mediated on coastal areas, but less consider how these impacts affect the emotions of their inhabitants. We engaged a participatory process in the Ebro Delta in October 2022, to inquire into stakeholders' perspectives on emotional attachment, adaptation strategies, and management approaches. To compare perceptions between local stakeholders and scientist, in March 2023, we conducted an online questionnaire for acquiring information on their perception and to explore their emotional responses, experiences and thoughts. We carried out qualitative research by using ATLAS.ti software, and developing semantic networks. Results showed a high level of concern about the current effects of climate change. Specifically, the most common feelings about the effects in their life/work were impotence, frustration, and anger. Also, they were worried about the uncertainty of the future of the delta, the inheritance that will be left to their children, and feeling of abandonment from public administrations. Initiatives that count with the active participation of stakeholders promote a new approach to our relationship with nature, raising awareness regarding environmental problems. Incorporating people's feelings about climate change into policy can be achieved by educational programs, social marketing for sustainable living, framing messages to resonate emotionally, and solicit stakeholder feedback to evaluate policy effectiveness.



# Enhancing climate mitigation in wetland management: flood gradient impact on greenhouse gas emissions

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Wetland management has traditionally been designed for the maintenance or enhancement of biodiversity. While not compromising their natural structural features, efforts are required to improve the functioning of these ecosystems. Wetlands can mitigate climate change, thus actions could also be taken to ensure or even increase this key ecosystem service. Within the LIFE Wetlands4Climate project, management and restoration actions focused on reducing greenhouse gas (GHG) emissions and enhancing the wetland's mitigating role were experimentally evaluated. Our study assessed how the flooding pattern influences CO<sub>2</sub> and CH<sub>4</sub> emissions. GHG fluxes were seasonally measured in managed and unmanaged areas of different temporary wetland types (inland freshwater and saline, and coastal), aiming to generate knowledge for a proper (following natural patterns) flooding-draining water management in these temporary ecosystems. Results showed higher CO<sub>2</sub> flux levels in dry areas (or time periods) with minimal methane emissions, while CH<sub>4</sub> were maximized at the wet-low water interface. The assessment of these and other measures regarding environmental values, wetland uses, and the carbon balance and ecosystem services was included and assessed using action/benefit matrices, which were provided to managers. These matrices considered both the importance and the magnitude of each management action with respect to the wetland variables considered. Quantifying the effect of each integrated management action in the matrix allowed managers to evaluate the management proposals included in plans and define the possibilities of maximizing climate mitigation of Mediterranean wetlands through adaptive management.

# Adaptive co-restoration: an integrated approach for the restoration of socio-ecosystems facing global change

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Traditional ecosystem management face critical challenges derived from the complexity and unpredictability of socio-ecosystems, which often result in unexpected responses that may trigger or stabilize undesired system states. These two characteristics (complexity and unpredictability) are largely inherent to socio-ecosystem dynamics, due to their openness, multi-scalarity, path-dependency and non-equilibrium dynamics; but they also result from the disregard that traditional, command-and-control approaches have for stakeholder agency in politicized decision arenas. These risks are intensified in ecological restoration projects, especially under current climate and global change – owing to ecological processes associated to anthropogenic disturbances and interventions (e.g., priority effects and empty-niche dynamics resulting in long-lasting path dependencies; state shifts triggered by disturbances, particularly when they closely follow key interventions). Adaptive-co-management provides a methodological framework for the long-term design of ecosystem and natural-resource management strategies. It operates by accepting the aforementioned uncertainties at face value and focusing on facilitating knowledge co-production, joint decision-making and sustained learning as key element of socio-ecosystem adaptation and resilience. In this communication, we lay out the key elements of adaptive co-management; identify specific factors of ecological (and, more specifically, wetland) restoration that reinforce some of the challenges addressed by it; and propose a framework for adaptive co-restoration that could help address these challenges.

# Participatory science for raising awareness and citizen action in favor of the human well-being provided by wetlands

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Citizen science has great potential to develop outreach and environmental education, to enter fully into the democratization of civil action based on scientific evidence that is also experienced. In this paper, we report on a participatory science project that addresses two types of wetland ecosystem services.

Decades of fertilizer application and surface spreading of animal manure led to significant increases in nutrient concentrations in groundwater worldwide. High nitrate concentrations in groundwater can lead to health risks and pollution contamination. Literature reports that restoring strategically placed wetlands that add only 1% of the watershed area potentially reduces 50% of nitrate in runoff. Hence, we led the first participatory mapping of nitrates in the Campo de Calatrava aquifer system, together with 50 citizens of the area. It reveals the power of volcanic wetlands and river meadows in reducing up to 90% of nitrate concentration. Besides, citizen participation led to a 33% increase in nitrate data in the area.

In addition, we explored the mental and physical well-being provided by the observation of biodiversity through an ornithological itinerary and an entomological workshop focused on the work and life of “The Priest of the Bugs”, i.e., the locally renowned naturalist José María de la Fuente. These activities reinforced the population’s sense of identity with their wetland.

We evaluated the effectiveness of the activities through post-pre surveys, which identified the participants’ prime motivation for participatory science, the improvement of their perception of their wetland services, and the prospects for further citizen science activities.

# Poster Communications

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# OSI

## Microbial Ecology



## Land use changes and denitrification in headwater streams

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Population growth and crop intensification are the main land-use drivers altering biodiversity and biogeochemical cycles. Eutrophication is a major impact of land use intensification on aquatic ecosystems caused by excess nitrogen (N) and phosphorus (P) from urban wastewater and agricultural leachates. Preventing eutrophication or restoring eutrophic ecosystems is currently a major challenge. In this context, improving our understanding of denitrification is crucial, since it is the main process that removes definitively reactive N from the ecosystem to the atmosphere as N<sub>2</sub> or N<sub>2</sub>O. We studied denitrification in sediments of headwater streams located in catchments with dominance of four land-use types: natural vegetation, forestry, urban and agricultural use, with three streams per type. We hypothesized that the contrasting sources of C and N across land-use types could have a significant effect on the specific way of the denitrification process. For this purpose, we collected water and sediment samples from the streams in the winter-spring season. Water samples were analyzed for nutrient contents and physical characteristics (temperature, electrical conductivity and dissolved oxygen). Sediment samples were analyzed for redox potential, particulate organic matter quantity and major cations and anions. We used molecular methods (RNA extraction and qPCR) to analyze the transcriptional abundance of the main bacterial denitrifying genes (NarG, NirS, NirK, NorB and NorZ) of the fine sediment portion. Preliminary analyses have detected the presence of these genes in the different streams. We expect to find a higher denitrification rate in streams with more anoxic sediments, these being urban and agricultural.

## Combining metagenomics and stable isotopes to understand biogeochemical processes in sediments of Iberian saline lakes

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Inland saline lakes are highly complex systems. Salinity decisively influences water chemistry and imposes major physiological barriers to microorganisms. Therefore, the biogeochemical processes that maintain the ecological functioning of these ecosystems are strongly conditioned but still poorly understood. The diversity of saline lakes in semi-arid inland areas of the Iberian Peninsula represents an opportunity to unravel the singular biogeochemical processes that govern the ecology of these ecosystems, a knowledge that is essential for their conservation and management. We have studied twenty-one inland lakes, which cover important salinity and anthropogenic impact gradients, through the analysis of lake sediments, which integrate multiple signals of key biogeochemical processes. A complementary analysis of sediment composition and stable carbon and nitrogen isotope signatures, along with the use of metagenomic techniques to characterize the gene and metabolic pool based on Metagenome-assembled genomes (MAGs) and functional annotation, allowed for an integrated view of the biogeochemistry of the lakes, the conditions imposed by salinity and its interrelation with the main environmental parameters. Finally, water availability and salinity condition the carbon content of the sediment, which in turn regulates the nitrogen and phosphorus cycle in the water column, also the nitrogen isotopic signature is a reliable indicator of the human impact of wastewater on the studied ecosystems. This work was supported by the project CLIMAWET-CONS (PID2019-104742RB-I00), funded by Agencia Estatal de Investigación and the Ministerio de Ciencia e Innovación (Gobierno de España), as well as by the project LIFE Wetlands for Climate (LIFE19 CCM/ES/001235) funded by the European Union.

# Biotic and abiotic drivers of Aquatic Hyphomycete assemblages of Sierra Nevada streams

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Riparian vegetation produces shade that limits primary production within the river and, at the same time, provides litter, which is processed at stream ecosystem by the combined action of decomposers and detritivores. Aquatic hyphomycetes (AH) are the most important microbial decomposers (saprophytic fungi) and facilitate the utilization of litter resource, through microbial conditioning, for ulterior consumers. However, the ecological knowledge of this conspicuous group is still rather limited, being usually described as generalists with low substrate specificity. From the ecosystem functioning point of view, their functional redundancy precluded further analysis of their ecological role. However, the lack of basic knowledge on this group may hinder the acquisition of important information for understanding the ecological integrity of headwater streams. In order to fill this scientific gap, we studied conidia of the AH assemblages in water column and natural foams from 41 stream sites located in Sierra Nevada National-Natural Park and surrounding areas, and fully characterized environmental conditions from local -stream reach climate, water chemistry, benthic organic matter and riparian vegetation- to catchment -vegetation and landuse- scales. Our preliminary results suggest that several AH species resulted characteristic for different stream typologies and clearly respond to the considered biotic (e.g., riparian community) and abiotic (e.g., climatology) features, suggesting that these fungal decomposers might be used as stream ecosystem bioindicators as we increase our scientific knowledge at the species level.



**OS2**

**Processes and Functioning  
of Ecosystems**



# Reciprocal stream–riparian fluxes: effects of distinct exposure patterns on litter decomposition

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Resource fluxes at the stream–riparian interface are a vital contributor to both systems' energy budgets. The effect of distinct litter exposure patterns—direction of the riparia–stream movement and duration of exposure at each habitat—however, remains to be elucidated. In this field experiment, oak leaves in fine and coarse mesh bags were either exposed to a stream-to-riparia or riparia-to-stream movement sequence for distinct periods (2:6, 4:4, or 6:2 weeks). After 8 weeks, ash-free mass loss, microbial activity, and fungal biomass were compared in leaves undergoing inverse movement sequences (e.g., 2-week exposure to the riparian area at the beginning vs. end of the colonization period). Mass loss in coarse mesh bags was negatively affected when leaves were previously exposed to a short terrestrial pre-conditioning period, despite higher microbial activity and fungal biomass, when compared to the inverse movement. This effect on mass loss was neutralized by longer terrestrial exposures that likely allowed for a more thorough conditioning of the leaves, through extended leaching and terrestrial microbial colonization. Our results suggest that terrestrial pre-conditioning periods of <2 weeks lead to litter-quality legacy effects in tough leaves, to which aquatic communities respond through lower substrate degradation efficiency, hindering stream decomposition. Contrastingly, oak aquatic pre-conditioning, regardless of duration, provides riparian communities with a high-quality resource, promoting litter processing through grazing behavior. As climate-induced hydrological shifts may result in altered provision/ quality of detritus subsidies at the stream–riparia interface, we suggest that assessments of decomposition dynamics should consider the entire litter conditioning history.

# Alder disappearance from riparian forests affects the quality and decomposition of litter from non-nitrogen-fixing tree species in streams

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Alder (*Alnus glutinosa*) is a common species in the riparian forest of many streams. As a nitrogen (N)-fixing species, alder has the potential to affect the leaf characteristics of neighbor non-N-fixing species. However, alder is currently being affected by the pathogenic oomycete *Phytophthora alni* across Europe. Therefore, alder dieback can have indirect consequences for forest streams that depend on leaf litter provided by the terrestrial vegetation, by changing the leaf litter characteristics of remaining tree species. In this study, we assessed the effects of alder disappearance on the leaf characteristics of three non-N-fixing species: oak (*Quercus robur*), chestnut (*Castanea sativa*) and beech (*Fagus sylvatica*). Leaf litter from trees in monoculture and trees in mixed plantations with alder were obtained from the BangorDIVERSE experimental plantations (Bangor University, UK), and litter was chemically and physically characterized using standard methods. Further, microbial-driven leaf litter decomposition was assessed in laboratory microcosms over 70 days. In the absence of alder, chestnut leaf litter was tougher and had higher phenols and lignin concentrations and lower C/N; oak leaf litter had higher lignin concentration; and beech leaf litter had lower lignin concentration and higher C/P. In accordance, leaf litter from oaks and chestnuts in monocultures decomposed slower than those from trees grown in mixtures with alder, while leaf litter from beech in monoculture decomposed faster compared to that from trees grown in mixture. In conclusion, in ecosystems strongly dependent on leaf litter, changes to its characteristics due to the loss of alder trees may affect stream functioning.

## CO<sub>2</sub> fluxes and their drivers during dry phase in temporary ponds

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Pond ecosystems play a key role in the global carbon cycle, potentially influencing it by acting as both carbon sink and source. The knowledge of CO<sub>2</sub> emissions in ponds including the relevance of dry phases has rarely been addressed. Climate changes in temperature and precipitation are altering pond hydroperiods causing many shifts from permanent to temporary or even permanently dry states, impacting carbon fluxes. Ignoring the CO<sub>2</sub> fluxes during dry periods could result in underestimating global CO<sub>2</sub> budgets. Here, we aim to assess the drivers of CO<sub>2</sub> fluxes on exposed sediment during the dry phase in 23 temporary ponds across Europe, distributed in Mediterranean (Spain) and temperate regions (Germany, Denmark, and Belgium). Our study focuses on environmental and local factors including sediment characteristics and organic matter composition to elucidate their role in CO<sub>2</sub> fluxes. We characterized sediment properties and analyzed fluorescence spectroscopy characteristics of water extractable organic matter using parallel factor analysis. Preliminary results indicate that ponds are net sources of CO<sub>2</sub>, emitting high rates during the dry phase, yet we obtained a few negative fluxes. The emissions ranged from 127 to 4888 mg C m<sup>-2</sup> day<sup>-1</sup> (mean ± SD 1406 ± 1205 mg C m<sup>-2</sup> day<sup>-1</sup>, median = 1078, n=30). Our first results indicate that sediment temperature and moisture are the most significant driver of CO<sub>2</sub> fluxes along with organic matter properties and sediment characteristics as conductivity. Our research highlights the importance of considering dry phases in ponds for accurate assessments of CO<sub>2</sub> budgets and the underlying mechanisms driving carbon dynamics.

# Biotic and abiotic drivers of Aquatic Hyphomycete assemblages of Sierra Nevada streams

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It has been proposed that light-limited lakes are characterized by the constraint of the autotrophic pelagic flux (the green pathway) and the dominance of the detritus-based heterotrophic flux (the brown pathway) in the food web. However, studies explicitly testing this assumption are scarce in natural food webs. Here, we estimated energy flux in a dystrophic light-limited shallow lake to investigate the importance of green vs. brown energy pathways to fuel the freshwater food web. In one year, we monthly estimated the intake energy flux for five fish trophic guilds (detritivores, top carnivores, meso-carnivores, herbivores, and omnivores). We found a high concentration of energy flux in the detritivore compartment, accounting for over 93% of the total energy flux through the food web in the year. A single detritivore species (*Cyphocharax voga*) dominated fish community, both in terms of biomass stock and number of individuals. There was a substantial increase in the detritivore energy flux during warmer months. We also found a contrasting scenario for herbivory (autochthonous green pathway) which accounts for insignificant amounts of total flux in fish community. These results illustrate a remarkable dominance of brown pathways, which support the energy flux through food web in dystrophic shallow lakes, reaching a maximum during the warmest season, point out a synergy between warming and brown pathways.

# The importance of bed substrate selection in Constructed Wetlands: implications in the ecosystem functioning

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The bed substrate in the subsurface constructed wetlands (CWs) are not only the physical medium for the biota growth, but also the main source of C and essential macro and micronutrients for plant and microorganism development. Selecting the adequate bed substrate is even more important in CWs treating irrigated agricultural water, because this wastewater type is commonly characterized by a strong C/N imbalance. Thus, the low C concentration constrains the nitrate removal via heterotrophic denitrification. The employment of C-rich substrates may help to deal with this C limitation but, yet, there are gaps of knowledge about the interaction between the bed substrate, plants and microorganisms in the CW ecosystem.

A subsurface CWs pilot plant with different bed substrates (gravel, gravel+soil and gravel+biochar) was monitored over the first two years of functioning to examine the effect of substrates on: i) plant growth and bacterial density; ii) plant litter decomposition and the leaf leachate quality as C-source for denitrification, and iii) net nitrate removal. Our results showed the bed substrate is a key factor that affects all the analyzed parameters. Specifically, the lack of C and nutrients supply on gravel beds limited the biota growth and nitrate removal. On the contrary, in gravel+soil and gravel+biochar beds the higher availability of C and nutrients had a positive repercussion on plants and microorganisms and, therefore, on nitrate removal. However, this biochar positive effect is short-lived. The main conclusion of this work is the addition of soil to gravel in CWs is very advantageous.

# Effect of global warming on the metabolic activity of microbial mats of a saline lake in mesocosms

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The aim of this work was to evaluate the effect of increasing temperature and atmospheric CO<sub>2</sub> concentration on the gross primary productivity (GPP) and community respiration (CR) that make up the microbial mats of saline lakes. The study was carried out with mats from the Chiprana Salt Lake (Zaragoza) transferred to 4 mesocosms (one of them control, in darkness without mats) in a controlled environmental chamber where they were exposed to temperatures of 14°C and 26 °C, and CO<sub>2</sub> concentrations of 416 and 1000 ppm. The initial hypothesis was that increasing the CO<sub>2</sub> concentration would increase the GPP of the mats and net community primary productivity (NCP), while increasing temperature, although it would also increase the GPP, would produce a greater increase in the CR, so that the NCP decrease. However, the results obtained differ from those expected. The CO<sub>2</sub> increase produced higher GPP and CR, but NCP was higher at the lower CO<sub>2</sub> concentration, as CR was higher with increasing CO<sub>2</sub>. On the other hand, temperature showed a negative effect on both variables (GPP and CR), with NCP being higher at 14°C than at 26°C. Due to the small sample size, these results are not extrapolable to other conditions of increased T<sup>a</sup> and CO<sub>2</sub>, nor to other types of microbial mats, but they point in an unexpected direction regarding the consequences of global warming on this type of microbial mats communities.

# The power of joining: Preferential lateral inflows are biogeochemical hot spots along Mediterranean fluvial networks

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Headwater streams play a critical role in global biogeochemical cycles because they transport, transform and retain large amounts of carbon, nitrogen and phosphorus. Yet biogeochemical fluxes within streams are poorly constrained, partly because of extreme spatial variability (patchiness) in water chemistry. We argue that this patchiness is partially generated by tributaries and discrete groundwater inputs that have different microbial communities, element concentrations and organic matter composition than the receiving streams. As a result, the junction of these preferential lateral inflows and streams can be biogeochemical hotspots in fluvial networks that are relevant to water chemistry and broader-scale biogeochemical fluxes. The project INHOT is the first explicit and comprehensive assessment of how preferential lateral inflows shape stream carbon, nitrogen and phosphorus dynamics in Mediterranean fluvial networks. By combining digital elevation models and radon analyses, we demonstrated that both tributaries and discrete groundwater inputs are widespread in three Mediterranean headwater fluvial networks with contrasting hydrology. In the next steps, we will perform synoptic surveys to analyze the microbial community composition and water chemistry of preferential inflows; as well as assess the biogeochemical responses of the receiving streams under different hydrological conditions (low vs high flow). Overall, this understanding is critical to create adaptation strategies to global change in fluvial landscapes, as well as to develop new guidelines for the conservation, management and sustainability of these essential ecosystems of our natural capital.



# High-mountain aquatic ecosystems of Sierra Nevada as natural purifiers of reactive nitrogen

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The intensification of the global N cycle implies an increase in atmospheric inputs of reactive nitrogen (Nr) to high altitude ecosystems. However, a seasonal and interannual monitoring of the high mountain lakes of Sierra Nevada (mainly La Caldera, reference lake) shows a consistent decreasing pattern of nitrate concentration ( $\text{NO}_3^-$ ) in the water column during ice-free periods of the last decades. This seasonal pattern is explained by denitrification, quantified as nitrous oxide ( $\text{N}_2\text{O}$ ) emission, and driven by an abundant and diverse community of denitrifying microorganisms in the sediments. Therefore, denitrification acts as a self-purifying mechanism for removing nitrates in high Mediterranean mountain lakes. In further investigation, we found that La Caldera, compared to its downstream river ecosystem, is more efficient at removing Nr as  $\text{N}_2\text{O}$  and molecular nitrogen ( $\text{N}_2$ ) or as  $\text{N}_2$  alone (“clean” Nr removal). These efficiencies are determined by abiotic factors of the sediments, mainly the concentration of  $\text{NO}_3^-$ , and by biotic factors, highlighting bacteria capable of carrying out complete denitrification (they contain the *nosZI* gene) or partial (they contain the *norB* gene), and secondarily, nitrifying archaea. In ongoing research, we study whether abrupt changes in atmospheric Saharan dust intrusions between consecutive years affect Nr removal through  $\text{N}_2\text{O}$  and  $\text{N}_2$  emissions and the microbial diversity of a selected set of Sierra Nevada lakes.

# Organic matter accumulation and hydrology as drivers of greenhouse gas dynamics in newly developed artificial channels

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Artificial channels, common features of inland waters, have been suggested as significant contributors to methane (CH<sub>4</sub>) and carbon dioxide (CO<sub>2</sub>) dynamics and atmospheric emissions, however the magnitude and drivers of their CH<sub>4</sub> and CO<sub>2</sub> (diffusive and ebullitive) emissions remain unclear. They are characterized by reduced flow, compared to the donor river, which results in suspended organic matter (OM) accumulation. We propose that in such systems, hydrological controls will be reduced and OM accumulation will control emissions by promoting metabolism and CH<sub>4</sub> production and thus emissions. Here, we monitored summertime CH<sub>4</sub> and CO<sub>2</sub> concentrations and emissions on six newly constructed river-fed artificial channels, from bare riparian mineral soil to lotic channels under two distinct flow regimes. Chamber-based fluxes were complemented with hydrology, total fluxes (diffusion+ebullition) and suspended OM accumulation assessments. During the first 6 weeks after the wetting, inflowing riverine water dominated the emissions over in-channel contributions. Afterwards, a substantial (≥50% of the channel's volume) accumulation of riverine suspended OM, boosted in-channel methane production and lead to widespread ebullition 10x higher than diffusive fluxes, regardless of flow regime. Our finding suggests ebullition as dominant pathway in these anthropogenic systems, and thus their impact on regional methane emissions might have been largely underestimated.

## Hotspots for CO<sub>2</sub> efflux from a saltpan's biofilm

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Our study is located in a saltpan in Marchamalo (Murcia, Spain), which is being restored after 28 years of disuse. In one of the evaporator tanks we observed the formation of a thick biofilm covering the whole surface. This biofilm presented convexities which held air inside them. We had the occasion to measure the CO<sub>2</sub> contained beneath the biofilm in two sampling campaigns. We proceeded placing a soil respiration chamber on top of the convexities connected to an infrared gas analyzer and making an incision in the biofilm layer, forcing the CO<sub>2</sub> flux into the respiration chamber. We estimated the CO<sub>2</sub> contained under the biofilm mat, being  $14.64 \pm 6.37$  g in spring and  $3.23 \pm 1.95$  g in autumn. This difference could be given because of the sediment water saturation level. Thus, in autumn the sediment under the biofilm was inundated leaving less pore space to hold gas in it. Also, sediment temperatures in Spring were significantly higher ( $p < 0.05$ ) leading to an increase in the CO<sub>2</sub> production. To assess whether the elevated CO<sub>2</sub> emission was due to its accumulation in preferential areas we also measured CO<sub>2</sub> fluxes next to the convexities. The mean efflux values in the vicinities of the biofilm's domes were  $0.42 \pm 0.08$  g m<sup>-2</sup> h<sup>-1</sup> in spring and  $0.003 \pm 0.03$  g m<sup>-2</sup> h<sup>-1</sup> in autumn, whereas the domes emitted  $167.03 \pm 215.04$  g m<sup>-2</sup> h<sup>-1</sup> in spring and  $14.16 \pm 19.64$  g m<sup>-2</sup> h<sup>-1</sup> in autumn.

# OS3

## Community Ecology



## Macroinvertebrates communities in low order isolated streams

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River metacommunities create dynamic assemblages that are more or less diverse depending on extinction and migration rates of species. Following MacArthur and Wilson's framework for island biogeography, we can anticipate that small streams that flow directly into the sea and are, thus, unconnected to large hydrological networks, should be less diverse than similar size streams that flow to larger rivers. They should, moreover, be dominated by taxa that are either very resistant to perturbations or that have high migration capacity. The study area includes various coastal regions in the Basque Country, focusing on specific sites representing isolated streams, streams directly flowing into the sea, and streams connected to larger rivers, with minimal variation in the rest of environmental variables. Preliminary results show substantial structural disparities in macroinvertebrate communities between the two types of systems, with isolated streams hosting a lower number of invertebrate taxa compared to connected counterparts. Additionally, the structure of communities largely differed between both types of sites, with isolated streams exhibiting higher relative abundances of freshwater amphipods and molluscs. These findings emphasize the vulnerability of isolated streams and urges the need for stronger conservation efforts when trying to protect their biodiversity.

**Keywords:** Macroinvertebrate communities, isolated rivers, connected rivers, community composition, biodiversity.

# Uncertain results on potential photosymbiotic activity in the valves of the freshwater ostracod *Eucypris virens*

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Crustaceans exhibit a wide range of organic pigments with different colorations and functions. However, little is known about the function of the characteristic green pigmentation that the podocopid ostracod *Eucypris virens* exhibits in its valves. In the present work, we hypothesize about the potential presence of photosynthetic symbionts or pigments in relation to this green colour. To test this hypothesis, we performed different experiments with the expectation of a reduction of the oxygen consumption rate and an increased uptake of <sup>13</sup>C-enriched inorganic carbon in conditions of light vs. darkness, using anaesthetized ostracods. Moreover, we used epifluorescent microscopy to check for the presence of autofluorescent pigments in the valves, so as different molecular analysis techniques to detect the presence of possible photosynthetic organisms in the ostracod microbiota. Our results on microscopy and molecular analyses, the <sup>13</sup>C incubation experiments and some of the respiration experiments show that the presence of photosynthetic symbionts in *Eucypris virens* might be plausible, even though some of the respiration experiments performed do not reflect any signs of photosynthetic activity, thereby suggesting an urge for future research aimed at clarifying these inconclusive results.

# Response of pond macroinvertebrates to a supra-seasonal drought: resilience of the community

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Summer droughts are natural events in Mediterranean steppe ponds causing water level fluctuations and, eventually, desiccation (temporary water bodies). In the last decades, the frequency of extraordinary (supra-seasonal) drought events has increased, even causing permanent ponds to dry out. This study describes the effects of a drought event (year 2017) on the macroinvertebrate community of a Mediterranean steppe pond which exceptionally dried out. The immediate effect (first year after drought) was a sharp decline in the abundances of several taxa (particularly, *Cloeon*, but also Coenagrionidae and some Trichoptera). Four years after drought, abundances of most of the groups had recovered and richness was close to pre-drought values.

# The planktonic community of three lakes in Lagunas de Ruidera Natural Park (Central Spain) during a summer and winter period

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Lagunas de Ruidera Natural Park is a protected area located in a semiarid region of central Spain (Castilla-La Mancha). The Natural Park encompasses a karst system formed by sixteen interconnected fluvial lakes. It is one of the most important wetlands in inner Spain and has a great impact on the regional biodiversity. Despite its importance, information about plankton in Ruidera lakes is scarce. Thus, we have studied the main plankton constituents (phyto- and zooplankton) of three lakes (Conceja, Colgada and Cueva Morenilla), during winter and summer 2022. Plankton was sampled with plankton nets and hydrographic bottles in the vertical profile. Main limnological variables and the concentration of nutrients and photosynthetic pigments were also measured. Previous studies showed different trophic status in the selected lakes. However, in our study, all lakes presented low nutrient and chlorophyll a concentration. Phytoplankton community was composed of 38 species, most of them belonging to Cyanophyta (9), Chlorophyta (8) and Dinophyta (7). Zooplankton community had 22 species consisted of rotifers (15), cladocerans (6) and copepods (1). Some dominant and frequent species were *Keratella cochlearis*, *K. quadrata*, *Polyarthra vulgaris* and *Bosmina longirostris*. Phytoplankton and zooplankton species richness varied among lakes and seasons, although abundances were generally low. Through a multivariate analysis, relationships among environmental variables and plankton species were detected. This work represents an effort to characterize the plankton within Ruidera lakes and highlights the need for more research along a spatial and temporal gradient to understand how the hydrologic conditions can impact the plankton community.



# Larval habitats of the sibling mosquitoes *Culex pipiens* s.l. Linnaeus, 1758 and *Culex torrentium* Martini, 1925 (Diptera: Culicidae) in temperate climate areas of northwestern Spain

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*Culex pipiens* s.l. Linnaeus, 1758 and *Culex torrentium* Martini, 1925 are considered sibling species since they differ only by a few subtle morphological characteristics. Both are of health interest as potential vectors of West Nile Virus (WNV), Sindbis and Ockelbo virus. In Europe, these species usually occur together and share similar ecological habitats, but further studies are necessary to clarify their population structure. The larval biotopes of these species have been characterized in temperate climate areas of Galicia (NW Spain) through a study carried out in 333 water bodies between May and October 2021 and 2022. The standardized dipping technique (350 mL dipper) was used to collect mosquito larvae. The specimens were morphologically identified and stored in 70% ethanol at the Laboratory of Aquatic Entomology in the University of Vigo (Galicia, Spain). Out of 5332 captured mosquitoes, 48% were identified as *Cx. pipiens* s.l. (n= 2556) and 12% as *Cx. torrentium* (n= 641), being present in 29% and 13% of the study area, respectively. They have been observed sharing breeding sites with other mosquito species, but they mostly occurred together, especially in artificial containers. *Culex pipiens* s.l. has also been found breeding in ponds, lagoons and puddles; while *Cx. torrentium* has shown more affinity for rockpools and no presence in lagoons. Although both species exhibit preference for breeding waters with similar physical-chemical parameters, *Cx. pipiens* s.l. has considerably higher tolerance ranges. The greater adaptability of this species to different water characteristics may largely explain its wide distribution in Galicia.

**OS4**

**Monitoring, Management,  
and Restoration of  
Aquatic Ecosystems**



# Flood risk management with Nature-Based Solution: Hidronet Baix Ter, as a case of study

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The Baix Ter alluvial plain suffered cyclical flooding from historical times. This is principally due to the Mediterranean seasonal rainfall and to the geomorphology of the area. In recent decades, the occupation and urbanization of the territory has increased the risk of flooding in urban areas and, as a consequence, the danger for the inhabitants.

The HidroNet Baix (HBT) project aims to reduce the risk of flooding in three neighbouring municipalities: Ullà, Gualta and Torroella del Montgrí (Girona, Spain) located in the Baix Ter alluvial plain, as a pilot study to later expand to the rest of the urban areas of the Ter river plain.

The HBT project applies Nature-Based Solutions (NBS) and it proposes the generation of temporary ponds in strategic areas and the restoration of riparian buffer strips. It will attenuate the impact of the extraordinary rainfall, storing and slowing the speed of stormwater, that existing drainage systems do not have the capacity to absorb.

This type of NBS actions also prevent soil erosion and reduce the load of nutrients and pollutants that reach the receiving aquatic ecosystems, thereby improving the ecological state of the aquatic systems that is part of the "Parc Natural del Montgrí, les Illes Medes i el Baix Ter".

Beneficial effects are expected both on human population and regarding aquatic biodiversity, since the creation of temporary ponds promotes the presence of habitats with hygrophite vegetation and the associated populations of amphibians and aquatic invertebrates.

# The importance of local drivers in community assemblages in Mediterranean salt marshes

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Understanding the role of colonization in community assembly after habitat restoration has mostly focused on three main drivers: species traits (i.e., dispersal ability), connectivity and environmental filtering. However, in certain ecosystems, hydrology may also play an important role in driving assembly dynamics. An example are coastal salt marshes, in which hydrology can determine environmental constraints such as salinity and nutrient concentration, but also connectivity due to confinement-flooding dynamics. In these cases, we hypothesize that local drivers, such as hydrology, could strongly determine community assembly in new or restored habitats. This study focuses on a restoration project carried out in “La Pletera” salt marsh in the Baix Ter Natural Park (NE Catalonia), where new lagoons were created with the aim of restoring the ecological functionality of the ecosystem. To assess the success of the restoration and the main drivers influencing community assembly, we sampled the macroinvertebrates from ten newly created and three already existing lagoons in three occasions: the first year after restoration (2016), the following year (2017), and six years later (2022). We studied changes in community composition in space and time, comparing lagoon age (new vs old) and hydrology (high connection vs low connection) since taxonomic and functional perspective. Our results showed that community composition changed accordingly to hydrological patterns during surveys, emphasizing the importance of considering it when assessing restoration success. These findings highlight the need to move beyond traditional perspectives and integrate local drivers into restoration strategies.

## Building climate resilience in streams and rivers: The AQUADAPT Initiative

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Southern Europe faces daunting climate future projections, with Portugal anticipating a potential +5°C rise in air temperature by 2100. Such shifts threaten to disrupt thermal and hydrological regimes, escalating the occurrence, intensity, and duration of extreme events, as droughts and floods. Consequently, the sustainable development of Portugal's inland region will inevitably depend on the ability to adapt to such climate-related changes. The AQUADAPT project aims to promote the resistance and resilience of stream and river ecosystems to climate change, through risk assessment and the construction of adaptation tools. We forge a robust high-resolution monitoring and warning system through modelling, forecasting, and spatial planning, particularly in tree case studies: –Sabor river, Ponsul river and Cardeira river. Our approach also implements and tests Nature-Based Solutions (NBS) in riverine restoration plans aiming to enhance resistance and resilience of streams and rivers.

The AQUADAPT gathers collective efforts of academia, governmental bodies, and industry partners, to provide stakeholders with skills and adaptive strategies, paving the way for a more resilient and sustainable future. Additionally, our project emphasizes knowledge dissemination and capacity-building efforts to ensure widespread adoption of sustainable practices.

# Macrolitter as a new tool for the ecological evaluation of the Leça River

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Rivers can be considered the primary source of vital ecosystem services for populations. However, river ecosystems encounter various anthropogenic threats, including macrolitter, with its impacts remaining inadequately understood. This study aimed to: 1) evaluate the ecological status of Leça River (northern Portugal) based on assessment elements defined by the Water Framework Directive (WFD), and 2) assess the viability of macrolitter characterization as a new ecological evaluation tool. WFD elements such as biological (benthic macroinvertebrates) and physical and chemical parameters were used to evaluate the ecological status of 7 sites (P1 to P7) along the river in three seasons (autumn, winter, and spring). Macrolitter was collected in the riverbank of each site, being quantified, categorized, and classified using OSPAR, J-Code, and TSG-ML reports. Preliminary results showed that WFD elements declined downstream. Macrolitter was abundant in all sites, with the items collected belonging to 72 litter categories. P6 exhibited the highest macrolitter abundance, aligning with the lowest ecological classification recorded. Our findings illustrate a downstream deterioration of the river, aligned with a heightened presence of macrolitter. This correlation suggests that macrolitter can serve as an indicator of deleterious anthropogenic activities. Further analysis has to be done to elucidate this relation, be it based on the type of macrolitter, be it based on the fact that, as a river degrades its quality, people tend to look at it as an adequate dumping site for garbage.

# Restoring a Mediterranean RAMSAR wetland by in-flow magnetic separation with a Halbach array configuration

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Although Mediterranean wetlands are hotspots of biodiversity, they are severely affected by eutrophication. Indeed, several internal features (i.e., shallowness and a typically high catchment area to lake area ratio) make Mediterranean wetlands especially sensitive to eutrophication that feedback on their condition. It is important to keep in mind that a large part of the external phosphorus load to these wetlands comes from the discharge of treated wastewater, which, although it meets the standards, represents an excessive input of nutrients. Accordingly, it is essential to develop new technologies for mitigating the problem of eutrophication by reducing phosphorus concentration in point sources that discharges into especially sensitive aquatic ecosystems. In this context, a promising two-step approach involves i.- the use of magnetic particles (MPs) as carriers where phosphorus is specifically absorbed on their surface and ii.- the removal of these MPs using magnetic field gradients. Up to date, the operation of high-gradient magnetic separators in batch conditions has been extensively reported in the literature. However, their understanding/use in flow conditions is still scarcely known. This work describes the design, construction and validation of a novel magnetic field separator for phosphorus removal in Fuente de Piedra wetland. The device involves a magnetizable grid placed inside a circular Halbach array that does not require a power supply. Finite Element Method magnetostatic simulations together with Computational Fluid Dynamics simulations are carried out to optimize the experimental setup using a magnetophoretic dimensionless (Mason) number.

# Changes in benthic diatoms communities from some streams in the Llobregat Basin (NE Spain) between 2015 and 2023

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Diatoms are one of the biological indicators of water quality in aquatic ecosystems. Following the Water Framework Directive, the study of diatom communities is widely used to monitoring rivers by the management entities. Catalan Water Agency monitoring program designs the set of samplings that will be carried out on the rivers in the Catalan River Basin District. From 2015 to 2023 samples of diatoms were analyzed in the Llobregat Basin obtaining the ecological state of the water bodies based in Specific Polluosensitivity Index (IPS). In this study, results of IPS were analyzed to identify streams with significant variance in their quality throughout these years, regarding the physicochemical variables and the diatom community composition. The main objective is to contrast changes in IPS index between years with changes in the diatom communities in these sites, especially in streams classified as Natural River following the ACA guidelines. Quality index lowest values are linked to species from eutrophic habitats which strong pollution. Diatom communities from each site were compared over the years, and it was found that disturbed years presented the most similar community composition represented by *Fistulifera saprophila*, *Craticula subminuscula*, *Sellaphora saugerressi*, and *Nitzschia inconspicua*. In conclusion, species from diatom community in each spot have the same dominance dynamics when facing physicochemical changes even in periods time apart.



# Phytoplankton community temporal evolution in reservoirs of two hydrological basins in the North of Spain

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Phytoplankton is a key indicator of water quality in aquatic ecosystems. In accordance with the Water Framework Directive, the study of phytoplankton communities has been widely incorporated into monitoring networks by management entities. The Bilbao Bizkaia Water Consortium (CABB) provides water to over 1 million people in the Basque Country. Considering CABB's water resources, the main contribution comes from the tandem reservoir system of the Zadorra river basin (Z), supplemented by smaller reservoirs belonging to the Ibaizabal-Nervi3n river basin (I-N). The main objective of this study was to contrast the phytoplankton community assembly processes in eight reservoirs of the CABB supply system for the 2017-2023 period. The biotic approach was based on Kruk's (2010) Morphologically Based Functional Groups (MBFG), for which community patterns were investigated and diversity indices calculated. Considering environmental variables, particularly ionic composition and hydromorphological features underscored climatic differences between basins. Although the overall composition and diversity indices were broadly similar for the phytoplankton communities across all reservoirs, cyanobacteria emerged as a differentiating element between the two basins. *Planktothrix isoethrix* (MBFG-3) was identified as the most relevant bloom-forming cyanobacteria in the I-N basin. Conversely, *Microcystis* species (MBFG-7) outstood in the Z basin. Overall, basin-induced environmental differences seemed to determine the dominance of potentially toxic cyanobacteria species within the phytoplankton communities, which rarely scaled to severe bloom episodes under local trophic conditions.

# Macrophyte communities of south-central Spanish rivers: diversity, indicator species, and application of macrophyte indices for the ecological status assessment

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Over the last two decades, the European Water Framework Directive 2000/60/EC (WFD) has been implemented to assess the ecological status of surface waters using physicochemical, biological and hydromorphological quality elements. The biological quality element (BQE) “aquatic flora” for rivers includes macrophytes and phytobenthos (diatoms). The term “macrophyte” refers to any autotrophic macroscopic organism but also to microscopic algae that form macroscopic aggregates or colonies. Thus, river macrophytes constitute a heterogeneous group made up of very diverse taxonomic groups and levels of organization. In this study we gather data obtained in macrophyte surveys carried out during the years 2001-2014. The studied river network was located at south-central Spain, in the limits of the administrative region of Castilla-La Mancha, including a great part of the Tajo, Júcar, Segura, Guadiana and Guadalquivir river basins. A total of 195 sites were monitored, belonging to 8 national river types. Specimens were determined at genus or species level depending of the taxonomic group; 32% of taxa collected were cyanobacteria, 31% eucaryotic algae, 20% bryophyte and 17% vascular plants of the total taxa. The main objectives of this study were to analyse the macrophyte diversity and distribution in the studied river types, to establish the environmental ranges of indicator species, and to assess the ecological status of rivers using macrophyte indices. Finally, we discuss the effectiveness of macrophytes as tools for assessing the ecological status of river ecosystems.

# Study of water quality in the Mero and Gobia rivers in the vicinity of the experimental farm of the Agricultural Research Center of Mabegondo (CIAM).

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The main objective of this study (2022-2023) was to investigate the water quality status to address compliance needs with the Nitrates Directive through Royal Decree 47/2022, of January 18, on the protection of waters against diffuse pollution caused by nitrates from agricultural sources. The results were used to define key points for future monitoring on the Mabegondo estate to control the entry of nutrients from crops and livestock into the Gobia and Mero rivers that border the estate, and consequently their entry into the Cecebre reservoir, located downstream of both rivers and supplying the city of A Coruña and its surroundings. Higher mineralization was observed in the streams draining the estate, with low levels of phosphorus. Although moderate concentrations of ammonia and nitrate were detected at some stations (especially related to agricultural and livestock activities), these values do not exceed the limit to consider waters affected by nitrates (Real Decreto 47/2022 de 18 de enero, sobre protección de las aguas contra la contaminación difusa producida por los nitratos procedentes de fuentes agrarias). Overall, a moderate to very good ecological status was observed in the Mero and Gobia rivers, with some variations due to specific metrics employed. The livestock and agricultural activity on the estate exert pressure on water quality, especially in the streams draining the estate. Continuous monitoring of nitrate concentrations is recommended to prevent negative impacts on the Cecebre reservoir.

## Knockin' on diatom's DNA metabarcoding door

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Diatom bioassessments have historically relied upon microscopy-based identifications to study the composition and abundance of algal communities. However, it is a fact that these analyses are time-consuming and limited to highly experienced analysts. In contrast, as a paradigm, metabarcoding of water column or biofilm DNA is expected to be a revolutionary tool to overcome inconsistencies in traditional diatom taxonomic identifications. Lower costs, fast turnaround and less technical expertise may spark the interest of water authorities for its implementation in water monitoring programmes. Still, it is important to stress that beyond the benefits, there are challenges related with the development of molecular-based metrics for ecological assessment that need to be resolved first. This study presents a five-year (2018-2023) diatom inventory from 650 water bodies spread throughout the Spanish Iberian Peninsula (Ebro, Guadiana, Miño-Sil, Segura and Tajo River Basin) to find out in a simple way if DNA metabarcoding is currently covering the most common diatoms recorded in our river ecosystems. Taxonomical analysis resulted in a total of 1134 species. After discarding taxa with a total abundance of less than 5% and a frequency of appearance less than 1%, 181 species were crossed with the DNA sequence information stored in the GenBank® database. Results showed that even though 64% of the taxa reviewed have molecular information, there is a 36%, plus another percentage of low-abundant and rare taxa, that are not sequenced up to date and therefore could be a limitation for its application in existing water quality monitoring programmes.

**OS5**

# **Exotic and Invasive Species**



# Invasion of a small peri-urban stream by the exotic New Zealand mud snail

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Invasion by non-native species is one of the greatest threats to freshwater biodiversity and ecosystem functioning. Disturbed ecosystems are more prone to invasion because chronic stress tends to erode ecological resilience, decreasing resistance to invasion over the long term. Among invaders, the New Zealand mud snail, *Potamopyrgus antipodarum*, has spread all over the world except Antarctica but with only nine records in Portugal. In 2004, the New Zealand mud snail was detected in a stream subjected to multiple stressors at relatively low abundances. Four sites were sampled irregularly in time over the next 10 years using a Surber sampler. Over the next 10 years, the cumulative density in the stream increased steadily, conforming to an invasion curve. The average density of the sites was used to calculate the cumulative density in the stream, and simple linear regression was used to test if time significantly predicted those values. The cumulative density in the stream increased steadily during the study period, from a global average across sites of 328 individuals/m<sup>2</sup> in January 2004, to 2100 in May 2009, and 3778 in April 2014. In the regression model, the overall population strongly adhered to a linear fit ( $R^2 = 0.99$ ,  $F_{1,7} = 1301$ ,  $p < 0.00001$ ) with an estimated mean initial size of  $N_0 = 372$  individuals/m<sup>2</sup>  $\pm$  150 (95% CL) and a monthly rate of increase of 28 individuals/m<sup>2</sup>  $\pm$  1.8 (95% CL). With these values, it would take 60 years from 2004 to achieve the carrying capacity predicted elsewhere for European streams

## LIFE INVASAQUA and Public Involvement: effective integrated management of invasive alien species

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Public awareness campaigns are crucial for effective management of Invasive Alien Species (IAS), particularly raising awareness about regulations and problems associated with IAS. Monitoring stakeholders' risk perceptions and behaviors through surveys is key to designing and implementing appropriate communication plans. LIFE INVASAQUA aimed to reduce the introduction and spread of aquatic IAS in the Iberian Peninsula, including the development of tools to improve management and early warning. Raising awareness was one of the keystones of the project, and surveys were created to monitor perceptions and behaviors related to IAS. Bilingual surveys were conducted on the Iberian Peninsula, targeting the general public, decision-makers and other stakeholders. During the project, more than 7,000 responses were obtained, with an increase in knowledge about the project over the last five years, currently standing at more than 60%. Of all the respondents, 56% knew what an invasive alien species was. Over the project period, the number of people who did not know what an IAS was decreased dramatically and there was a sharp increase in the number of people who knew what an IAS was. The results indicate different levels of perception of the impacts of IAS, with a greater awareness of impacts on biodiversity compared to impacts on human health and socio-economic activities. It was possible to disclose an increase in awareness of all IAS impacts caused by the INVASAQUA project actions, diminishing the differences in perception between biodiversity, socioeconomic and human health impacts, revealing the success of the LIFE INVASAQUA project.

# CLICKbaits: Digital tools to reinvent freshwater fish conservation in a changing world

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The Iberian Peninsula is a hotspot of freshwater fish biodiversity. This highly endemic fauna is exceptionally vulnerable to the spread of non-native fish species. Preventing introductions and targeting the early stages of invasion are still the most cost-effective management options. Because these approaches directly depend on people's attitudes and perceptions, insights from an increasingly data-rich world offer an opportunity to understand public interests and provide instruments to raise awareness of the risks posed by non-native fish. The analysis of online data regarding biodiversity allows us to uncover complex relationships between societies and the natural world. The main objective of this study was to identify what drives public interest towards Iberian ichthyofauna by assessing the species' online popularity. To do so, public interest was measured as the sum of Wikipedia page views between 2015 and 2023. Regression analyses were used to relate Wikipedia page views to species' attributes, traits and economic significance. Results showed that online popularity was mainly driven by geographic range, body size and economic interest (i.e., commercial fisheries, aquaculture, and aquarophilia). Additionally, predator, gamefish, and non-threatened species tended to be more popular among Iberian fish. While threatened species lacked public attention, this study highlights a preference towards economically valuable species or attributes that are common in non-native fish. Unveiling the factors underpinning societal interests in biodiversity is a crucial step for improving biodiversity conservation.



# Management actions applied to the invasive American red crab *Procambarus clarkii* in a core site of a Biosphere Reserve

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The aquatic ecosystem of the Abegondo-Cecebre reservoir, which is part of the “Mariñas Coruñesas” Biosphere Reserve, was ecologically transformed with the introduction in the 1990s of the American red crab (*Procambarus clarkii*). Its presence has caused a profound alteration of the aquatic vegetation and its associated fauna over several decades, especially noticeable regarding the reproduction of aquatic birds and amphibians. The Biosphere Reserve has carried out a series of actions to try to regulate this invasive species based on crab removal campaigns, and to assess possible benefits to currently affected species. These actions have taken into account the idiosyncrasy of the activity periods observed for the reservoir population, which, shows a 2-month delay in the activation and hibernation periods due to the effect of seasonal thermal constancy (thermal inertia). Our results were different depending on the removal point (n=2). Catches did not decrease over time around the reservoir dam in contrast to a second sampling and removal point. Amphibian reproduction (in the reservoir is currently very limited (*Hyla*, *Discoglossus*) but it has been verified (both for anurans and newts) in a small temporary pond located closer to the reservoir; and precautionary protocols have been established to prevent and detect its eventual colonization by crabs. A replicate of these management actions have also been applied at Beche reservoir (10 km from Cecebre), a much smaller water body, that will allow future comparisons of results.

# Thermal Tolerance of the New Zealand Mud Snail *Potamopyrgus antipodarum*

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*Potamopyrgus antipodarum* is a small freshwater snail native to New Zealand that has become one of the world's most successful invaders. Several characteristics contribute to this success, namely its asexual reproduction and high reproductive output, and its suggested wide tolerance to environmental factors. Assessing the tolerance of invasive species is, therefore, an important step towards predicting their invasive capacity in climate change scenarios.

This work aimed to assess the thermal tolerance of *P. antipodarum* and to evaluate how increasing temperatures can affect its reproduction, feeding behaviour and physiological fitness. Laboratory experiments were carried out in which the snails were exposed to constant temperatures of 20 and 24°C and also to a heat wave (temperature increase from 20 to 28°C). The following endpoints were then measured: a) the number of embryos produced per female, b) feeding rate, c) various biochemical markers related to physiological stress and energy metabolism.

The results showed that a temperature increase of 4°C had no effects on any of the endpoints evaluated compared to the control treatment. However, the heat wave significantly reduced the snail's reproductive output and feeding rates. Exposure to a transient heat wave also induced slight but significant reductions of aerobic metabolism (electron transport system) activity and a reduction of total glutathione content indicating activation of oxidative stress responses.

In short, *P. antipodarum* proved to be quite tolerant to temperature, and population level responses are not to be expected in warming scenarios although its abundance may decrease in the case of extreme events.

## One after the other: Inadequate policies applied to the exotic turtle market

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The commercial popularity of certain pet species generally predicts their potential invasiveness, with highly traded species being more likely to be introduced. Turtles are among the most traded, demanding close surveillance to prevent their introduction into non-native settings. In Portugal, various invasive turtle species have been recorded in the wild, emphasizing the importance of good monitoring programs. This study is the first official record of *Mauremys sinensis* in Portugal. Despite being critically endangered in its native range, *M. sinensis* has invasive tendencies outside of its native range. The discovery of two individuals—a mature male and a juvenile—in a lake near Évora, alongside native *Mauremys leprosa*, indicates an alarming invasion. Furthermore, the GBIF database revealed 14 citizen science reports of this species from 10 municipalities in Portugal since 2021, from the north to the south of mainland Portugal and even reaching Madeira Island. Invasive turtles pose numerous problems to native species, including competition, predation, disease transmission, and hybridization. Despite legislative attempts, the pet trade remains the primary route for these invasions, and it continues to operate without effective regulation. This study underlines the importance of comprehensive solutions, including whitelists and blacklists, to combat the invasive species pet trade, protect local ecosystems, and prevent future imports of species such as *M. sinensis*.

# Study on the populations of the Asian clam *Corbicula fluminea* (Müller, 1774) in the middle section of the Júcar River

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The Asian clam *Corbicula fluminea* (Müller, 1774) was detected recently in the middle reach of the Júcar River. This study updates the distribution of the species in the Júcar river from the Alarcón reservoir to the Molinar reservoir, and additionally, it analyzes the structure of three populations. Between May and October 2023, a total of 28 sites were visited to check for the presence/absence of the Asian clam. Their presence was detected in seven sites, selecting three of them with well-developed populations to carry out quantitative sampling. Bivalve samples were taken with a Surber type net with a 0.1 m<sup>2</sup> sampling area, equipped with a Nynet net with a 250 µm. Three replicates were taken in each site, in areas with different depth, substrate and current velocity, and samples were fixed in hermetic bags with ethanol 80%. At the same time, physicochemical data of the section were taken, such as pH, dissolved oxygen, water temperature and electrical conductivity. In the laboratory, specimens captured alive (closed valves) and dead (open empty valves) were counted, and the size of all individuals collected was measured (length of the anterior-posterior axis of valves). The results of density, biomass, and size structure of *Corbicula* populations in each site were analyzed, as well as its relation with physicochemical and hydromorphological variables.

**OS6**

**Biogeography and  
Evolution of Species**



## Potential distribution of minute moss beetles (Coleoptera: Hydraenidae) in Brazil

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Hydraenidae is a cosmopolitan family of small water beetles, commonly known as “minute moss beetles”. The family is one of the most speciose among water beetles, with more than 1,600 described species arranged in 40 genera worldwide, of which four are currently recorded in Brazil: *Adelphydraena* Perkins, *Hydraena* Kugelnann, *Ochthebius* Leach and *Parhydraenida* J. Balfour-Browne. In this study we analyzed the potential distribution of these four genera in Brazil using ecological niche modeling (ENM). Within Brazil, Hydraenidae genera exhibit distinct distribution patterns. By utilizing current records and model projections, we have delineated areas of habitat suitability for each genus. Our results indicate that *Hydraena* and *Ochthebius* have a wide potential distribution, covering a large part of the country. Otherwise, *Adelphydraena* would be restricted to the Amazon and *Parhydraenida* to the mountains of the Brazilian Shield. The knowledge on the Brazilian hydraenid fauna is still very poor. There are significant knowledge gaps, and many areas have been under-sampled, which should be addressed in future studies.

# A time-calibrated 'Tree of Life' of aquatic insects for knitting historical patterns of evolution and measuring extant phylogenetic biodiversity across the world

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The phylogenetic history of aquatic insects remains largely elusive, and our understanding of their chronology is fragmentary and incomplete at best. Here, after gathering a comprehensive data matrix of 3,125 targeted rRNA and protein-coding gene sequences from nine independent gene portions, we built a well-supported time-calibrated phylogenetic tree comprising almost 1,200 genera that represent a large proportion of extant families of dragonflies and damselflies (Odonata), mayflies (Ephemeroptera), stoneflies (Plecoptera), and caddisflies (Trichoptera). Molecular dating using the birth-death model of speciation, with a lognormal-relaxed model of sequence evolution informed by transcriptomic constraints, suggested that (i) dragonflies and damselflies first radiated approximately 220 million years (Ma) ago and most extant lineages thrived independently after the Triassic–Jurassic (Tr–J) extinction event; (ii) mayflies underwent bursts of diversification during the Cretaceous; (iii) ancestral divergence separating the stonefly suborders Arctoperlaria and Antarctoperlaria was consistent with geographical isolation after vicariant fragmentation and tectonic splitting of the supercontinent Pangaea around 170 Ma ago; and (iv) the most recent common ancestors of caddisflies extended back to the time of Pangaea, supporting the earliest offshoot of the 'retreat-making' Annulipalpia. Our 'Tree of Life' of aquatic insects also resolved shallow phylogenetic relationships related to the convergent evolution of exophytic oviposition in dragonflies or the Jurassic origins of the burrowing lifestyle in mayflies. The tree is available for download from the Zenodo repository (DOI: 10.5281/zenodo.7990254) as a Newick file, a format that can be easily viewed in freely available programs like R, Dendroscope, and FigTree.

# Evolutionary trajectories of body size over geological time in world's extant Odonata and Trichoptera (Insecta)

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Body size is a fundamental property of organisms, and documenting the evolutionary trajectories of body size variation is an important endeavour for the biogeosciences alike. For instance, Cope's rule attempts to summarize body size trends over evolutionary time suggesting that species tend to increase in size from a smaller ancestor. In this study, we investigate global patterns of body size evolution using a recent timeline for different monophyletic groups of extant aquatic insects (i.e., dragonflies, damselflies, and caddisflies). We compiled body size data for world's extant odonate and trichopteran species in our database using public datasets and published articles and books. Specifically, we estimated ancestral sizes for each node of our best-scoring time-calibrated phylogeny and plotted the trajectory of body size evolution as a 'traitgram'. Deep-time patterns of body size evolution of damselflies, dragonflies, and caddisflies were found to be complex and non-linear. This result suggests that selection for large body size in aquatic insects can eventually be counterbalanced by opposing evolutionary forces.



# The first Ralfsia-like brown alga (Phaeophyceae) in freshwater streams: records from Spain

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A novel brown alga was found inhabiting freshwater streams in Spain. Preliminary molecular analyses indicate that it represents a new lineage of the order Ralfsiales at the genus and possibly the family level. This is the first report of a non-marine species for this order. It is an epilithic crustose species, dark to light brown, sometimes with yellowish-orange bands, soft but hard texture. Colonies are usually orbicular with a radial growth pattern and dichotomous fan-like branching of filaments. The prostrate filaments bend upwards as they grow, building circular or semilunar protrusions in concentric bands that can overlap at different heights from the substrate. Old colonies can reach up to 0.5 cm thick and more than 30 cm in diameter. The new species mainly inhabits 1st to 3rd order, low-medium altitude, mineralized, calcareous streams and rivers. Most of them were located at the mountainous Cantabrian coast in Northern Spain, although the species was also recorded in southeastern Spain. The streams were well oxygenated and oligo-mesotrophic, with medium to high current velocities. It inhabits stony substrates of riffles and runs, where it may cover from small to big stable boulders and rock outcrops. This discovery also raises questions about the mechanisms underlying the adaptation of brown algae to freshwater environments, particularly in terms of salt tolerance. In conclusion, the finding of this non-marine ralfsial alga represents a paradigm shift in our understanding of brown algae diversity, evolution, and ecological adaptations.

# First genome assembly of the euryhaline ostracod *Cyprideis torosa* using HiFi long-reads

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Despite the increasing availability of genome resources from a wide range of aquatic invertebrates, there is still a lack of accurate genome assemblies for certain taxa, such as ostracods. In particular, only three draft genomes, based on Illumina short reads, are currently available for Ostracoda. *Cyprideis torosa* is a model organism in ecology and paleoecology due to its wide tolerance to salinity and hypoxia, and morphological variation. Here, we present the first genome assembly obtained through highly accurate (HiFi) long-read sequencing. We extracted the DNA from a pool of ca. 100 *C. torosa* individuals collected from Salines de Santa Pola Natural Park. We applied a PacBio SMRT sequencing protocol, obtaining three million HiFi reads. After cleaning the reads from adapters and bacterial contamination, we built a genome assembly using Hifiasm. The new assembly includes 10,519 contigs, an N50 of 79kb, and a BUSCO score of 88%. It represents a significant improvement with respect to the previous draft genome of *C. torosa*, with a tenfold reduction in number of contigs/scaffolds and larger contig length (N50 = 19kb in the previous assembly), although our BUSCO score is slightly lower. The new assembly will be fundamental to investigate transposable element evolution and the genetic basis of osmoregulation and oxygen tolerance in ostracods.

# Genetic Characterization of Freshwater Bivalves in the Area of the Alqueva Multipurpose Project

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The Alqueva MultiPurpose Project (EFMA) is a structural project in southern Portugal, which ensures the availability of water to an area of approximately 10,000 km<sup>2</sup>.

EFMA involves a water transfer from the Guadiana to the Sado basin, and one of the potential environmental impacts is the passage of freshwater mussels from the Guadiana to the Sado water bodies, by means of its larvae and early life stages, and the consequent risk of miscegenation and hybridization between populations from the two basins. Therefore, establishing the baseline situation of the gene pools characteristic of native populations of both basins is of particular importance.

In addition to the set of samples collected specifically in the framework of the water transfer risk evaluation, this work benefited from a collaborative work being carried out to share samples and resources with the project “MUSSEFLOW: Host-dependent evolution, ecology and conservation of freshwater mussels under varying hydrological consequences of climate change” (funded by FCT), allowing to obtain more complete and reliable data.

The results demonstrate that the Guadiana populations are genetically distinct from the populations in the bordering basins (Tagus, Sado and Mira), although morphologically similar. Moreover a strong differentiation between subpopulations in separate rivers within the Sado and Guadiana basins was recorded.

The genetic differentiation between populations may have been conditioned by the paleogeomorphology of the aquatic systems they colonized thousands of years ago and, more recently, by the availability and mobility of their hosts and the characteristics of the habitats where they occur.

**OS7**

**Ecology of Wetlands,  
Lakes, and Estuaries**



# Unveiling the hidden world: zooplankton in peatlands of O Forgoselo (Galicia, NW Iberia): preliminary results

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Peatlands are fragile ecosystems highly sensitive to climatic conditions and anthropic pressure. This is especially concerning in Southern Europe, where their occurrence is scarcer and water stress is more pronounced; therefore, there are many initiatives to establish protection areas and promote their preservation. Peatland conservation and restoration in the current context of global change it is also important both, in preventing the release of stored carbon to the atmosphere and in providing habitats for plant and animal species, contributing to biodiversity conservation at the regional level. Zooplankton is one of the most widely used bioindicators to assess ecological status in wetlands because it is highly sensitive to changes in water chemistry, pH, and trophic conditions. However, the composition and ecological dynamics of this group in Iberian peatlands is poorly known. For this purpose, the composition of zooplankton together with water chemistry, limnological parameters and phytoplankton in different microhabitats (i.e. meadows, ponds) of five minerotrophic acid peatlands of the O Forgoselo peatland complex (Galicia, NW Iberia) have been studied. Under values of pH between 3.5 and 6.5, and electrical conductivity between 28 and 167  $\mu\text{S cm}^{-1}$ ; zooplankton is dominated by acidophylic species as the rotifers *Brachionus sericus*, *Keratella serrulata* or *Lecane stichaea* and cladocerans of the Genus *Alonella* and *Chydorus*. Rotifers presented more than 20 species, with densities around 30 ind/l; cladocerans and copepods were also represented by at least 15 species and densities around 20 ind/l. We have found differences in the zooplankton composition among the studied microhabitats and peatlands.

# Development of a semi-empirical algorithm for the estimation of carotenoid concentration in the Albufera coastal lagoon (Valencia, Spain) using remote sensing

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Carotenoids, accessory pigments of some classes of phytoplankton, play a crucial role in the modulation of light absorption, transferring the captured energy to chlorophyll-a, the main photosynthetic pigment. In August 2023, the Albufera of Valencia experienced an unusual shift to an orange-red coloration due to a strong increase in the concentration of carotenoids, more pronounced than that of chlorophyll-a. This change coincided with exceptional meteorological conditions characterized by low rainfall and high temperatures. The need to monitor these changes motivated the development of a semi-empirical algorithm based on Sentinel-2 satellite images to estimate the concentration of carotenoids in the Albufera, using field data collected between 2018 and 2023. The spectral index  $R783/(R490+R560)$  gave the best results, with an nRMSE of 12.76% in the validation. Algorithms using the R740 band also showed good performance. These new algae have different spectral characteristics from classical algae, with a higher correlation in the red-edge region close to the NIR. Turbidity could influence these differences, such as the blue and green absorption attributed to carotenoids. These results have potential synergies with microscopic and molecular approaches, allowing a more complete understanding of changes in phytoplankton populations. The developed algorithm will facilitate past, present, and future satellite monitoring of these events and help identify possible causes and ecological implications.

# Assessing the effect of Spadefoot toad tadpoles (*Pelobates cultripes*) on aquatic macrophytes through field and mesocosm experiments

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Anuran tadpoles can have important effects on aquatic systems, affecting decomposition, periphyton abundance or nutrient cycling. Nevertheless, how they interact with aquatic macrophytes has remained relatively understudied. Focusing on the temporary pond network of Doñana National Park (Southwestern Spain), we evaluated how the herbivorous tadpoles of the Spadefoot toad (*Pelobates cultripes*) affected the biomass of three different macrophyte species (*Ranunculus peltatus*, *Myriophyllum alterniflorum* and *Callitriche brutia*), through mesocosm and field experiments. In mesocosms, we tested the effect of two tadpole densities on plants. Within a natural pond, a field experiment consisted of exclusion (no tadpoles), inclusion (fixed tadpole density) and open (natural tadpole densities) plots. In the mesocosms, tadpoles significantly reduced plant biomass of the three plant species. Interestingly, the effect was independent of the larval density in the mesocosms, with tadpoles at lower densities growing larger and exerting a similar herbivory impact compared to higher density, when tadpoles that were smaller due to competition. In the field experiment, *Ranunculus peltatus* almost disappeared when tadpoles were present, whereas *Myriophyllum alterniflorum* biomass only decreased in the inclusion plot. Tadpoles of *P. cultripes* have a strong impact on the biomass of aquatic macrophytes, but do not affect all plant species equally. This consumption of macrophytes likely has cascading effects on the whole system, influencing primary production, spatial complexity and interactions with other organisms. Moreover, we highlight the importance and complementarity of mesocosms and field experiments, the former allowing more experimental manipulations, while the latter provides more realistic representations of the natural processes.

# Harriers as sister umbrella species in agriculture-wetland ecosystems

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Species-specific and fence-focused approaches to wetland conservation can lead to neglect of important ecological linkages between these ecosystems and neighboring agricultural ecosystems. The prospects of climate change call for a more focused research effort in these ecotones. Montagu's and marsh harriers (*Circus pygargus* L. and *C. aeruginosus* L., respectively) are morphologically and ecologically similar umbrella species that live in sympatry. Although both harriers would use common resources in agricultural-wetland ecosystems, in a competition scenario, they do not share the same trophic niche. Previous findings inspired this work, namely (i) the control of long-term population dynamics of the marsh harrier by habitat changes in a semi-arid Mediterranean wetland, and (ii) the medium-term spatial synchrony of the same species between different temporal and shallow wetland locations. Here we explore habitat controls (land use, wetland vegetation, meteorological variables, etc.) on the common occurrence or not of Montagu's harrier and marsh harrier, as well as their implications for an integrated conservation effort at the territory scale. We pay special attention to the social issues of conservation in depopulated rural areas, where we have recorded important changes in crop intensification and industrial meat production.



# Pollen and non-pollen palynomorphs in the modern sediment of Lagoa dos Nenos, Islas Cíes (Ría de Vigo, NW Iberia)

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Active coastal lagoons are unstable geological formations that may be considered as ephemeral sedimentary systems formed on coastal areas during the Holocene sea-level rise. They are usually connected to other ecologically valuable coastal wetlands, and due to their configuration and vulnerability are listed as priority habitats. Currently are subject to progressive threatened because of human interaction, sea level rise and eutrophication.

Modern sediments in these lagoons are important deposits of pollen and non-pollen palynomorphs (NPP), their main contributions coming from pollen rain and water transport. This evidence allows us to study the representation, productivity, dispersion, and abundance of the different local wetlands and regional plant communities. Therefore, modern pollen in this type of fluvio-marine surface sediments can be used in the calibration of models for fossil data.

The Cíes Islands archipelago is located at the mouth of the Ría de Vigo. The Lagoa dos Nenos lagoon is behind the beach and the dune system, all of them forming a barrier-lagoon complex. Different zones of deposition for several pollen and NPP types can be distinguished, due to the configuration of the lagoon, the vegetation composition of its surroundings, and the tides. Particularly, *Erica* pollen can be found in modern sediment, which does not come from local/extralocal vegetation, because this genus is currently extirpated from the islands. The closest formations of this type of vegetation are located at Cabo Home and Cabo Silleiro. The new data obtained suggest that this pollen type is mainly transported by marine currents.

# Daily functional variability of phytoplankton in the littoral zone of a large shallow subtropical coastal lake

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Studies have shown that different phytoplankton functional approaches have particular sensitivities, providing divergent responses between compartments of the same environment. Lakes provide a wide diversity of habitats for aquatic organisms, with the littoral zone of shallow lakes being considered extremely dynamic due to climatic effects and adjacent contributors. This study aimed to understand the functional response of phytoplankton in the littoral zone of a shallow lake, using three functional-based classifications: Reynolds functional groups (RFGs); morphology-based functional groups (MBFGs); and life forms (LFs). Data were collected daily for 25 consecutive summer days (2016) at two marginal sites of Mangueira lake, a polymictic and oligo-mesotrophic subtropical system in southern Brazil. The CNF life form dominance was registered, comprising mostly colonial cyanobacteria species. The MBFG classification demonstrated stability during the studied period: Group VII, represented by mucilaginous colonies, prevailed at both sampling stations. The RFGs J and K were predominant, together reaching values near 50%. The rapid daily dynamics of important resources for phytoplankton lead to the stability of functional forms demonstrated by the three categorizations, favoring colonizing abilities. Community response time, in functional terms, suggests aspects of functional stability, as was the case for MBFG VII, RFGs J and K and life form CNF, and/or even functional turnover. The recruitment of functional forms in a daily period evidences that ecological filters may be acting on the functional arrangement of littoral phytoplankton of Mangueira lake.

# Effects of climate change on the large shallow subtropical coastal lake based on the cumulative intensity heatwave index

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Although lakes represent only 0.26% of the total freshwater on Earth, society makes use of them through essential environmental services such as public supply and irrigation. These ecosystems must therefore be monitored using parameters that express their physical, chemical, and biological characteristics. One of the parameters typically used in lake management is water temperature, as it regulates the physical, chemical and biological processes in these ecosystems. To explore the effects of climate change in Mangueira lake, a shallow subtropical coastal lake, this study applied an approach to assessing heatwaves in lake ecosystems. Among the indices proposed by this methodology, determined using water temperature data, the cumulative intensity index was used, which provides important information on the duration and average intensity of heatwaves in lakes. Using an observed time series with daily air temperature data measured since 1961, the Air2Water water temperature estimation model was calibrated with surface water temperature data obtained via remote sensing. Then, based on climate projections obtained via global climate models, Air2Water was used to generate a time series of water temperature in the historical period and the SSP5-8.5 future scenario. All the projections associated with the cumulative intensity of water temperature indicated that Mangueira lake will be subject to more intense and/or longer-lasting heatwaves, implying potentially serious effects on the aquatic communities living in this environment. Heatwave indices, such as cumulative intensity, can therefore be used as a control and monitoring tool for lakes, with a view to the proper management of these ecosystems.

## Coastal habitats mapping within the framework of a state level cartography project

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Currently, there is no a homogeneous and updated state-level cartography of natural and semi-natural habitats in Spain. Harmonizing existing maps from the Autonomous Communities cartographies is complex, due to differences in scale, habitat classifications, and interpreting criteria. The project “Mapping of terrestrial habitats: improvement of territorial information at a national scale”, funded by the Ministry for the Ecological Transition and the Demographic Challenge (MITECO), aims to create a coherent national habitat cartography applying, to each polygon, two legends: the Spanish Habitats Checklist (EUNIS based) and the Habitats of Community Interest (EU Habitats Directive). The project is organized by habitat groups, being coastal habitats one of the most challenging due to dynamic natural processes such as waves, tides, and sea level changes, which leads to identification and delimitation issues. The broad classification of these habitats in the EUNIS system, including coastal dunes, salt marshes, and rocky shores, adds complexity to accurate mapping. Additionally, the significant heterogeneity across Spain’s territory, exemplified by the contrasting characteristics of its Atlantic and Mediterranean coastlines, presents another challenge. Moreover, the scale at which coastal habitats should be properly delimited is highly detailed. Auxiliary data such as LiDAR, geological maps, and fieldwork are essential to validate and improve the final product, resulting in a homogeneous and enhanced cartography of coastal habitats. The final map will cover all habitat groups with an interoperable legend at regional, national and European scale.

**OS8**

**Global Change and  
Aquatic Ecosystems**



# Assessing the impact of SALinization on the River GamBIA (SALBIA)

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Freshwater salinization is a global environmental challenge that poses a risk to ecosystems and human welfare, especially in those countries that are highly vulnerable to climate change such as The Gambia. Within this context, the project SALBIA proposes a multifaceted and interdisciplinary approach to assess the impacts of freshwater salinization on the socio-ecological system of the river Gambia. The 3 subprojects composing SALBIA cover three key facets to diagnose those impacts on: biodiversity, carbon cycling and gas emissions, and local communities. Accordingly, each facet assessed by SALBIA builds up basic knowledge needed to address a most-pressing societal need to face global change: biodiversity loss, greenhouse gas emissions and carbon cycling, and impacts on local communities and their adaptation. By assessing the relationships between those facets, SALBIA will not only generate new knowledge of broad interest to the international scientific community, it will also help to develop adaptation strategies that can be useful in many regions affected by freshwater salinization around the world. SALBIA will also contribute to significantly advance our understanding of freshwater salinization by targeting a region that has been largely neglected and aspects of it that have been rarely assessed (e.g. impacts on carbon cycling and nature contributions to people). This contribution aims to show the main objectives of SALBIA, together with the methodologies that will be used, the activities to be carried out (including capacity building) and some preliminary results mainly focused on the analysis of available data coming from previous studies in the region.

## Simulating warming effects on stream biodiversity (SIMBIO)

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Food webs integrate biodiversity based on feeding habits. The maintenance of biodiversity and associated ecosystem processes of these food webs (i.e., their persistence and robustness) depends on its structure. The integration of these webs when studying the functioning of ecosystems is essential due to the unprecedented loss of biodiversity because of global change. Of special concern are the effects of warming on the biota of headwater streams, given their high ecological value. Climate change reduces diversity, increases the rates of physiological processes, modifies the interactions between species and alters the rates of ecosystem processes. Highly realistic simulation tools (Next-Generation Individual-Based Models, NGIBMs) represent a great opportunity to study complex systems. We will simulate food webs with maximum structural realism, including Metabolic Theory of Ecology, individual growth curves, and genetically based traits. Therefore, it would link empirical data to the functioning of the food web and associated ecosystem processes, as a function of temperature. By means of simulation, we aim to investigate the persistence and robustness of headwater streams food webs in forecasted scenarios of global warming, as well as its consequences on the ecosystem functioning.

# Microplastics dynamic in a sub-urban river: spatial and seasonal variation

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Microplastics (Mps) have been recognised as one of the most important emerging pollutants and one of the main threats to biodiversity conservation worldwide. Early scientific research on MPs focused on marine environment rather than freshwater ecosystems. Nevertheless, recent studies have shown that rivers play an important role, as they are the main pathway for transporting litter and MPs from land to the ocean. These advances suggest that the number of MPs in freshwater ecosystems may be at least equal to that found in oceans, but some authors suggest that it can be even higher. This work aims to understand which are the most important factors that can explain the spatial and seasonal variability of MPs concentration in water and sediments in the Louro River (Galicia). Five sampling points were established along the river and eight sampling campaigns were carried out. At each location, the river flow was measured and one sample of 1000 cm<sup>3</sup> of sediment and three samples of MPs in the water column were collected from both riverbanks and from the centre of the river section to quantify the MPs concentration. The preliminary results showed that the MPs concentration in water ranged between 0.13 – 6.29 MPs m<sup>-3</sup> in the water column and between 26 – 679 MPs Kg<sup>-1</sup> in sediments. Although the concentration in water and sediments did not follow a marked spacial pattern, the results showed that the total amount of MPs transported by the river increased from the headwater points to those located near the mouth.



# Warming rate influence the increase on thermal tolerance of the cosmopolitan green microalgae *Chlamydomonas reinhardtii*

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Global warming is affecting the composition of freshwater phytoplankton communities due to the different capacities of species to adapt to increasing temperatures. Eco-evolutionary theory proposes that the rate of environmental degradation influences the probability of evolutionary responses to selective pressures. Thus, could rapid or slow warming rates affect differently the thermal resistance limits of freshwater phytoplankton? And would this warming stress affect primary productivity? An experimental evolution design was applied to the cosmopolitan green microalgae *Chlamydomonas reinhardtii* to test if different temperature increments (slow, +2 °C, and rapid, +4 °C) affect the ability of this species to survive over its initial limit of thermal resistance (LTR, 37 °C). However, after 5 months of cultivation (128 generations), none of the eight populations tested, four for each rate, was able to surpass its initial LTR. Nevertheless, an increase in thermal tolerance was observed: the derived populations showed growth and photosynthetic rates at 37 °C higher than the ancestral populations, that were maintained at 25 °C. Besides, when the increment was slow, all populations grew at 37 °C as much as the control at 25 °C. However, when the increase was rapid, only one population did. Another experiment suggested that, although the LTR was not exceeded in the rapid-warming derived strain, some adaptive mechanisms could be present in this population. Meanwhile, survival at 37 °C under slow warming could be related to acclimation. Our results show that slow warming rates might increase the tolerance to temperature of this phytoplanktonic species, enhancing primary productivity.

## Presence of anthropogenic debris in *Larus fuscus* excreta in a protected wetland

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In recent years plastic pollution has become an emerging threat to ecosystems integrity. Plastic debris have been found in food webs of both terrestrial and aquatic ecosystems. Generalist species are especially susceptible to ingest plastics and other anthropogenic debris. In this study we quantified and classified microplastics and other anthropogenic debris found in faecal samples of Lesser Black-backed Gull (*Larus fuscus*) collected in a protected wetland (Fuente de Piedra lagoon, Málaga, Spain) close to a landfill. For this, fresh excreta samples were taken from the wetland. Then, samples were disaggregated using distilled water and sieved to separate particles in logarithmic size classes (2000  $\mu\text{m}$ , 1000  $\mu\text{m}$ , 500  $\mu\text{m}$ , 250  $\mu\text{m}$ , 125  $\mu\text{m}$ , 63  $\mu\text{m}$  y 32  $\mu\text{m}$ ) in the laboratory. The particles retained in the different sieves were identified and quantified using binocular magnifying glass. Anthropogenic debris were sorted by class (i.e., plastic, glass, metal, aluminium foil), and colour. Microplastics and other anthropogenic debris were found in the 100% of the examined samples. The most frequent debris class was plastic sheets, and the most abundant particle colour was blue. Results reveal that the ingestion of anthropogenic debris by Lesser Black-Backed Gulls and the subsequent excretion in the wetland area constitutes a source of microplastics and other pollutants into the ecosystem. This process should be taken into consideration for conservation management strategies and land-use plans.

## Effects of water salinization and substrata quality on shredder performance

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Freshwater salinization is of major global concern disturbing biotic communities, ecosystem functions and services. Despite important lethal and sub-lethal effects on shredder invertebrates, it remains unknown if consequences result from direct exposure to contaminated aquatic medium, and/or from indirect effects of reduced substrate quality through fungal conditioning in salinized media. Here, chestnut and oak leaves were conditioned in reference (Cond0, 0 g/L NaCl) or salinized (Cond3, 3 g/L NaCl) media before being offered to the shredder *Schizopelex festiva* maintained in reference (Inv0) or salinized (Inv3) media. Invertebrates' consumption rates were higher for oak than chestnut, and in Inv0 than Inv3, but were not affected by conditioning media, despite lower fungal biomass associated with Cond3 leaf litter. Growth was only affected by invertebrate media (Inv0 > Inv3); Inv3 also led to the lowest survival. *S. festiva* preferred Cond0 over Cond3 oak leaves only in Inv0. Results strongly suggest that direct exposure to salinized media is the main pathway of salt toxicity to shredders, through a generalized reduction in invertebrates' metabolic rates when facing salt stress. Salt contamination may result in an energetic investment in osmotic regulation at the expense of consumption and growth, with consequences for invertebrate survival. Potential negative effects of salinization on shredders' ability to select more nutritious food items may contribute to cascading effects throughout the stream food webs, particularly in streams lined by more recalcitrant leaf litter.

# A comparative study on drought responses of freshwater fish and invertebrate communities using biotic integrity indices

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Droughts have an important role in shaping lotic ecosystems. Biotic indices are widely applied to monitor the ecological health of ecosystems, as established by the Water Framework Directive. However, their responses to drought disturbances, which lead to gradual adjustments of invertebrate and fish communities as habitats become dewatered, remain largely unknown. According to the Standardized Precipitation Index, in Catalonia 2020 was a year of abundant rainfall, in contrast to 2023 which was notably dry. To draw comparisons between the two years, several fish (IBICAT, EFI+, and percentage of exotic fish) and macroinvertebrate (IBMWP, EPT, IMMI-T, and DEHLI) indices were calculated at 14 sites (NE Iberian Peninsula). Overall, macroinvertebrate indices exhibited a more pronounced response to drought conditions compared to fish indices. The EFI+ showed a slight decrease and the percentage of exotic fish increased during drought, but the differences between the wet and dry periods were not statistically significant. The IBICAT remained nearly unchanged. Conversely, three of the four macroinvertebrate indices analyzed - DEHLI, EPT, and IMMI-T - experienced a significant decrease during the dry period. These indices appear to be more sensitive to drought than the IBMWP, which did not significantly differ between the wet and dry periods. The differential response of macroinvertebrates and fish to dry conditions could be attributed to differences in life span cycles, implying that invertebrate communities react more rapidly to environmental changes than fish communities. On the other hand, fish communities appear to be more resilient, provided that minimum survival conditions are not exceeded.

# Temporal succession of prokaryotic communities in a model Mediterranean high-mountain lake using sedimentary ancient DNA

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Mediterranean high-mountain lakes are particularly vulnerable to global warming. Paleoecological studies have contributed to understanding changes in chironomids, cladocerans, diatoms, and sedimentary algal pigments in high-mountain ecosystems of Sierra Nevada (Granada, Spain). However, few studies have examined changes in prokaryotic communities over geological scales in these areas. We examined temporal dynamics of prokaryotic communities in a lake of Sierra Nevada by studying variations in the abundance, diversity, and composition of bacterial and archaeal communities in response to changing environmental conditions using a sedimentary ancient DNA approach. Gradual increases in the absolute abundance of archaeal and bacterial communities were detected in the sediment core over the ~150-year record. While the number of observed archaeal taxa remained stable, gradual increases in the values of the Shannon and Simpson indices were observed during this period. Increases in alpha diversity indices were detected for the bacterial community from core bottom toward modern intervals as well as continuous and gradual changes in the composition of archaeal and bacterial communities. Random Forest models showed that, depending on the prokaryotic community, the variations found in the abundance, alpha-, and beta-diversity of archaeal and bacterial communities were explained by different paleoenvironmental variables as organic sedimentary carbon and chlorophyll-a and annual precipitation, among others. Overall, this study shows that prokaryotic communities are sensitive to variations in paleoenvironmental conditions that occurred over the past 150 years in a Mediterranean high-mountain lake and that archaeal and bacterial communities respond differently to these varying conditions.

# Fish Assemblages in Seville Supply Reservoir System: Implications for Management in a Water Scarcity Scenario

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During the current dry period, with low rainfall since 2019, the management of the reservoirs is more complex due to the reduction in the volume of water stored and the alteration of its quality. The fish fauna of the reservoirs is a factor to be considered in the management and therefore in 2022 the Metropolitan Water Company of Seville (EMASESA) promoted intensive assessments of their fish fauna. Using direct sampling gears (multimesh gillnets and boat electrofishing) and hydroacoustic methods, the fish assemblages of the 6 supply reservoirs were quantified and characterised.

Eight species were identified: 2 autochthonous (Andalusian barbel and Guadiana nase); and 6 alien: black bass, common carp, bleak, black bullhead, wels catfish, pumpkinseed and eastern mosquitofish. The prevalence of alien and invasive species is common to all reservoirs, and species richness varies from four to six species. Estimates of fish density and biomass range from 6.50 to 130.76 fish/dam<sup>3</sup> and from 5.98 to 69.78 g/m<sup>2</sup>, respectively.

The methodological approach used in these studies has provided useful insights into the abundance, biomass and composition of fish assemblages in reservoirs, and could be used to develop adaptive strategies for the management of the reservoir ecosystems, for the control of biological invasions and environmental risks and for scientific research purposes.

Further studies in reservoirs are recommended over the next 10 years to monitor the evolution of the fish assemblages and their response to the management actions and the water storage and use schedules.

# Effects of traditional ditches on macroinvertebrate communities of headwater streams in the Sierra Nevada National Park

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The importance of the Sierra Nevada National Park as an ecosystem service providing units for people, is linked to the regulation of the hydrological cycle through an ancestral water management system, the so-called acequias de careo. This system recharges sloping aquifers through ditches, which harvest water from headwater streams during snowmelt, increasing spatial and temporal availability of water for aquatic and terrestrial ecosystems and traditional agrosystems. The decrease in precipitation and snowfall, and the concomitant increase of water withdrawal with global warming, poses a need to assess the effects of flow reduction on biodiversity in stream reaches downstream of water abstraction. Here, we evaluated the effects of water withdrawal by traditional irrigation ditches on diversity of benthic macroinvertebrates communities of six headwater streams of Sierra Nevada. To this end, we compared reaches upstream and downstream ditch intakes in autumn and spring. Preliminary results exhibited a general decrease in streamflow in downstream reaches. Regarding invertebrates' community, no differences were observed for  $\alpha$ - nor  $\beta$ -diversity between upstream and downstream reaches, but  $\gamma$ -diversity turned out to be higher upstream.  $\beta$ -diversity of both groups was related to "species turnover" and local contribution to  $\beta$ -diversity was ca. 50% for both groups of reaches. Moreover, downstream reaches displayed less complex community structures and negative effects on regional diversity. Our results should be interpreted cautiously given the long-lasting drought suffered during the study period. Further studies covering diverse climatological conditions are necessary to better understand the effects of ditches' water withdrawal on headwaters of Sierra Nevada.

**OS9**

# **Extreme Aquatic Ecosystems**





# Diversity of planktonic and benthic communities in the high-altitude Salar de Pedernales basin, Atacama Desert, Chile

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Salar de Pedernales basin is located at 3300 meters above sea level in the Atacama Desert, Chile, covering an area of 3000 km<sup>2</sup>. This ecosystem is characterized by high exposure to ultraviolet radiation, low humidity, and extreme thermal gradients. Despite these extreme conditions, the habitat has rich autochthonous fauna and flora. Aquatic biodiversity in the basin is still poorly documented. Our study assessed alpha and beta-diversity patterns of planktonic and benthic communities across several salt flats (Pedernales S-O, Piedra Parada, La Laguna), natural water courses (wetland and creeks), and stream and artificial lagoon (Pedernales N-E). It examined their relationship with local physicochemical parameters. The basin was found to be a highly heterogeneous ecosystem; environmental variability was principally associated with changes in conductivity, pH, magnesium, calcium, potassium, and sulfate. Diverse communities were distributed across all locations except for the artificial stream. Phytoplankton and phytobenthic communities had higher species numbers ( $24.9 \pm 9.5$  and  $19.9 \pm 5.8$ ) than zooplanktonic and zoobenthic communities ( $4.6 \pm 1.8$  and  $4.9 \pm 2.0$ , respectively). In general, photosynthetic communities were spatially similar. Zoobenthic communities strongly differed between locations, while zooplanktonic communities were more homogeneous except in Salar de Pedernales N-E. Distance-based redundancy analysis revealed that phytoplankton and phytobenthos were mainly influenced by nitrate and sodium, respectively. Zooplankton communities were also affected by sodium, while zoobenthic composition varied with changes in both nitrate and lithium. Our data highlight the remarkable biodiversity of the natural water bodies of the Salar de Pedernales basin and the potential effect of anthropogenic intervention on the biota of these extreme habitats.

**OSII**

**Aquatic Ecotoxicology and  
Environmental Risk Assessment**



# Innovative Pilot Microplastics and Micropollutants in Water in rivers of Kanton Aargau (Switzerland)

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The project organized by Ambio GmbH and FHNW aimed to develop comprehensive sampling and analysis strategies for investigating microplastics and micropollutants in rivers, specifically within canton Aargau Switzerland. Employing non-destructive spectroscopic methods (Laser Direct Infrared Spectroscopy and  $\mu$ -Raman Spectroscopy) alongside with Pyrolysis Gas Chromatography Mass Spectrometry (Py-GC-MS), the study identified styrene as the predominant microplastic, originating from various polymers. Despite low concentrations, the study emphasized the necessity for accurate quantitative analysis, leading to the proposal of a new sampling strategy. Using a continuous sample collector, the project collected 12 liters of river water over two weeks, subjecting it to a 24-hour digestion process and subsequent Py-GC-MS screening. If microplastics were detected, secondary analysis with LDIR and RAMAN provided size and shape information. Standard operating procedures were established for consistency. The study unveiled microplastic pollutants in rivers, with Py-GC-MS and LDIR yielding matching results. Challenges included addressing contamination by subtracting laboratory blank samples. The combined use of Py-GC-MS, LDIR, and RAMAN proved valuable, highlighting their complementary nature for future research. Recommendations were made for further investigations into river sampling methodology, microplastic concentration, and gas phase reactions in Py-GC-MS, particularly regarding polyvinyl chloride (PVC). These proposed strategies and findings will be further explored in an upcoming bachelor thesis and potential InnoSuisse project.

# The ecotoxicological effects of metformin in aquatic standard species, in a climatic change scenario

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Pharmaceuticals' dissemination throughout the environment has resulted in their emergence as environmental contaminants. Metformin (MET) is frequently used as an oral antidiabetic drug for Diabetes type-II mellitus treatment, and is discharged in significant amounts into the environment, endangering aquatic ecosystems. Therefore, studying the effects of pharmaceutical contaminants on aquatic environments, along with the effects of climate change, such as rising temperatures, is crucial as concerns about these contaminants grow. This study aims to assess the ecotoxicological effects of MET in different aquatic standard species under predictions of a global average temperature increase. Growth inhibition assays were conducted on *Raphidocelis subcapitata* (3 days; 0.00 to 1000.0 mg MET/L) and *Lemna minor* (7 days; 0.00 to 200.0 mg MET/L), at  $24 \pm 1$  °C (OECD guidelines) and  $28 \pm 1$  °C (worst-case IPCC scenario), following OECD guidelines. Results showed that *L. minor* growth was affected by increasing metformin concentration, with an EC<sub>50</sub> (24°C) = 50.8 mg/L and an EC<sub>50</sub> (28°C) = 45.8 mg/L. A decline in fronds number and fresh biomass was observed at both temperatures. For 28°C exposure, these effects were visible starting from the lowest concentration tested (24.52 mg MET/L). Additionally, pigment content alterations were observed and an increase in antioxidant defense activities, for both temperatures tested in *L. minor*. Low toxicity of MET at the microalgae was observed (> 100 mg/L). The results show that, as the temperature increased, *L. minor* responded more sensitively to metformin. Further aquatic organisms will be tested to provide more ecological and relevant data.

# Exploring the plastisphere: Colonization dynamics of microalgae on plastics in European freshwater ecosystems

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Plastic pollution is identified as a common stressor affecting aquatic ecosystems. Global efforts aim to understand the interactions between plastics and aquatic biota that influence lower trophic levels. Plastics provide a new potential habitat for organisms, which is commonly referred to as the “plastisphere”. Interactions between microalgae and plastics remain understudied, especially in freshwater ecosystems. Here, we present the first results of the “PhytoPlastic” project, which won 4th collaborative FreshProject. Within this study, we examine these interactions by investigating the temporal scale of phytobenthos establishment on two of the main industrial use plastics, polyethylene terephthalate-PET and polyethylene-LDPE, in 17 lakes across Europe. Experiments were carried out installing the aforementioned plastic polymers in bodies of water for 3, 7, 15 and 30 days seasonally during a one year cycle. The research included the analysis of phytobenthos, with a specific focus on the microalgal component, and identification of variations in biomass by analyzing chlorophyll-a and Ash Free Dry Mass over time, across seasons, and under different treatments. To our current knowledge, this is the first field experiment investigating microalgae-plastic interactions on a broad geographic scale and also with high variability in environmental characteristics. This preliminary study will contribute to a better understanding of the consequences of the presence of plastics in freshwater ecosystems and identify key factors influencing the colonization process of microalgae.

# Understanding toxic cyanobacterial benthic mats from protected high-mountain rivers

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Toxic cyanobacterial mats have been related to animal deaths worldwide. This study aims to characterize mats from two high-mountain rivers which play an essential role in the Guadarrama Mountains National Park's ecosystem. Sampling was performed since late spring-summer 2021 and mats abundance was periodically monitored until June 2023. A comprehensive genetic analysis (PCR-based screening and metabarcoding) based on 16S rRNA and cyanotoxin-synthesis genes revealed two *Microcoleus* species in mats from both rivers. Besides, notable differences in dominant genotypes were found between the communities of both systems. *anaF* gene (anatoxins-related) from *Microcoleus* sp. was confirmed in both rivers. However, only Manzanares river presented genes linked to microcystins and saxitoxins. Some *Microcoleus* species are globally recognized as prominent anatoxins-producers being those the sole toxins detected in low levels in both rivers, with maximum concentrations found in Manzanares including two chemical variants (anatoxin-a and homo-anatoxin). This toxicity becomes significant in terms of risk assessment considering the recurrent mats proliferation and the high biomass reached in Manzanares where more than 20% of the riverbed was covered by mats at least on one occasion in above 80% sampling sites. The correlation between limnological factors and differences found among mats distribution and diversity will be discussed. These systems are highly frequented by wildlife and domestic fauna and act as destination for leisure activities. Globally, these results reinforce the need to understand the toxicity and ecology of these communities to carry out effective assessment and management of risks.

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# Exploring the ecotoxicity of the biopesticide pyrethrum extract on *Daphnia magna*

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Biopesticides are natural compounds considered safer and more sustainable for the environment. Nevertheless, a comprehensive evaluation is imperative due to the potential influence on non-target organisms, as certain evolutionarily conserved metabolic pathways and physiological functions need careful consideration. Pyrethrum extract (PE) is a widely employed biopesticide in agriculture, veterinary, and aquaculture practices. This study aimed to assess acute (0.6 – 40.0 µg/L; 96 h; E(L)C50 toxicity) and sub-chronic (0.7 – 1.1 µg/L; 10 d; life-history parameters) effects of PE on *Daphnia magna*. Furthermore, a comprehensive biomarkers approach was employed, encompassing antioxidant and biotransformation defenses, lipid peroxidation (LPO), neurotoxicity, and energy reserves content. Results of the acute exposure showed mortality and alterations in swimming behavior increased with increasing PE concentrations. PE was very toxic to *D. magna* [ $EC_{50}$  (96h) = 0.094 µg/L and  $LC_{50}$  (96h) = 23.3 µg/L], affecting also oxidative stress, lipid peroxidation, and neurotoxicity. In the sub-chronic assay, an increase in the activities of antioxidant defenses (superoxide dismutase, catalase, glutathione peroxidase), energy reserve content (glycogen content), and LPO indicated an imbalance in oxidative metabolism induced by PE exposure. This study underscores the potential of toxic effects in *D. magna* by PE, highlighting the vulnerability of non-target organisms to this biopesticide. Studies with new species can be essential to complete and make a review of the Safety Data Sheet (SDS) with a focus on environmental safety and ecotoxicity assessment of PE since this information is scarce and uncertain.

# Temporal dynamics of nutrients excess and pesticides exposure: Impacts on River Biofilm communities

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The temporal dynamics of stressors could produce diverse impacts on the biodiversity and functions of river microbial communities. Notably, nutrient excess and water pollution remain as significant stressors affecting river ecosystems. In this study, we investigated how the effects of the temporal occurrences of a pesticide's mixture (including herbicides, insecticides, and fungicides; stressor 1) influenced biofilm communities under different exposure patterns of occurrence (press vs pulse), against two constant nutrient concentrations (low vs. high; stressor 2). We conducted an experiment using 18 circular mesocosms (6 treatments), where biofilms previously colonized on artificial substrata under different nutrient concentrations, were exposed for 8 days at different pesticides temporal dynamics. Our findings showed that biofilm biomass and functions would be the most impacted after receiving pulse-occurrence compared to control and press treatments. Contrary to our initial expectation, we observed that microbial communities exposed to nutrient excess exhibited more pronounced impacts than those under low nutrient concentrations, which showed higher resilience to pesticide exposure. Our results underscore the complex interaction among multiple stressors and the importance of considering the temporal dynamics of stressor occurrence in river ecosystems.



# Does Artificial Light at Night (ALAN) affect the reproduction of an aquatic insect?

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Due to urban expansion, light pollution has become an environmental concern. Artificial Light at Night (ALAN), has been reported to adversely affect the behaviour and physiology of several organisms, including insects. However, studies are lacking in assessing the long-term effects of ALAN, which is of concern given the expected prolonged exposure of organisms to this anthropogenic stressor. Aquatic insects inhabiting urban environments, like *C. riparius*, are good model organisms for research on ALAN as they are prone to be exposed as larvae living in aquatic environments and later as imagoes (aerial stage) where they can be attracted to light sources. Moreover, *C. riparius* swarming behaviour may be affected by alterations in light intensity and spectrum, which may compromise their reproduction. This study aimed to assess the effects of ALAN over two consecutive generations (Parental - P and Filial - F1) of *C. riparius*. Organisms were exposed to 0, (dark night – control), 1 and 10 lux during the nighttime. Endpoints such as mean emergence time were evaluated for both generations, while reproduction and population growth rates were assessed for the parental generation. Preliminary results suggest that organisms from P generation may emerge slightly earlier when larvae were exposed to ALAN. However, ALAN exposure seems to reduce the fertility of *C. riparius*. Ongoing work is assessing the effects of ALAN on F1 generation giving insight on possible carry-over effects of ALAN. Data will allow inferring about the potential fitness consequences of ALAN exposure on aquatic insects.

# How to use mesocosms and multiparametric measurements based on biofilm responses in the environmental assessment of novel technologies for water treatment in the mining activity

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The mining industry may cause serious environmental damage to their surroundings, with considerable impacts on aquatic ecosystems. One of these impacts are the release of metals and the acid mine drainage. Several technologies based on membrane technologies and electrocoagulation have been developed to mitigate the impact of mining effluents. However, there is not a standardized method to assess whether these technologies suppose a significant improvement in water and environmental quality. We have developed an easy and cost-effective mesocosms approach based on the fast measurement and analysis of multiple functional (photosynthetic efficiency) and structural responses (community composition by in situ fluorescent quantification, biomass, and content in Chl. a) in freshwater biofilms. Biofilms from pristine rivers are grown in experimental methacrylate artificial streams with manipulable conditions of light, flow, and temperature to reproduce the conditions of the potentially impacted water bodies. Several trials were performed, aimed to assess the environmental quality of the effluents treated with these innovative technologies, obtaining high prediction capacity on their environmental benefit. The use of communities taxonomically diverse and the measurement of functional and structural parameters helps in the obtention of ecologically relevant results in the environmental assessment of these technologies because: i) this approach covers different modes of action of different stressors at different levels of biological organization; ii) key communities for the sustenance of aquatic food webs are assessed; iii) acute and chronic responses are integrated; iv) analyses are easy to be measured, cost-effective and reliable.

# Green and conventional synthesis of gold nanoparticles: a toxicity comparison using the microalgae *Raphidocelis subcapitata* in a context of climate change

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Due to their unique properties, gold nanoparticles (Au-NP) have been extensively explored in biomedicine, among other applications. Typically, these colloids are produced by conventional chemical synthesis (e.g. citrate method), though green-synthesis using plant extracts have been subject of interest as alternative methods praised by their environmental friendliness. Therefore, this work aimed to assess the toxicity upon green microalgae *Raphidocelis subcapitata*, of Au-NP obtained by the well-established citrate method and those derived from the synthesis using extracts of *Salvia* sp., *Ephedra* sp. and *Geranium*. Considering the current climate change scenario, it was also intended to assess the influence of temperature on the toxicity of the Au samples. Firstly, the colloidal Au-NP were fully characterized and then, *R. subcapitata* were exposed, for 72 h, to a range of concentrations of the four types of Au-NP at 20 °C, 23 °C and 26 °C. At the end of the assays, yield and population growth rate were monitored. When exposed at 23°C, microalgae were more sensitive to Au-NP synthesized with plant extracts as compared with those synthesized with citrate. Regardless the colloidal sample, the Au-NP induced significant effects on microalgae's yield or population growth when exposure occurred at 20°C. However, with an increase in temperature to 26°C, the lowest concentrations of Au-NP with citrate induced a higher growth rate on *R. subcapitata*, comparatively to the control. This effect was no longer visible for Au-NPs synthesized with plant extracts.

# Dynamics in potentially toxic cyanobacterial community from a drinking water reservoir

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This study aims to characterize the physico-chemical properties and cyanobacterial community from a Spanish reservoir used for drinking water supply. We focus on understanding the community dynamics and its potential toxicity. For it, biological samples were collected at different sites and depths of the water column, environmental data were also recorded over a year (from March to December 2023). A set of methodologies were employed to properly describe the system, including microscopy, fluorometric methods and genetic techniques (molecular screening by PCR and metabarcoding sequencing).

The morphological characterization revealed that the community was mainly dominated by 6 genera: *Planktothrix*, *Raphidiopsis*, *Anabaenopsis*, *Dolichospermum*, *Microcystis* and *Aphanizomenon*, being *Microcystis* and *Aphanizomenon*, previously identified as cyanotoxin producers in Spanish freshwaters. Indeed, the PCR-based screening for genes involved in the biosynthesis of the main cyanotoxin groups, revealed the potential presence of genes related to microcystins, anatoxins and saxitoxins. In the reservoir the highest diversity of potential toxins was found in October, coinciding with the maximum values of cyanobacterial biomass measured by chlorophyll a (>50µg/L Cyanobacterial Chl-a).

An in-depth microscopic analysis recollecting microphotographs are currently in process with the purpose of training algorithms based on artificial intelligence (IA) that could be helpful for automatic taxonomic identification.

Given the pivotal role of this reservoir in water supply, these results emphasize the need for intensifying monitoring endeavors and undertaking thorough investigations to accurately address the risk posed by potentially toxic cyanobacterial blooms in this environment.

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# Deciphering the ecotoxicological impacts of 4-chloroaniline chronic exposure on *Daphnia magna*

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The intensifying concerns regarding the impact of industrial compounds on aquatic ecosystems require a thorough comprehension of their ecotoxicological effects. This study explores the potential risks induced by the aromatic amine, 4-chloroaniline (4-CA), considering its growing use in various industrial processes and products. Moreover, 4-CA emerged as a candidate for inclusion in the 4th Watch List under the Water Framework Directive, predominantly due to its frequent occurrence in aquatic ecosystems. In this sense, chronic exposure of *Daphnia magna* to environmentally relevant concentrations of 4-CA was performed, where reproductive effects and biochemical biomarkers were evaluated. Results indicate that first brood fecundity and fecundity were significantly decreased at 9.645  $\mu\text{g}$  4-CA/L; and 0.294 and 9.645  $\mu\text{g}$  4-CA/L, respectively, while somatic growth increased above 0.942  $\mu\text{g}$  4-CA/L. These results indicate an impact on growth and reproduction performance. Biomarker analysis revealed significant alterations in several biochemical responses. Manganese superoxide dismutase activity increased  $\geq 0.092$   $\mu\text{g}$  4-CA/L, while copper-zinc superoxide dismutase activity also decreased in the lowest 4-CA concentrations tested (0.092 - 0.941  $\mu\text{g}$  4-CA/L). An increase of selenium-dependent glutathione peroxidase (0.092 and 9.645  $\mu\text{g}$  4-CA/L) was recorded indicating enhanced antioxidant defense mechanisms. Moreover, there was an increase in the genetic damage index ( $\geq 0.294$   $\mu\text{g}$  4-CA/L) with a dose-effect response, suggesting DNA damage. These findings underscore the complex nature of the ecotoxicological effects of 4-CA on *Daphnia magna*. By elucidating these mechanisms, our study provides valuable insights for assessing and mitigating risks associated with aromatic amine contamination in aquatic ecosystems.

# Pesticide contamination in the Albufera Natural Park during rice cultivation period (Spain)

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The Albufera Natural Park is a protected wetland on the Mediterranean coast dominated by rice paddies since the 18th century. It covers an area of 211.2 km<sup>2</sup>, between the mouths of the Turia River (North) and the Júcar River (South), including in its central area the Albufera de Valencia, a shallow hypertrophic polymictic coastal lagoon with a surface area of 23.2 km<sup>2</sup> and an average depth of one metre. The hydrology of the lake and wetland is linked to rice cultivation and is based on the connectivity of aquatic habitats (irrigation ditches, rice paddies, drainage canals and coastal lagoon). Pesticides used in agriculture are one of the main sources of accelerated pollution in this aquatic ecosystems and it was intended to study the distribution of pesticide pollution to obtain information on the current pollutants of the Albufera Natural Park. Twenty-one pesticides were found in water and sediment samples from aquatic habitats in the Albufera Natural Park. Seven of them were already banned by European law before 2016. The water and sediment samples showed 17 pesticides in common, related with rice cultivation. The most ubiquitous and dominant pesticides found in water and sediments in aquatic systems were bentazone and triclazole. The pesticides were estimated in the rice fields and the Albufera lagoon based on their actual volume of water and area of sediments flooded in 2016 (from the hydrological balance). This result is dangerous to the impact on the habitats of Mediterranean ecosystems under agricultural pressure such as this wetland.

# OSI2

## Ecosystem Services



# Litter decomposition in ponds across a large latitudinal gradient

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Ponds play an important role in the processing and decomposition of plant litter from both allochthonous and autochthonous sources, which in turn contribute to the global carbon cycle. The aim of the present study was to quantify microbial litter decomposition in ponds located along a large latitudinal gradient in an effort to predict changes in litter decomposition as a response to global change. For this purpose, we quantified the potential decomposition rate in 250 ponds located in eight countries (Belgium, France, Germany, Spain, Switzerland, Turkey, United Kingdom and Uruguay) along a gradient of land use intensity using the Tea Bag Index (TBI) approach. Selected ponds included permanent and intermittent ponds. We found lower potential decomposition rates in lower impacted ponds but the stabilization factor (indicator of the inhibiting effect of environmental conditions on the decomposition of the labile fraction) was not affected by land use intensity. Moreover, we found differences among pond hydroperiod for the stabilization factor, but not for the potential decomposition rates. Latitude had positive and negative effects on the potential decomposition rate and the stabilization factor, respectively. The observed unexpected latitudinal pattern could be explained by the specific latitudinal range included (within the temperate zone), where probably other local factors such as organic matter and nutrient input to the pond may play a major role. Our work contributes to the assessment of global organic matter decomposition across large geographical areas, which is key to understanding the effects of anthropogenic stress and climate change on these ecosystems.



# Litter decomposition in ponds across a large latitudinal gradient

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Dry rivers only flow after heavy rainfall or snowmelt. Despite providing many benefits to people, they are among the most altered fluvial ecosystems in the world. So far, the literature examining these alterations has focused on two direct drivers of change as the main causes: land-use change and climate change. However, it is not known how other direct and especially indirect drivers contribute to these alterations. This study aims to identify the drivers of change affecting the benefits of dry rivers, as well as the influence relationships between drivers. We selected three dry rivers in south-eastern Spain with high natural and cultural values. We conducted 37 interviews with the representatives of the main social groups in the study area. We asked them about perceived changes in the benefits of dry rivers in recent decades and about the causes of these changes. We identified 39 different drivers: 17 direct and 22 indirect. Indirect drivers were reported equally or more than direct drivers. Most drivers were perceived by interviewees as altering benefits (e.g., rural exodus), some as promoting them (e.g., public awareness), and others as both altering and promoting them (e.g., environmental policy) due to the different value systems of individuals. Not only do indirect drivers influence direct drivers (34 relationships), but also vice versa (5), and there are also influencing relationships between indirect drivers (36), and between direct drivers (13). The balance between the drivers altering and promoting the benefits of dry rivers seems to help maintaining quality of life.

**OSI3**

**Urban Aquatic Ecosystems**



## Urban stream ecosystems provide a key habitat for the maintenance of biodiversity within cities

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Freshwater ecosystems have been critically impacted by anthropogenic changes, and particularly by the effects of urbanization. Nonetheless, river and stream ecosystems within cities can provide key natural areas (blue and green) and ecological corridors, essential for the preservation of various biological communities. In this study we aimed to understand the potential of urban stream ecosystems for the maintenance of overall biodiversity. We surveyed 14 urban stream ecosystems of the city of Coimbra, Portugal and assessed their biological elements, focusing on diatoms, macroinvertebrates, fish and birds, as well as abiotic variables (n=10) and land-use indicators/urbanization indexes (n=8). The study sites displayed a high number of taxa (ca. 300) including sensitive macroinvertebrates (i.e., Plecoptera and Trichoptera), fish listed as Endangered on the national Red List of Threatened Vertebrates (*Anguilla anguilla*, *Gasterosteus aculeatus*) and emblematic birds as the kingfisher (*Alcedo atthis*). Diatoms showed the higher mean richness and diversity, while fish presented the lowest. Biodiversity indexes based on fish were negatively correlated with water nutrients content (i.e., Phosphates and Ammonia) and positively correlated with water depth (Spearman  $p < 0.05$ ). Birds showed the higher mean evenness within the study sites, which was positively correlated with the Habitat Quality Assessment obtained from the River Habitat Survey (Spearman  $p < 0.05$ ). The community structure and composition of the several biological elements was differently associated to environmental parameters and land-use (BIOENV and dbR-DA analysis). Our results indicate that different biological groups reflect dissimilar environmental impacts and point out to the importance of preserving the biodiversity of urban aquatic ecosystems.

## Satellite observations as a proxy of urban streams ecological quality?

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Healthy urban stream ecosystems are essential to safeguard biodiversity preservation and ecosystem services that contribute to cities sustainability and human health. The ecological assessment of European rivers is obtained through indices based on aquatic communities and hydromorphology, and water quality, which requires a high investment in field work, taxonomic expertise and sacrificing many organisms. Satellite observations could be a useful alternative if they are able to translate the ecosystem quality. However, the small size of streams and the complexity of the urban environment are a challenge. We investigated if spectral indices addressing vegetation and water (8 spectral bands and 5 spectral indices at 3m resolution, daily revisits) were correlated (Spearman) with the ecological quality ratios (EQR) derived from the Portuguese Index for Invertebrates (IptIS) and metrics, as well as with hydromorphological quality, water pollution and land use. We collected invertebrate communities from 16 urban streams in Coimbra, Portugal and used corresponding cloud-free imagery. The EQRs were not significantly correlated with any bands and spectral indices tested, demonstrating their inability to reflect the complexity of invertebrate communities. However, 3 bands (Green I, Red and Red-Edge) were significantly correlated with biotic index ASPT. Spectral bands Red and Red-Edge provide the most comprehensive assessments, being correlated with the biotic quality, urbanisation area, alterations in stream morphology (HMS), and habitat quality (HQA). Future work should explore via OneAquaHealth Geospatial and Satellite Information Platform the integration of these indices into the stream health assessment AI predictive models.

# Effects of Artificial light at night (ALAN) on the feeding behaviour of freshwater detritivores

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Urban aquatic ecosystems are commonly exposed to artificial light at night (ALAN) that can affect organisms' physiology and behaviour organisms. Nevertheless, there is a lack of data on its effects in benthic invertebrates that urge be addressed. In this study, we tested whether exposure to low levels of ALAN (1 and 10 lux) affects the feeding rates of three detritivore species: the dipteran *Chironomus riparius*, the caddisfly *Sericostoma vittatum* and the invasive snail *Potamopyrgus antipodarum* using a simple feeding assay where organisms feed on conditioned alder leaf discs for 7-10 days.

Results indicate that only *C. riparius* were responsive to ALAN with exposed larvae showing reduced feeding rates and larval growth. Both *S. vittatum* and *P. antipodarum* exposed to ALAN did not show any differences in their feeding rates when compared to control conditions (i.e. dark nights). Given that biochemical responses related with oxidative and general stress response have been observed before in *C. riparius* exposed to ALAN, *S. vittatum* and *P. antipodarum* organism have been preserved for measurements of their physiological condition. Those results will help assess organismal level consequences for these species with longer life cycles. Our findings highlight species-specific differences in sensitivity to ALAN. Furthermore, results demonstrate how these simple feeding bioassays can be used in laboratory and in situ as a behavioural endpoint to predict impacts of light pollution on benthic detritivore species and on ecosystem functioning (organic matter decomposition).

# Llegim el riu, a participatory initiative to assess and improve urbanised rivers through citizen science: identifying key issues in each community

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The co-creation sessions of the Llegim el Riu project aimed to address several environmental issues in the Llobregat basin, including river degradation, waste accumulation, water pollution, hydrological changes and regulation of public use of these ecosystems. After identifying the main problems in each municipality, specific measures were developed to mitigate these problems. Feasible measures were prioritised based on available financial resources or local group volunteerism, resulting in 1-3 actions to be implemented within 1-3 years. The success of the project was measured in terms of participation rates, satisfaction levels and real impact on local decision-making. The reasons for non-participation can be grouped into the following five themes: work overload of a tandem member (i.e. environmental and library technicians from each municipality), lack of local tandem coordination, political decisions of the council, lack of citizen participation and lack of public trust in the council. However, in municipalities where the project was completed, different degrees of success were observed. While some communities saw the formation or reactivation of volunteer groups and the resolution of past conflicts, others struggled to sustain the momentum or initiate further action beyond the end of the project. Overall, the project demonstrated mixed success in mobilizing community engagement and facilitating meaningful change in river management practices. Efforts to enhance participation, such as implementing online workshops, aim to address barriers to involvement and improve project outcomes in future iterations. Despite challenges, municipalities with ongoing local groups show promise for sustained environmental stewardship and collaborative decision-making in the region.

# Conservation, threats and vulnerabilities of the Culebrón urban wetland (Coquimbo Bay, Chile).

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Urban wetlands can provide important ecosystem services, along with maintaining the biodiversity and sustainability of cities. The Culebrón has been one of the wetlands in Coquimbo Bay (Chile) with the greatest impact and pressure due to urban development, the presence of debris, micro-garbage dumps, mining tailings and two roads that cross the site. The objective of this work was to document the threats and vulnerabilities of Culebrón (275983E-6683395N), in order to contribute to the restoration and conservation of this wetland. To identify and assess pressures, we carried out a review of the literature and media, informal conversations with experts and key actors, and field trips. The pressures were valued in scope and severity, and their sources in contribution and irreversibility. Several threats were identified that affect this wetland, including: garbage, vehicle traffic, excess visitation, exotic and invasive fauna, flow alteration and urbanization. The overall rating of the pressures varies from Very High to Medium, only hunting pressure had a Low rating. Even so, the vertebrate taxonomic richness was high: 4 classes, 12 orders, 29 families and 89 species, of which 8% have conservation problems. This wetland is home to 87 native vertebrate species, of which 57% are endemic to Chile. Among the natural threats to Culebrón wetland, the increase and intensity of storm surges and coastal erosion stand out, which together with its small size, make it highly vulnerable to global change. Funded by PFUE-RED-21992.

**OSI4**

**Biodiversity and Conservation of  
Aquatic Ecosystems**





# Status and perspectives of a highly threatened species; situation of the pen shell (*Pinna nobilis*) in the western Mediterranean Sea

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The fan mussel (*Pinna nobilis*), a large bivalve endemic to the Mediterranean Sea, is undergoing a catastrophic decline, possibly due to the protozoan *Haplosporidium pinae* and bacterial and viral diseases. This decline led to a population reduction throughout the Mediterranean region. Since September 2016, the extinction has spread rapidly across southeastern Spain, extending to Spain's Mediterranean coast, France, Italy, Greece, and beyond, resulting in nearly 100 % mortality of affected populations. Experts have warned of the risk of its spread and extinction due to the species' unique ecology and vulnerability, and consequently, the Spanish government has upgraded the species' conservation status to in danger of extinction. A rescue program began in November 2017, with the aim of keeping individuals in captivity for research and breeding purposes to better understand the disease. Although some individuals show resilience, predation and persistent disease pose significant challenges to their survival. Human activities such as poaching, habitat destruction and environmental pollution have already contributed to the decline of fan mussel populations, and pressure from cultural practices and commercial demands continues in the Eastern Mediterranean. Previous conservation efforts, including marine reserves and European policies, had helped the population recover from extinction. The results presented here, focus on the Ebro Delta population (NE Iberian Peninsula), are part of a LIFE project (LIFE PINNARCA NAT/ES/001265) aiming to advance our understanding of important aspects of fan mussel biology, and population status. This information is critical for developing conservation and management strategies to preserve the species.

# The diet and genetic structure of the pyrenean desman (*Galemys pyrenaicus*) populations in the catalan pyrenees

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The Pyrenean desman (*Galemys pyrenaicus*) is a semi-aquatic mammal endemic to the mountain rivers of the north and centre of the Iberian Peninsula that has recently experienced a strong decline. The use of molecular techniques allows to perform non-invasive studies from faeces, providing invaluable information of population genetics and diet, essential tools for the conservation of this elusive and endangered species. In this study, we investigated the genetic structure of the Catalan Pyrenees population using 24 polymorphic microsatellite loci from 113 faeces. The Bayesian analysis revealed 2 genetic and geographic clusters. For the diet analysis we applied a metabarcoding approach using the COI-Gillet primers on 228 faeces. We obtained 424,000 reads per sample, 56% of which belonged to the host, *Galemys pyrenaicus*, and 35% provided information on prey content. Aquatic taxa dominated with 78% of the reads, while 22% were assigned to terrestrial taxa. The diet of Pyrenean desmans was mainly composed by insecta (87.4%), gastropoda (3.3%), diplopoda (3.1%) and malacostraca (2.4%). Within the insect class, the most represented orders were diptera (24.1%), plecoptera (24%), ephemeroptera (22%), trichoptera (15.5%) and coleoptera (6%). There were seven species of insects that were present in more than 90% of the samples, which included MOTUS from the genera *Protonemura*, *Rhithrogena*, *Dinocras*, *Hydropsyche*, *Ecdyonurus*, *Epeorus* and *Baetis*. Together they made up 41% of the diet of *G. pyrenaicus*. In addition, the wide variety of prey taxa suggests a generalist diet for the Pyrenean desman.

# Understanding regime shifts in a shallow lake of As Brañas de Sada (Galicia, NW Iberia)

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As Brañas de Sada is a complex of coastal shallow meromictic lakes of semi-natural origin in which there are periodic regime shifts alternating between dark phases, dominated mainly by duckweeds (i.e., *Lemna minor*, *Spirodela polyrhiza*), and clear phases, dominated by submerged aquatic vegetation (i.e., *Potamogeton pusillus*, *Ceratophyllum demersum*). This wetland complex is a natural laboratory from which we can better understand these transition phenomena in regime shifts that entail functional changes and completely restructure this aquatic ecosystem. Therefore, we monitored water chemistry, stable isotopes (C, N) and interactions between biological communities during a regime shift episode in one of the shallow lakes, paying special attention to changes in composition, structure and biomass of zooplankton and the main groups of primary producers: phytoplankton, duckweeds and submerged macrophytes. A multivariate statistical analysis with abiotic and biotic parameters showed that phosphorus levels and zooplankton changes could play an important role in the transitions from clear to dark phases in the limnological dynamics of these aquatic ecosystems. Zooplankton plays a key role in the biogeochemical cycles of aquatic ecosystems, linking primary producers and the microbial loop with higher trophic levels, therefore constituting the main trophic regulator of phytoplankton competing for nutrients with duckweeds and submerged macrophytes, which decisively influences regime shifts and the dominance of duckweeds in these ecosystems.

# Knitting patterns and correlates of zooplankton functional diversity in mountain and lowland ponds

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We tried to disentangle the spatial patterns and correlates of zooplankton functional diversity along an altitudinal gradient across central and northwestern Spain. We identified key environmental controls of zooplankton functional diversity across a set of lowland and mountain ponds. We assessed if replacement or richness differences drove overall functional diversity patterns and evaluated the relative contributions of geographical and environmental distances to spatial variation in functional diversity.

Our findings highlighted the importance of environmental filtering with increasing elevation, leading to a potentially concomitant decrease in functional diversity values. However, altitudinal variation was not related to changes in functional diversity patterns, which were mostly the result of the pure loss or gain of species traits both in lowland and mountain ponds. We suggest that eutrophication associated to agriculture was the main factor underlying functional homogenization of zooplankton communities in lowland ponds. Local environmental filtering, rather than geographical distances, was the main driver of functional diversity and its dominant trait richness difference component.

This study supports the prediction that elevation fosters changes in different aspects of variation of zooplankton community functions. Our results highlighted the importance of local conditions determining the functional architecture of zooplankton communities, both in extreme environments and in lowland areas suffering from agricultural pressures and associated eutrophication processes. However, the fraction characterizing relationships between functional diversity and environmental correlates was generally low. This suggests the prevalence of idiosyncratic responses and random stochastic events in structuring variation of trait composition in freshwater zooplankton communities.

# Freshwater biodiversity of temporary streams from the Canary Islands in a context of water scarcity and invasive species: the CONACAN and BIOACUANA projects.

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Freshwater biodiversity is disappearing at an accelerated rate as a result of human pressures. This situation is especially relevant in oceanic islands, where native communities are characterized by high levels of endemism but also high vulnerability to human impacts. Besides, the knowledge we have on freshwater biodiversity in islands is much lower than in continental areas. Given this, it is possible that many undescribed freshwater species might have been already lost. The Canary Islands are characterized by severe water scarcity associated with climate change and a strong human water demand (e.g. banana plantations). The BIOACUANA and CONACAN projects focus on three of the Canary Islands (Tenerife, La Palma and La Gomera). The main goals of these projects are: 1) to hydrologically characterize the presence/absence of water in the sampled streams of the islands; 2) to produce basic information on the taxonomic and genetic diversity of aquatic diatoms and macroinvertebrates; 3) Evaluate the current status of aquatic invasive species; 4) to determine the vulnerability of the species to global change; and 5) to identify priority areas for the conservation of diatoms and aquatic macroinvertebrates insect biodiversity using systematic planning tools. Thus, these projects will help to improve basic knowledge of freshwater diatoms and aquatic macroinvertebrates in the Canary Islands, which include many endemic and vulnerable species. In addition, these projects will promote conservation strategies in the face of global change.

# Determining the role of mesohabitats and seasonality on the native species *Barbus meridionalis* fish population size structure. Case of study of the Ter River (NE Catalonia)

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Spatial diversity patterns of stream fish communities are broadly defined along the downstream-upstream gradients, but less is known about the fish habitat distributions at a mesoscale. Mesohabitats are defined as small spatial units within a stream reach and can be key to help establishing conservation measures for threatened fish species. Given that stream structure offers numerous mesohabitat opportunities, stream fish species can be spatially distributed due to habitat conditions and biotic interactions.

The objective of this study was to assess the role of mesohabitats and seasonality in the spatial distribution of the native fish species Mediterranean barbel (*Barbus meridionalis*) in a Ter River stretch, where systematic electrofishing surveys were conducted seasonally along 2021. We assessed how population abundance and body size structure of *B. meridionalis* responds to prey resources, non-native fish species, and environmental conditions. We hypothesize that *B. meridionalis* at different life stages would use differently the sampled mesohabitats along seasons to avoid intraspecific competition.

The poster will offer preliminary results of the main drivers affecting the spatial distribution of *B. meridionalis* at a mesoscale. We will propose future directions of research based on data modelling, and we highlight the use of mesohabitats for the potential application on river restoration.

# Distribution and larval habitats of Anophelinae (Diptera, Culicidae) in aquatic ecosystems of the Valencian Autonomous Region (Spain)

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Anopheline mosquitoes (Diptera, Nematocera, Culicidae) are biologically and medically important species not only because females bite and suck blood of birds, wild mammals, domestic animals and humans, but also because they can act as transmitting vectors of pathogens such as malaria to humans.

Fourteen species of the subfamily Anophelinae have been previously reported from Spain, seven of which have been recorded from the Valencian territory: *Anopheles algeriensis* Theobald, 1903, *Anopheles atroparvus* Van Thiel, 1927, *Anopheles claviger* (Meigen, 1804), *Anopheles maculipennis* Meigen, 1818, *Anopheles marteri* Senevet & Prunelle, 1927, *Anopheles petragrani* Del Vecchio 1939 and *Anopheles plumbeus* Stephens, 1828. The larval developmental sites of these species comprise a wide variety of biotopes always characterized by clean and fresh waters.

The aim of present communication is to update the knowledge of the *Anopheles* species present throughout the three provinces of Valencian Autonomous Region (Castellón, Valencia and Alicante). New data about the geographic distribution, location from sea level to high altitudes and types of aquatic developmental breeding sites where each species inhabit are offered. In addition, the physical-chemical parameters of larval habitats of each one of the species are described and species comparative maps are provided. The adult generalist or specialized feeding tendency and their potential vector risks for human populations are discussed.

# Macroinvertebrates as indicators of water quality in the Segura Basin

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Aquatic macroinvertebrates constitute an extensive group of animals that play a crucial role in freshwater ecosystems. They form a highly diverse group, characterized by a wide geographic distribution and various life strategies. Additionally, they are key players in substrate decomposition and nutrient dynamics, making them essential in food webs. They are quite useful for assessing the quality of freshwater as they act as bioindicators.

To conduct this study, a comparative analysis of the years 2021, 2022, and 2023 has been established on the presence and abundance of macroinvertebrates between two sections of the Segura River with very different ecosystem situations. The first section corresponds to the headwaters and the Fluvial Natural Reserve, with very good biological quality; the other section, located in the middle floodplain, is subject to various anthropogenic pressures and has poor biological quality. After the study, it has been observed that the water mass subjected to various pressures has a very low species richness, at 15%, compared to the headwater mass, which has a richness of 66%. Abundance is another noteworthy factor, as in the headwater section of the river, families are more abundant (<9000 occurrences), indicating predominantly waters with good environmental quality. Whereas in the more degraded section of the river, abundance is much lower (>150 occurrences), and species are more generalist, with a wide range of adaptation to various habitats.

**Keywords:** Macroinvertebrates, water quality, richness, abundance.



# New data on the taxonomic richness of the diatom genus *Asterionella* Hassall in the Duero and Miño-Sil basins (Spain)

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*Asterionella* Hassall is a genus of colonial pennate diatoms that inhabit the plankton of freshwaters. *A. formosa* Hassall is the most reported species of this genus globally, and particularly in Spain. In this work we studied the morphological heterogeneity of *Asterionella* populations from 17 lakes and reservoirs distributed in an area of NW Spain (part of the Duero and Miño-Sil basins) where only *A. formosa* had been reported, in order to check the presence of other species of the genus. Phytoplankton samples of each site were observed under light microscopy (LM) at 1000x magnification, and data of the main morphometric and morphological characteristics of *Asterionella* were recorded from randomly selected cells in valve or girdle view. Scanning electron microscopy (SEM) was also used for some selected populations. The cells from sites located in the Tera River subbasin (Duero basin) and nearby sites of the Miño-Sil basin were highly heteropolar in valve view and had a distinctive morphology that conform, both in LM and SEM, the characteristics of *A. ralfsii* var. *americana* Körner, a diatom known from acidic and oligotrophic lakes. The remaining cells were less heteropolar in valve view and most of them were identified as *A. formosa*, although some cells showed unclear characters and could not be assigned to a specific taxon. The attributes in girdle view were not as valuable for taxa differentiation than those in valvar view, which is challenging for determination in routine phytoplankton monitoring, as cells linked in colonies are usually observed in that position.

# The mystery of *Cyclotella alvarniensis*, modern or fossil diatom?

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*Cyclotella alvarniensis* is considered a fossil diatom species dated to the Upper Miocene. Sparce frustules have been found in Spain, France, Switzerland and, more recently, in Senegal. Although it has always been considered a fossil since its discovery, the causes of its aerial dispersion and geographical distribution are not clear. During a river diatom monitoring program in the Sant Llorenç del Munt i l'Obac Natural Park (Barcelona, Spain), we recorded the presence of *C. alvarniensis* frustules for the first time in Catalonia. Thirteen frustules were found in different habitats sampled from 2019 to 2023: in stream biofilms, artificial pools, disconnected pools, and rehydrated dry sediments. Although since its discovery, the records of *C. alvarniensis* have automatically being assumed as its fossil form, we discuss the possibility that it is could be also a modern species. This is because there are no closer diatomite exploitation areas nearby the Natural Park, which is usually the main reason for its appearance. As diatoms samples are usually treated to identify to species level, living samples are rarely observed, losing the opportunity to verify whether there are modern or fossil forms. In this study we try to answer the mystery of the appearance of *C. alvarniensis* in our samples, and we encourage researchers to observe living individuals in their samples in locations where this species can occur.

# Large Branchiopods as key prey for larval newts: a case of study on *Triturus pygmaeus*

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Globally, 41% of amphibian species are threatened with extinction. Temporary ponds are crucial ecosystems for amphibian breeding where large predators are typically absent, therefore reducing the predation pressure on their larvae. Also, in these ecosystems large branchiopods (LB) proliferate, becoming a significant food source for several species. This study aims to assess the potential of LB as food source for urodeles, especially for the development of pygmy newt larvae (*Triturus pygmaeus*), a species of conservation concern. In a controlled experiment, three groups of newt larvae were fed in independent aquaria with three different diets: water fleas (*Daphnia magna*), mosquito larvae (Chironomidae), and LB (*Streptocephalus torvicornis*, Anostraca) for 16 days. At the end of the experiment, we measured body mass, length, crest + tail area and height, and we recorded tail tissue samples of these larvae and diets for stable isotope analysis (i.e.  $\delta^{13}C$ ,  $\delta^{15}N$ ). Also, we recorded samples of potential food sources and measured and took tail tissue samples of ten newt larvae captured from the same pond as the experimental larvae. Preliminary findings indicated the treatments influenced tail morphology and growth of the larval newts, suggesting different hunting behaviour among newts of different monodiets, and the role of these LB as good quality preys, respectively. Isotope analysis indicated different signature for newts with different food source, highlighting the complementarity of the diets used. Temporary ponds have suffered a notable degradation in Europe during the last decades, also putting at risk the use of LB as prey for aquatic-terrestrial organisms.

# Primary productivity studies in the wetlands of the Peñalara Massif (Sierra de Guadarrama National Park, Central Spain) for monitoring the hydrological and ecological status

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The results of a primary productivity study in the wetlands of the Peñalara Massif (Sierra de Guadarrama National Park) are presented. The identification, counting, and determination of phytoplankton biovolume were carried out on 50 wetland samples collected between 2010 and 2021, along with the qualitative/quantitative analysis of photosynthetic pigments in 13 samples using HPLC-UV/vis. A total of 534 taxa were identified, specifically 359 in Lake Peñalara and 307 in the rest of the wetlands. Lake Peñalara exhibited lower diversity, density, and biovolume than the rest. Usually, the highest values of these parameters coincided with the summer period. Chlorophyta s.l. was the dominant group in Lake Peñalara, with predominantly small to medium-sized taxa exhibiting mixotrophic characteristics, while larger taxa of desmids and dinoflagellates were observed in other wetlands. In general, the wetlands exhibited an oligotrophic nature, although some of them showed signs of higher trophic levels during the summer period. The pigment analysis reflected some discrepancies with the phytoplankton results, indicating the need for a more specific study to review the pigments associated with each phytoplankton group. The results obtained in the Sierra de Guadarrama are compared with those available in other mountain systems of the Iberian Peninsula.

# Assessing the impact of Llegim el riu on citizens using mental models

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Through interactive and experiential learning activities, such as field trips, hands-on experiments, and discussions, citizens gradually build mental models that help them understand the complexity of river ecosystems and the challenges they might face. In this context, our study aims to analyse the mental models of citizens who participated in the participatory project called Llegim el Riu (i.e. Reading the River), conducted in 12 municipalities of the province of Barcelona (Catalonia, Spain). Specifically, we want to observe how mental models change before and after the citizen science activity using the RiuNet app, which allows participants to assess the hydrological and ecological status of rivers through on-site sampling. To this end, a simple exercise in which participants drew both how they imagined an unimpacted and impacted river was conducted before and after the activity, as well as two months after this activity during the co-creation session. Additionally, a short survey was also conducted to assess how participants' backgrounds influence their mental models, adding another layer of depth to our analysis and allowing us to explore potential correlations between demographic factors and perceptions of river ecosystems. Overall, our study seeks to provide valuable insights into the effectiveness of participatory approaches in promoting public understanding and engagement with river ecosystems, and their potential impact on collaborative decision-making processes.

**SSI**

**Advances in eDNA for the study  
of freshwater ecosystems: from  
technical to ecological aspects**



# Can eDNA metabarcoding be used to develop a biological quality index for disconnected pools in temporary rivers?

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Temporary rivers are fluvial ecosystems that alternate between different aquatic phases: flow, disconnected pools (DPs) and dry riverbeds. Rarely included in regular biomonitoring and conservation programmes, DPs are important refugia for aquatic biodiversity during dry-phase and contribute to communities during rewetting through recolonisation processes. Traditional macroinvertebrate-based biological indices developed for perennial rivers perform poorly in DPs, mainly because natural drought interferes with human disturbance. In addition, DPs tend to have small habitat sizes and high invertebrate densities. Consequently, local macroinvertebrate communities can be severely affected by traditional, highly invasive sampling methods. Given the high conservation interest of DPs, alternative methods for sampling should be explored. Besides improving taxonomic resolution over traditional biomonitoring methods, eDNA-based methods allow sampling without invasive techniques. Our aim was to investigate whether eDNA metabarcoding could represent macroinvertebrate changes along a gradient of human impact in the DPs, with a view to the eventual development of specific biomonitoring tools. We collected sediment eDNA samples from 55 DPs in Catalonia and processed them using fwhF2-EPTDr2N primers and Illumina high-throughput sequencing. We determined the anthropogenic impact gradient using the anthropogenic impact index (i.e. population pressure, land use, infrastructure and access) and identified indicator species along the gradient. We used these indicator species to develop a multimetric index of biological quality. Our study represents the first attempt to develop a biological index of DPs using molecular methods. This index is timely and useful as climate change and increasing water demand will make DPs the dominant habitat in many river systems worldwide.

# Environmental DNA (eDNA) and field surveys as complementary tools for monitoring amphibian communities

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Amphibians are the most threatened vertebrate class on Earth. Environmental DNA (eDNA) is an emerging technology that holds a great potential for improving biomonitoring programs of these conspicuous faunas. We assessed the degree to which eDNA metabarcoding can be used to infer amphibian community composition and species-specific relative abundance values compared to conventional field methods. Moreover, we designed and validated a cytochrome-b qPCR assay for detecting the Iberian ribbed newt *Pleurodeles waltl* Michahelles, 1830, and estimate its relative eDNA concentration in a set of Mediterranean ponds. We further compared both qualitative and quantitative data provided by qPCR with metabarcoding and conventional acoustic and visual surveys. eDNA metabarcoding retrieved all amphibian species detected by conventional field surveys, as well as a few additional taxa that could have been missed during acoustic and visual inventories. However, mean differences in community composition did not significantly diverge between the two monitoring methods. Interestingly, quantitative estimates obtained from eDNA metabarcoding seemed virtually independent of population density across all amphibian species. Nevertheless, our results showed a significant association between eDNA concentration and population density estimates for the Iberian ribbed newt, which may be related to the increased specificity of the qPCR approach. Although our results indicate that eDNA-based assessments can complement conventional approaches, further research is still needed to test the applications of these molecular approaches for addressing amphibian diversity and distributions in different ecogeographical contexts.



# Effectiveness of diatom metabarcoding to assess the ecological status of rivers in the Guadiana basin

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Benthic diatoms are one of the most studied biological groups, used in the evaluation of water quality. Their ability to respond to pressures that affect the environment in which they live, makes them an ideal biological indicator of water quality. However, they represent a highly complex taxonomic group, thus their morphological identification under the microscope is time-consuming and requires a high degree of specialization. Identifying taxa using a molecular-based approach, such as DNA metabarcoding, would be a promising alternative. In this preliminary study, we tested the use of metabarcoding to assess water quality with diatoms in the Guadiana Hydrographic Demarcation, comparing its performance to that of morphology-based identification of species. We applied both the traditional and molecular methods on 49 biofilm samples. The metabarcoding data were obtained using a diatom-specific *rbcl* DNA barcode. We assessed the effectiveness of the molecular approach to produce a species list, IPS scores and ecological status class comparable to those obtained with the traditional morphological approach. A new IPS index developed specifically to better adjust to the conditions of rivers in the Mediterranean basin, was used for this study. Overall, more taxa were found with metabarcoding than with microscopy. The correspondence between taxonomic lists detected by the two methods was relatively low but increased considerably when considering dominant taxa. Morphology-based indices did not show a significant correlation with sequence-based indices; in many cases, a status shift from good to moderate occurred. Further development of the metabarcoding method is needed for its use in environmental assessment.

**SS2**

**Exploring ways of Integrated  
land and water resource  
management to achieve  
temporary pond conservation**



# Remote sensing as a tool for the conservation of temporary ponds in Madrid, central Spain

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Mediterranean temporary ponds are priority habitats for conservation in Europe (Habitat Directive). However, despite huge efforts to build a cartography, the information about temporary ponds in peninsular Spain is incomplete, scattered across different databases, and without a proper classification as temporary or permanent ponds. Hydroperiod indicates the water permanence, being determinant for the species inhabiting temporary ponds. Nowadays, the shortening of pond hydroperiod and even desiccation are precisely serious threats to temporary ponds. We created a hydroperiod layer of the Comunidad de Madrid for the 2019-2020 hydrological cycle. First, we elaborated an inedited cartography of temporary ponds of this region, using different available cartographies, literature, and photointerpretation. Second, we constructed a hydroperiod layer over this vectorial data using satellite imagery of the Sentinel 2, resulting in the number of inundated days for each pixel (10 x 10 m). Finally, we assigned hydroperiod only considering the polygons in the vectorial layer of temporary ponds. We applied this tool to the Wetland Plan of Madrid (Decreto 26/2020), assessing the role of pond maximum hydroperiod and cumulative surface area of ponds in the richness of aquatic invasive alien species (IAS) in the protected wetland areas. We detected that only the maximum hydroperiod significantly influenced the richness of IAS after variation partitioning statistical analysis, revealing higher richness in protected wetland areas that included long hydroperiod/permanent ponds. We suggest wetlands including abundant temporary ponds spreading large surface areas could harbour high biodiversity also preventing the introduction of IAS, enhancing the pondscape resilience and conservation.

**SS3**

**Water management of  
reservoirs: the challenges  
of the future generations**



# Sentinel-2 Imaging System for Water Quality in Two Reservoirs in Algeria

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In the context of reservoirs to supply populations, it is crucial to assess the ecological status and understand how it affects biodiversity and ecosystem services. Sentinel-2 satellite imagery offers a valuable opportunity to obtain spectral information at the pixel level and monitor ecological changes. Variables such as chlorophyll a, suspended matter, and the depth of view of the Secchi disk are important indicators for assessing the good status of water bodies. The bio-optical algorithms applied to these images can contribute significantly to early risk management in the context of Global Change. To this end, a system for obtaining satellite images in near-real time is used, which allows semi-quantitative information to be obtained on the variables of interest already indicated, based on the adjustment with field data. The case study is the Kerrada and Cheliff reservoirs located in northwest Algeria. The result provides us with a normalized Root Mean Square Error of around 15%, which is an acceptable value for the estimation of these variables: Chlorophyll-a between 8 and 100 mg/m<sup>3</sup>; and transparency less than 1 m. With the algorithm obtained, we can see the time series of the variables and changes in spatial heterogeneity when an image is obtained from satellite.

# Remote sensing analysis of chlorophyll concentration, total suspended solids, and transparency in the lakes of Sangay Natural Park (Ecuador)

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The quality and quantity of water in the high Andean lakes of Sangay National Park (SNP) in Ecuador are essential for human life and the conservation and improvement of the environment. However, their study is challenging due to the difficulty of accessing many of them and the lack of human and material resources to carry out sampling campaigns and laboratory analysis. Therefore, remote sensing is shown as a valuable tool for their study. This work shows the use of automatic products obtained from Sentinel-2 satellite images for water quality observation and temporal study. These images provide us with free information on variables such as chlorophyll-a (Chl-a) concentration, Total Suspended Solids (TSS), and water transparency through the depth at which 90% of incident radiation is absorbed (KDZ90max). Using the C2RCC (Case 2 Regional Coast Colour) processor included in the free SNAP (Sentinel Application Platform) software developed by the European Space Agency for image processing, it is possible to obtain maps of these three variables that allow us to observe both their spatial distribution and temporal variation. An analysis has been conducted from 2016 to 2022, showing that Chl-a varied between 0.1 and 1 mg/m<sup>3</sup>; TSS between 0.1 and 0.7 g/m<sup>3</sup>, and KDZ90max between 10 and 30 m.

# Drivers of plant community composition and diversity in the shoreline of a freshwater reservoir in NW Spain

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Freshwater reservoirs are hydrological infrastructures with unstable ecological conditions and a variable flow regime. These artificial systems are colonized by species of flora and fauna that take advantage of the niche singularity. Here we have studied the plant community of the Abegondo-Cecebre reservoir near A Coruña, NW Spain. Our aims were i) to characterize the community and main drivers of composition and diversity, ii) to evaluate the potential restoration of a native community to increase stability and water quality, and iii) to measure the impact of invasive species in the community. We studied morphologic (slope, width), edaphic (granulometry, pH, nutrients) and biotic (tree cover, dead wood cover, total plant cover) variables in twelve study sites at regular intervals in the shoreline of the Abegondo-Cecebre reservoir near A Coruña, NW Spain. Twenty 0.5m<sup>2</sup> plots were placed in each site to measure occurrence and cover of all vascular plants. A range of multivariate techniques were used to address changes in the plant community, and how this relates to environmental factors. The results showed that the dominant herbaceous community, resulting from the physical and hydromorphological conditions of the reservoir, constitute a potential critical point for the establishment of invasive species. We argue that the high ecologic, economic and social detrimental effects urge to increase vigilance to prevent the introduction and spread of new invasive aquatic species.

# Cyanobacteria as indicators of water quality in multisystem cascade reservoirs: community, cyanotoxin and molecular analyzes

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Toxic cyanobacteria in public water supply reservoirs are a serious health concern since they can release cyanotoxins into the water. Among the cyanotoxins produced by cyanobacteria are microcystins (MC – hepatotoxin) and saxitoxins (STX – neurotoxin). In Brazil, the presence of STX in drinking water reservoirs is very frequent. The Cantareira System is composed of six multisystem cascade reservoirs, responsible for supplying 9 million people (SP), as such, samples were taken from the Jaguari (JG) and Jacareí (JC) reservoirs, the most eutrophic and upstream ones. This research aimed to estimate potentially toxic cyanobacteria through community measurements (cell density and biovolume), STX concentrations (ELISA assays) and detection of saxitoxin production genes, *sxtA*, and *sxtS*, through PCR analysis alongside genome sequencing. Cyanobacteria biovolume measured between 4.1 mm<sup>3</sup>/L – 8.9 mm<sup>3</sup>/L (dry season) and 4.8 – 12.0 mm<sup>3</sup>/L (rainy season). STX concentrations varied between 0.073 - 0.64 mg/L. Cyanobacteria biovolume and STX concentrations are significantly correlated ( $r = 0.87$ ). The PCR analysis corroborated these results through the detection of *Raphidiopsis raciborskii* genes. Both *sxtA* and *sxtS* genes were detected in both reservoirs, indicating that the present cyanobacteria species were capable of synthesizing SXT. However, neither *sxtA* and *sxtS* genes were expressed in the JC during the dry season. Representative genomic sequencing was performed for *R. raciborskii*. The sequences for *sxtS* in JG and JC were nearly 100% similar. For *sxtA* sequences presented high similarity for JG and JC with around 87% similarity.

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# Exploring the Influence of Climate on Hydrological Dynamics in Ebro Watershed Reservoirs (Spain)

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The hydrological regime water is an important factor to study as it allows us to understand the mechanisms that govern continental waters, whether they are lentic or lotic. This study analyses the hydrological dynamics of reservoirs within the Ebro River basin in Spain. The study aims to investigate the relationship between renewal time, hydroperiod and climatic variables, which has not been extensively explored before. It examines different climatic classifications according to the Köppen system to establish correlations between climate patterns and hydrological variables, particularly water renewal time and hydroperiod in reservoirs. To achieve this, a Python function was developed to generate a dataset that correlates all study data for the hydrological year (October-September) from 2010 to 2017. The distribution of hydroperiods among reservoirs varies annually, with significant fluctuations observed in 2013 and 2014. The study, also categorizes reservoirs based on their retention time, highlighting the differences between multiannual, annual, and hyperannual reservoirs. Correlation analysis between retention time and hydroperiod shows weak inverse overall relationships, with some exceptions during specific seasons. Reservoirs of certain climatic types show stronger correlations between retention time and hydroperiod, indicating climatic influences on hydrological dynamics. The study explains the differences in water retention times among the reservoirs in the Ebro River basin, which are affected by their volume and human activities, especially spring outflows for irrigation. Although larger reservoirs are more resistant to hydrodynamic changes, their management must consider ecological implications and prioritize adaptive strategies in response to climate change.

# Beyond standard parameters: the role of microplastics in Alto Rabagão and Aguieira water quality

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According to the Water Framework Directive (WFD), the ecological potential of reservoirs is determined based on specific physical, chemical, and biological parameters. However, microplastics (MPs; 0.001-5 mm) are pollutants, that are not included in water quality evaluation. To assess the influence of MPs on water quality, we conducted a study analyzing the MPs presence in two Portuguese reservoirs: Alto Rabagão and Aguieira. Furthermore, the obtained results were utilized to identify potential sources of these pollutants. For this purpose, three sampling sites were selected in each reservoir and monitored for one year. Subsurface water samples were collected for quantification and identification of MPs based on size class, color, and shape and to determine specific parameters following WFD guidelines. Additionally, the reservoirs surrounding areas were characterized by land use, soil occupation, and anthropic pressures. Fibers with a size between 0.1 and 0.5 mm were the dominant type of MP recorded in both reservoirs. In Spring season, Alto Rabagão showed higher MPs concentration compared to Aguieira (94 and 20 MPs/L, respectively). Blue and black were the prominent MPs color observed, but grey was also highly recorded in Alto Rabagão. Aguieira reservoir showed many anthropic pressures such as WWTP's discharges and surface water catchments while Alto Rabagão only has trout farms. Nevertheless, both reservoirs are used for recreation and leisure activities, which also may contribute to the results obtained. However, these results showed the high importance of integrating MPs evaluation on water quality assessment, since these aquatic ecosystems are used for human purposes.

# Anthropogenic impact on a subtropical reservoir (São Paulo, Brazil): classic profile and remote sensing study

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Limnology and Remote Sensing have been used to study reservoirs, but very few have been studied using simultaneously both approaches. Barra Bonita reservoir (São Paulo, Brazil), with a flooded area up of 310 km<sup>2</sup>, is formed by two arms, the Tietê and Piracicaba rivers, located in the most populous and industrialized region of Brazil. Tietê receives exceptional water contributions from of São Paulo city, while Piracicaba from small cities and a large agricultural area, with sugar cane being one of its main crops. Due to the large organic load that these rivers enter, profile studies show that their water masses can have an anoxic layer varying between 2 and 10 m in depth. A monthly time series analysis, for a period of seven years, demonstrated that the average surface chlorophyll (Chl<sub>a</sub>) concentrations for the Tietê arm ( $31.89 \pm 15.41 \mu\text{g/l}$ ) are stabilized and for the Piracicaba arm ( $17.65 \pm 10.72 \mu\text{g/l}$ ), a monthly increase trend of  $0.167 \mu\text{g/l}$  was observed ( $p < 0.001$ ). For these Chl<sub>a</sub> estimates, the SNAP product chl\_conc was validated, using Sentinel 2 images with 60m resampling, after C2RCC-C2X-Nets atmospheric correction. There is also a marked seasonality, with higher Chl<sub>a</sub> at the period of greater precipitation, attributed to the input of N and P into the arms, with lower monthly values during periods of lower precipitation. The approaches used in this study demonstrate the extreme anthropogenic impact on Barra Bonita reservoir and together they allow different and complementary perspectives on this quality.

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# Assessing the Impact of Land Use Changes on Suspended Matter in South Pantanal Rivers through Sentinel-2 Imagery

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The Brazilian Pantanal biome is increasingly threatened by anthropogenic activities, leading to significant land use changes. Specifically, the expansion of pasture and agricultural activities, resulting in the reduction of vegetation cover, adversely affects the quality of water in the Pantanal area. These human-induced activities, particularly deforestation and habitat fragmentation, are believed to exacerbate soil erosion processes. Consequently, an increase in the total suspended matter (TSM) transported by the rivers is anticipated. To investigate this hypothesis, a study was conducted from January 2016 to January 2024, focusing on the Areião River and the mouth of the Miranda River on the Paraguay River. Annually, two cloud-free Sentinel-2 images, from the rainy and dry seasons, were selected, totalling 17 images. Images were downloaded from the Copernicus Browser without atmospheric correction (AC). The SNAP program was used for resampling, to obtain a subset of the study area and apply Case2eXtrem-Complex neuronal net for AC, due to its lower specificity. The TSM values derived from the AC were analysed to observe temporal variations. This analysis was performed in three regions of interest (ROIs), each comprising 400 pixels, located along each river. For each image, the mean TSM value of each ROI was calculated, yielding 17 TSM values per river. The Mann-Kendall trend test was then applied to assess the trends of TSM values for each river. An increase in estimated TSM values from 2020 onwards was observed in all rivers, but the increase being statistically significant for the Areião River only.

# Phytoplankton changes in “L’Albufera de València” during 2023 are uncovered through their colour changes

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The hypertrophic status of “L’Albufera de València” is characterised by the dominance of cyanobacteria that give to the water an intense blue-green colouring.

Both, nutrients available and environmental conditions influence the composition and temporal evolution of phytoplankton populations. These factors control the distribution and abundance of algal populations as well as the appearance of new dominant species.

During winter 2023, the algal groups that reached the most significant percentages were respectively, in bio-volume, chlorophyceae 15%, and diatoms 40%, matching cyanobacteria with this percentage. During spring succession, the increased growth of filamentous cyanobacteria (*Cylindrospermopsis*, *Planktolyngbya* and *Pseudanabaena*), modified water colour that reached its most acidic green tones. In summer, cyanobacteria remained the dominant group with an increase in carotene-type pigments but without changes in taxonomic composition. Water colour gradually changed and in October, coinciding with exceptional environmental conditions of high temperature and wind, a very thin acicular filament (40 × 0.7 µm) bloomed, also coinciding with a strong reddish-brown (magenta) Albufera. This new colour triggers a great interest and excitement for all the sectors related to the lake and its visitors in general. However, the origin of this colour must be sought by the composition change of phytoplankton, as a result of the appearance and adaptation of new species to the new and lasting environmental lake conditions.

For this work, together with the classic techniques it has been necessary to apply other tools such as electron microscopy, high-resolution confocal microscopy and flow fluorocytometry.

**SS4**

**Research and management  
challenges on non-perennial  
rivers**



# Addressing the complexity of non-perennial rivers in environmental flow planning

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The extraction of surface and groundwater to meet human demands has always been in conflict with the conservation of freshwater ecosystems. In this vein, environmental flows (e-flows) are a widespread solution to protect, restore, and rehabilitate river ecosystems by limiting water withdrawal or regulating river flow. Initially, e-flows were conceived as a constant regime of minimal flow, but that idea has evolved to recognize and embrace the hydrological variability of river systems and the dependent ecological and social attributes. The e-flows concept and its related methodologies were originally designed and have evolved from and for perennial rivers, thus most existing methodologies focus on the habitat needs of fish in perennial rivers and in the regulation of river flow through dams. However, the majority of the drainage system in Mediterranean regions consists of non-perennial rivers in which existing methodologies cannot be applied due to the complexity of the flow regime of these ecosystems, the importance of groundwater withdrawal and the vertical connectivity, and the need to select ecological indicators other than fish. This contribution has a double goal: i) to propose an holistic framework to establish the hydrological, biogeochemical, ecological, and sociocultural bases for e-flows assessment in non-perennial rivers and ii) to share our experience applying this framework on the non-perennial streams of Menorca island.

# Hydro-morphological indicators for the assessment of temporary rivers during the dry phase

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Hydro-morphology is a key factor in river systems in terms of functional, ecological and environmental values. Despite being one of the three-quality assessment elements included in the Water Framework Directive, its use in temporary rivers has limitations because existing metrics does not capture the variability between flowing and dry phases. In Mediterranean river networks, where temporary rivers might constitute more than 60% of their length, hydro-morphological deterioration by human impacts is especially relevant because the dry phase is often undervalued or ignored by managers and citizens. The DRY-Guadamed project aims to develop advanced tools for the assessment of the ecological status of temporary Mediterranean rivers during the dry phase (including disconnected pools and dry riverbeds), and to address existing management challenges and needs. In this context, the identification of key hydro-morphological components during the dry phase for the development of a dedicated metric is crucial. To this end, we developed a hydro-morphological protocol that was applied to different Mediterranean basins in Spain (Segura, Guadiana, Tajo, Guadalquivir, Júcar, Ebro and internal basins of Catalonia). This protocol was structured in two sections and included: i) hydro-geomorphological parameters (channel morphology and connectivity), and ii) the presence of anthropogenic pressures, both in-channel and riparian zones. Main findings showed similarities and differences in the application of the protocol according to different hydrological states (disconnected pools and dry riverbeds) and basins. These results will guide the development of a hydro-morphological index suitable for the assessment of the ecological status of temporary rivers during the dry phase.



# Trends in the ecological status of temporary streams of Menorca

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Temporary streams in Menorca (Balearic Islands) rely heavily on groundwater due to their small watershed size. A comprehensive study was conducted to assess the ecological status of Menorca streams, which involved assessing macroinvertebrate communities and water physicochemistry in 14 campaigns over 5 years, spanning from 2005 to 2008 and 2022 to 2023, at 10 different temporary streams. The study aimed to identify temporal trends in water chemistry, ecological status, and invertebrate metrics. The study identified the importance of previous months' accumulated precipitation for stream discharge, with higher values corresponding to higher macroinvertebrate metric values. Although nutrient concentrations in the streams are high, results showed a decreasing trend in 2022 and 2023. The ecological status metrics showed low, but slightly higher, values during the 2022-2023 period, with high values at Best Available streams. These observed improvements in ecological status can be attributed not only to the observed decrease in the stream's nutrient contents but also to higher precipitation levels during the second period.

**SS5**

**Justice, Equity, Diversity, and  
Inclusion (JEDI) in Limnology**



## Is my teaching gender fair? Towards gender-fair teaching in Limnology

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Women scientists, as well as researchers belonging to groups historically excluded from academia, have vastly contributed to our understanding of aquatic ecosystems, but their contributions are still neglected in university curricula. Increasing their visibility is a matter of justice, and also a way to make visible diverse role models, which can help remove barriers for future scientists. Teaching practices and students' evaluations can also be gender biased.

Perhaps you think that your teaching practices are gender-fair, but are you sure? Just stop by for 10 minutes and take this self-assessment questionnaire that we have prepared for you to figure it out. This poster includes a QR code that invites you to respond to a self-assessment questionnaire about the inclusion of the gender perspective on university teaching on Limnology or Aquatic Sciences and applicable to any other Geoscience. The questionnaire includes aspects about self-awareness, teaching preparation, teaching practices, and evaluation practices. Based on your answers, the questionnaire will provide you with a general evaluation as well as guidance to improve the inclusion of gender perspective on your teaching. This educational resource has been developed by the Gender & Science Group of the Iberian Association of Limnology (AIL), as part of the Gender LimnoEdu project (<https://www.genderlimno.org/>). We have also developed open ready-to-use resources that you can apply to your university courses to improve the visibility of women scientists.

# The Gender & Science Commission of the Iberian Association of Limnology - Ten years advancing together towards gender equity

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The Gender & Science group of the Association of Iberian Limnology (AIL; <https://www.genderlimno.org/>) was established in 2014 with the main goal to promote gender equity in freshwater research. We are a group of dynamic researchers from diverse backgrounds and institutions that work voluntarily and collaboratively on various projects and activities. Our main objectives are: (i) to serve as an external observer of gender biases in freshwater sciences (ii) to conduct gender-related studies in the field, (iii) to enhance the visibility of women within the scientific community and society, and (iv) to initiate actions to improve gender equity in freshwater sciences. Our collaborative efforts involve interdisciplinary collaborations spanning social sciences, media, and art. We have curated two exhibitions: one delving into the historical and the contemporary contributions of female researchers in limnology, and the other exploring the impacts of climate change on freshwater ecosystems, with a focus on the engagement and vulnerability of women. Additionally, we have published five peer-reviewed articles and two book chapters, participated as speakers and organized over 12 special sessions in numerous conferences and seminars. We got two awards for “The most inspiring talk” and “Contribution to diversity and inclusion in academia. Overall, our activities have changed the attitude and consciousness of many researchers by amplifying the women’s voices, being an example of how self-organized groups can drive structural changes in scientific societies. We invite you to visit our website and Instagram profile, and welcome everyone to joining our highly motivated team.

# Diversity and inclusion on scientific societies: the case of the Iberian Society of Ecology

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The Iberian Society of Ecology (SIBECOL), founded in July 2018, groups professionals dedicated to the study of all areas of Ecology (theoretical, terrestrial, marine, and inland waters). The Commission on Diversity and Inclusion is one of the SIBECOL working groups, which aims to celebrate, protect, and raise awareness about diversity within SIBECOL members. The priority of this commission is the inclusion of those groups that have been historically excluded from the scientific community, namely due to their ethnicity/racial identity, sex, sexual orientation, gender identity, gender expression, physical or mental difference, national origin, ideology, religion, family situation, age or socio-economic status. The Commission's specific objectives include: 1) to act as an observatory of the diversity within SIBECOL; 2) to propose and develop activities and policies that improve the visibility and inclusion of minority groups in SIBECOL; 3) to develop studies on diversity and inclusion directly or indirectly related to the field of Ecology; and 4) to collaborate with other scientific societies, professionals, groups, and entities to achieve these objectives. Here, we present the different initiatives, activities and actions that the Diversity and Inclusion Commission of SIBECOL has carried out since 2020. This communication aims to start a debate and exchange of ideas on how we can improve diversity and inclusion within scientific societies to learn from different experiences in order to avoid the perpetuation of discriminatory situations for future generations, so they can be personally and professionally successful through a supportive environment.

# Emotional state, research activity and support for limnologists during the COVID-19 pandemic: Learning from lockdown

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With the emergence of the Covid-19 pandemic, researchers faced unexpected disruptions in both their professional work and personal spheres. Researchers had to deal with that challenge from an already tensioned profession, mainly due to precarious positions, unequal opportunities, and overworking. The Gender & Science group of the Iberian Association of Limnology (AIL) and the research group SINTE-Lest investigated how the Covid-19 pandemic impacted on limnologists in Spain. We focused on the pandemic's impact on the emotional state of the researchers, their research activity and their support needs since the lockdown began, and whether gender influenced their experiences. Data were collected through an online survey which was launched in July of 2020. Thirty respondents (20 females and 10 males) completed the full questionnaire. The results showed that negative emotional states were clearly intensified by the lock-down for both males and females, but females experienced a significantly stronger negative impact on their feelings than males. Many research activities were negatively impacted regardless of gender, for example: meeting deadlines, planning activities, and collaborating with other researchers. However, women suffered stronger impacts than men in rhythm of work, productivity, dissemination of works and reconciliation of research with other tasks. Regarding the support needed and received, women reported more frequently their need for emotional/personal support than men. Our results suggest women being a more sensitive to pandemic crisis in the academic domain. Finally, based on our findings, we provide recommendations for institutions to overcome the reported Covid-19-related challenges during potential future similar situations.

**SS6**

**Exploring the effects of global  
change on freshwaters: The  
crucial role of LTER studies**



# Heating up: Increasing water temperatures in high mountain lakes with diverse depths and geomorphological characteristics

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Environmental warming is the most conspicuous and relevant effect of ongoing climate change. However, the increase of a lake water temperature is not easily predictable as warming effects may co-occur with a myriad of other confounding climate factors such as changes in precipitation and droughts or changes in lake morphological features – effects that are not easy to untangle. Based on long-term time-series observations, we explore the increase in water temperature of four small high-mountain lakes in the Sierra Nevada (southern Spain) with different depth and geomorphometric features. Our hypotheses are that warming is occurring in all the lakes of this high-mountain region, summer water-column stability is increasing, duration of ice-cover period is decreasing, and that inter-lake differences are due to variation in lake depth and local geomorphology. Utilizing high-frequency in situ temperature data from years 2009 (Lake La Caldera and Laguna Larga) or 2011 (Río Seco and Aguas Verdes) to 2021, and multiple discrete measurements as far back as 1984, we will test the above hypotheses, describe the findings, and discuss their relevance to the ecology of mountain lakes in the Sierra Nevada.



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