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CENTERS OF RESEARCH

Water research in the Mediterranean: challenges and perspectives. The Catalan Institute for Water Research (ICRA)

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Summary. The mission of the Catalan Institute for Water Research (ICRA) is to implement a holistic, multidisciplinary approach to water research, taking into account all components related to freshwaters and human-used waters. The ICRA has developed a coordinated, cooperative organization structure to allow this multidisciplinary approach across a broad spectrum of water related sciences and technologies. ICRA's main goals are to provide scientific and technological perspectives and tools to issues derived from water needs, scarcity, and ecosystem conservation in the Mediterranean context. [**Contrib Sci** 10:207-220 (2014)]

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Outlining integrative solutions for humans and ecosystems

In relation to other areas of the world, the Mediterranean region is one of the most vulnerable to global changes, as well as to potential alterations in water availability. Dryland regions are particularly water-thirsty, and overpressures on water resources are the rule of thumb. The conjoint result of climate and human-related activities (including damming and abstraction) affects the run off of basins and thus water availability, and it results in structural water scarcity. Due to hydrological alterations, drainage networks

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experience variations in geomorphological dynamics (e.g., accelerated channel incision, habitat simplification). The chemical quality of the water is also affected, with higher nutrient and pollutant concentrations under lower flows in areas subject to strong human pressures. Biological communities respond to harsher environmental conditions with lower diversity, the arrival of invasive species, and a lower efficiency of biological processes (e.g., nutrient uptake and primary production, decomposition). Water scarcity and its derived effects concern both the fate of ecosystems and human well-being.

Potential alternatives to rising demands and harmful effects are neither simple nor easily decided upon. Growing human population demands and the prospect of global change suggest an incipient, but evident conflict with ecosystem needs, as well as effects on water quantity and quality. Addressing both human and natural system demands from a perspective of shrinking water resources requires a multidisciplinary approach, one that takes into account the complete water cycle (from ecosystems to humans and back). Ultimately, changes in water availability are likely to have a detrimental impact on economic activities.

Water issues in the Mediterranean setting include the existence of multiple stressors affecting the health of aquatic ecosystems, and the need to maximize water resources for human needs. Addressing these issues properly requires in-depth scientific, technological and management contributions to achieve a reliable approach to the analysis of the ecological quality of rivers, lakes, and seas, the development of new wastewater and drinking water treatment technologies, and the study of resource availability strategies that take into account sustainability, social perceptions, and related (financial and social) costs. This complex scenario, encompassing (and usually confronting) ecosystems and humans, guides the research mission of ICRA, the Catalan Institute for Water Research.

The Mediterranean basin is characterized by highly variable river flows, and scenarios of climate change forecast an increase in the frequency and magnitude of extreme events [8]. Warmer temperatures and reduced river flows will likely increase the physiological burden of pollution on the aquatic biota; biological feedback between stressors (e.g., climate change and nutrient pollution) may lead to outcomes. Ranging from limiting biodiversity to hindering economic activities in the region, thus threatening ecosystems health and directly impacting citizens and economic sectors that fundamentally depend on water, such as agriculture, tourism, industry, energy, and transport. These shortcomings will also raise sewage emissions, with bacterial activity increasing with temperature, thereby diminishing the quality of wastewater. Sulfide and methane emissions from wastewater treatment plants (WWTPs) have yet to be considered in the integral management of urban water systems, despite impacts at local and global scales ranging from unpleasant odors to direct emissions of greenhouse gases.

In this context, water reuse is becoming one of the most promising practices and can give birth to new alternatives for the reduction of fresh water consumption. Promoting the concept of a *closed water cycle* in highly water-intensive economic activities is one of the new frontiers to be explored. The development, testing, and dissemination of a closed water cycle require the development of innovative technologies that must be sustainable. The Mediterranean area constitutes, in this sense, a perfect laboratory, as it provides a market open to the rapid uptake of the proposed technologies and an unmatched visibility at the global level.

Water-related issues are not exclusive to the Mediterranean, but have a European-wide perspective. Annual water withdrawal for the whole European continent is projected to rise from the current 415 km^3 to ~660 km^3 by 2070 (for comparison the total annual runoff of the Rhine River is 73 km^3); whereas in areas with water scarcity an increase from 20% to 35% is predicted. At present, about 45% of the water is used for industry, 41% for agriculture, and 14% for domestic purposes (EEA 2007). In European regions such as eastern Germany, western Poland, and England, water demand exceeds water availability, and water scarcity has become an important management issue [1,9]. This scenario enhances the necessity of improving water management, water pricing, and water recycling policies to ensure the water supply and to reduce tensions among regions and countries [3].

Water supply and sanitation are already part of the day-by-day issues in industrialized countries. Innovative technologies for water treatment are being developed and introduced, but water-scarcity solutions also require new ways of thinking. The management of the whole water cycle has to be optimized, integrating green technologies. At the same time, the environmental and socio-economic effects must be exhaustively evaluated. Furthermore, technology is not the only issue as, for instance, decisions on the types of crops and on food trade have a much higher impact than any decision to build a new water infrastructure or improvements in irrigation efficiency.

The occurrence of multiple stressors, the response of the ecosystems, and the technological issues addressing human needs and supporting ecosystem preservation need to be integrated within complex socio-economic systems. Due to the shortcomings of current knowledge and, consequently, in management practices and policies, interactions between potential stressors have been neglected. Most policies regulate the pressure component of given stressors, but not their impacts, as is the case in the EU Waste Water Directive (91/271/EEC). Interactions among stressors and the resulting complex effects must also be considered, rather than only the pressure itself. An additional important shortcoming in current management practices and policies is the neglect of the influence of multiple stressors on ecosystem services. Thus, information on the biophysical and economic value of the ecosystem and its ongoing alteration because of global change limit the incorporation of ecosystem services into the improvement of current policies.

The role of ICRA in the Mediterranean context

The ICRA tackles water-related issues from a comprehensive scientific and technological perspective. ICRA was established in October 2006 by the Catalan Government within the framework of the Research Centres Network Programme (CERCA). ICRA was created with the mission of implementing a holistic, multidisciplinary approach to water research, taking into account all components related to freshwaters and human-used waters. ICRA is a foundation whose trustees are the Department of Innovation, Universities and Enterprise of the Catalan Government (DIUE), the University of Girona (UdG) and the Catalan Water Agency (ACA). ICRA receives support from several organizations, such as the Council for Scientific Research in Spain (CSIC), the ICREA institution, and the University of Lleida. The Institute's official headquarters, the H2O Building, has been in operation since October 2009 (Fig. 1).

ICRA has developed a coordinated, cooperative organizational structure that allows for a multidisciplinary approach to a broad spectrum of water related sciences and technologies. This multidisciplinary approach is one of the Institute's differential and referential features (Fig. 2). In particular, ICRA's work seeks to provide scientific and technological perspectives and tools for a number of issues related to water needs, scarcity, and ecosystem conservation.

ICRA's main areas of research

Research at the ICRA is organized in three main areas: *Water Resources and Ecosystems*, *Water Quality*, and *Water Technologies and Assessment*, with interlinked goals and interests (Fig. 2). It has provided an update on its research goals and main achievements since its foundation.

Water resources and ecosystems

This area investigates the spatial and temporal dynamics of water resources, whether surface or ground waters, and their potential effects on the structure and function of continental aquatic ecosystems. Special emphasis is given to irregularity in water resources and the effects of land uses and

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Fig. 1. The H_2O building of the Catalan Institute for Water Research (ICRA), Girona.

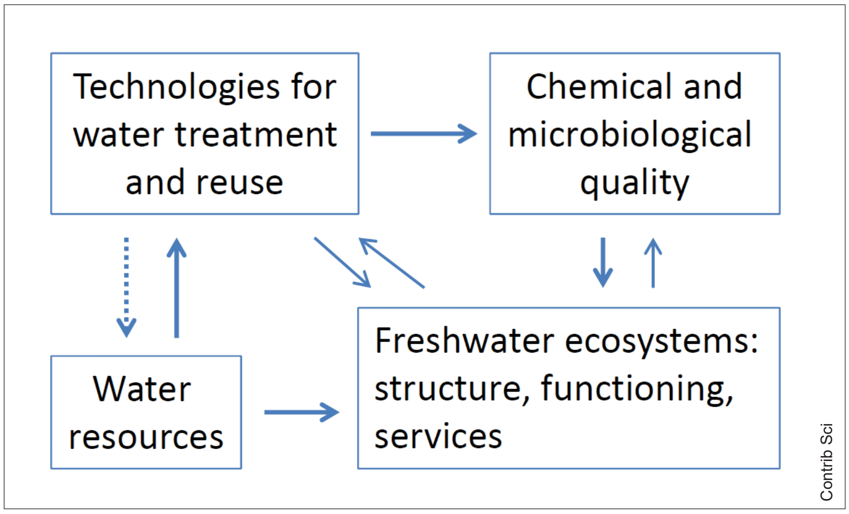

Fig. 2. Conceptual links of ICRA's main research objectives.

climate change on water availability and ecosystem dynamics. Research in this area relies on a strong field component. Gathering data through field measurements and experiments to analyze water-sediments-biota interactions in a range of fluvial environments is a key element in developing a strategic plan for the area. Measurements are based on permanent field stations, where components of the water cycle are recorded, and on *ad hoc* campaigns to observe and acquire data on physical and biological phenomena.

River hydrodynamics. Dams regulate the flow regime of rivers and have been constructed to satisfy water demands. Regulation is especially high in areas that experience strong climatic variability, such as the Mediterranean region. The flow regime controls the basic physical and ecological aspects of the channel's form and processes, including sediment and nutrient loads. Rivers in dry land environments tend to be more regulated than their humid counterparts. Dams, regardless of their use (hydropower production, irrigation, urban demand), alter the downstream flow regime of rivers. The resultant hydrological alterations include changes in flood frequency and magnitude, reduction in overall flow, changes in baseflows, and flow fluctuations as a consequence of the altered timing of releases. These alterations have a wide range of effects on riverine ecology. Within this context, our main goal is to analyze hydrological and geomorphological processes in Mediterranean river basins, especially those impounded by medium-to large-scale reservoirs and affected by changes in land use. On-going research is aimed at diagnosing physical processes in river basins to support predictive models and eventually achieve a realistic management of the physical environment [2]. This research line investigates hydrological processes in drainage basins, water resources, effects of changes in land use and upstream human activities on runoff and erosion, and especially morphosedimentary dynamics in the river network; special attention is paid to alterations in process magnitude and variability. The Muga and the Segre basins constitute the initial research areas. The core of our work is to construct water and sediment budgets through a research axis defined by measurement, modeling, and management. Work and experience are assembled from various fields of river science, including basin hydrology, fluvial geomorphology, freshwater ecology, and environmental and hydraulic engineering.

Dynamics of hydrogeological systems. Local flow systems, mainly those located in alluvial aquifers, are used to supply water resources to meet human needs. The evaluation of this relationship with respect to regional, large scale aquifer systems provides additional insights that may ease human pressures on presently exploited aquifer layers. Forthcoming climate changes will modify the water budget; therefore research is currently focused on evaluating the response of this impact at different hydrogeological scales. Quality issues, such as nitrate pollution and the newly detected contaminants in groundwater, are other actual topics of interest. These must be addressed to provide sound knowledge that will allow decision-making by managers and stake-holders regarding water resource planning. Nitrate in groundwater still poses a large threat to the safe use of water resources for domestic use. Its diffuse nature and widespread occurrence demand both clear planning and assessment strategies that consider vulnerability issues and potential actions based on the idiosyncrasies of each basin and an evaluation of the cost of any feasible solution. This implies a truly interdisciplinary approach that needs to be developed from a holistic perspective. Other chemicals of interest in groundwater quality research and management include antibiotics, fertilizers, and those originating from other human activities. Interest in these compounds is two-fold: they indicate the quality of the resource with respect to its intended uses and, more importantly, they reveal the flow-path from the surface to deeper aquifers, thus acting as tracers of natural or human-altered subsurface hydrodynamics. Nevertheless, the processes that control the migration and fate of these chemicals are poorly understood. Further, advanced field and laboratory research is needed to understand and predict the transport of such pollutants and their effect on groundwater quality. Similarly, microbial communities in aquifers are also considered in our research goals, as indicators of a complex world of biogeochemical processes that must be studied not only from a pathological perspectives of water-related illnesses, but also as indicators of aquifer properties and processes such as pollution degradation, all of which are of interest in water resources management.

Ecology of fluvial systems. The effects of intermittent water flow on river biogeochemistry and biota, the effects of temperature regime alterations in the processing of organic carbon, and the effects of global change on ecosystem services are the main river ecology aspects developed at the ICRA. The continued work in the intermittent stream Fuirosos has provided insights into the long-lasting relationship between hydrology, biogeochemistry, and ecosystem functioning in Mediterranean rivers [26]. The effects of climate extremes on the delivery of ecosystem services in the Llobregat and Ebro basins and the balance between water supply and the provisioning of water services are a substantial part of the Consolider-Ingenio SCARCE project. Within this project, a large field survey at the river segment downstream of the WWTP of Puigcerdà has allowed the chemical fate of several pharmaceutical compounds and the relative impact and response on the biota to be determined.

Research on the dynamics of carbon in fluvial systems is part of the MINECO project CARBONET, which studies the implications of global change for carbon transport and processing dynamics in river networks, with a particular focus on their lentic and lotic parts [22]. Field surveys at 21 sites within the Fluvià and Muga basins (NE Catalonia), and permanent monitoring equipment in several places within these basins, are the main tools to achieve a detailed budget of carbon transport and processing.

A recently awarded project (GLOBAQUA) will approach the relevance of water shortage and scarcity on freshwater ecosystems at the European scale. This project interlinks aspects on chemical and microbiological water quality, sediment dynamics, biodiversity, ecosystem functioning, and socio-economy, which define river basins under the pressure of water scarcity. Although mostly focused on Mediterranean basins (with partners in France, Italy, Greece, Spain, and Morocco), field and data analyses will also include areas from Germany, England, and even Canada, as they share similar problematic of excess demand with respect to the availability of resources. The final objective of GLOBAQUA (to be carried out from 2014–2018) includes providing a better comprehension (and correcting measures) of how to present policies and management practices influence the delicate equilibrium of the intensive use of water resources and the necessity to protect freshwater ecosystems.

Lacustrine and reservoir systems. Research focuses on carbon cycling in Mediterranean reservoirs, the impact of climate change on their water quality, and the effects of antibiotic pollution on reservoir bacterial assemblages. Interdisciplinary and cross-scale approaches are distinctive traits these studies. Organic and inorganic carbon dynamics in reservoirs and weirs, as well as the effect of global changes on the water quality of these storage systems, are part of the CARBONET project. In addition, the presence of antibiotics in reservoirs and its effects on planktonic communities in reservoirs has been investigated using an interdisciplinary approach that includes analytical chemistry, sequencing techniques in microbiology, and ecology [14]. The detailed descriptions of the effects of allochthonous organic matter on the metabolic balance in reservoirs and the consequences for water quality management in those systems are part of the ongoing discussion within the group joined by the COST action "Networking Lake Observatories in Europe" (NETLAKE).

Modeling of ecosystems and basins. Models are ideal heuristic platforms for stimulating critical thinking and to generate new hypotheses in environmental sciences. State-of-the-art modeling techniques are used in the detection and assessment of the effects of global changes in the functioning of Mediterranean fluvial basins and the management of water quality in man-made reservoirs. Activities within SCARCE allow: (i) modeling of the emerging pollutants at the watershed scale using the GREAT-ER model, with special emphasis on the processes occurring at the river reaches;

(ii) the inclusion of in-stream processes in the watershed-scale model InVEST, an ecosystem services evaluation model platform, in close collaboration with its developers (Natural Capital Project, Stanford); and (iii) the study of nutrient retention in river networks, including impaired streams using the SPARROW model. Also, the study of vulnerable regions in terms of water quality changes under scarcity conditions across the Iberian Peninsula, using state-of-the-art, computing intensive statistical tools (MINE and DFA) in Undarius, the ICRA's High Performance Computing cluster, is currently in progress.

Water quality

Research in this area is related both to water chemical contamination, particularly by emerging organic micropollutants, and to the impact of microbial diversity and activity on water quality.

Chemical contamination. Both of the main topics of research focus on water quality with regard to contamination by anthropogenic compounds; the first investigates contaminants in the aquatic environment and the second, the chemical quality of water in wastewater and drinking water treatment processes. Their broader aim is to provide a better understanding of the sources and processes that control the distribution of contaminants in ecosystems and their potential effects on these ecosystems and human health.

As the availability of analytical methods for the determination of trace emerging contaminants is the prerequisite for proper risk assessment, both lines of research continuously work on the development of advanced analytical methods for the determination of emerging contaminants in complex matrices such as wastewater, sediments, and biota using state of the art LC-MS/MS instrumentation. Among the methods employed are those for the simultaneous determination of 53 antibiotics [12], methods for the analysis of endocrine disruptors and related compounds in water and sediment [11] and biota [16], methods for the analysis of pharmaceuticals in biota [14], and methods for the analysis of cytostatic drugs [10]. Special attention is also paid to the transformation processes occurring during wastewater treatment and in the aquatic environment that affects the fate and behavior of emerging contaminants.

These techniques have been applied within the multidisciplinary project SCARCE. In this project, the environmental quality of Iberian rivers (Llobregat, Ebro, Guadalquivir, and Júcar) is investigated with respect to the presence of emerging contaminants. The evaluation of the presence of these contaminants in different environmental compartments will allow an assessment of the current condition of these Mediterranean rivers and the effects that global climate change, including climate change, could have on their chemical and ecological quality. Here, a notable result is the pioneering study on the accumulation of pharmaceuticals in fish from Iberian rivers and the widespread detection of diclofenac in fish tissue, both of which have captured the attention of the media nationwide [15].

The bioaccumulation of emerging pollutants in aquatic organisms is being investigated by ICRA researchers in another newly launched European project, ECsafeSEAFOOD, in which target contaminants are monitored in seafood collected worldwide. Results obtained from this ambitious sampling strategy together with the relevant information gathered from literature and national monitoring programs will enable risk assessment studies and the implementation of mitigation strategies to reduce the impact of contaminants that pose a risk to human health. For relevant contaminants, fast screening/detection methods tailored to suit stakeholders needs and to promote consumers' confidence are also being developed.

The project SEA-on-a-CHIP, which began its activities in 2014, aims at the development, validation, and implementation of early warning systems to assess chemical contamination in estuarine and coastal areas. A miniaturized, autonomous and remote immunosensor platform that can provide extreme sensitivity and selectivity for the real time analysis of up to eight target contaminants will be tested and implemented in aquaculture facilities.

The aim of the project DEGRAPHARMAC is the development of a treatment process for pharmaceuticals in sewage and sludge using lignolytic fungi, which have a powerful non-specific enzymatic system capable of degrading a wide range of xenobiotic compounds. Analytical methods are being developed for the determination of emerging contaminants, such as endocrine disruptors, and several families of pharmaceuticals (antibiotics, analgesics, cytostatics, etc.) in order to evaluate the capacity of treatment technologies based on these fungi real effluents such as those from reverse osmosis concentrate, wastewater from an urban hospital, a veterinary hospital, and a university residence, and in WWTP sludge.

The development and implementation of an innovative and efficient decontamination strategy are also the objectives of the European project ENDETECH. In this project, a novel technology based on enzymes able to eradicate pharmaceutical compounds and endocrine disruptor pollutants from wastewater, will be applied in bioreactors using tailored immobilization supports to enhance the stability and efficiency of those catalytic enzymes. Researchers from the ICRA are evaluating the efficacy of the enzyme-based treatment processes by means of chemical analyses and ecotoxicological hazard assessments of the selected pollutants and some of their transformation products.

Microbial diversity and activity. The effect of antibiotics on aquatic bacterial communities is one of the two main lines of research of microbiologists at the ICRA. A particular interest is the environmental distribution of genes conferring antibiotic resistance and how anthropogenic inputs affect their spread [15]. The presence of antibiotics, even at low concentrations, may stimulate the emergence and dissemination of antibiotic resistance genes (ARGs) among environmental bacteria, causing major environmental and health problems. We have therefore developed and optimized real-time PCR assays for the quantification of plasmid-mediated quinolone resistance (PMQR) genes in the environment [18] in order to study their abundance and dynamics in aquatic ecosystems influenced by WWTPs and hospital discharges. We have also used culture-independent approaches to determine the prevalence of ARGs and to examine how bacterial communities from biofilms and sediments respond to the discharge of WWTP effluents in the receiving river [19]. In addition, we have isolated a multidrug-resistant strain of the genus *Aeromonas*, which provides direct evidence that bacteria from aquatic environments affected by wastewater inputs constitute a reservoir of ARGs [20].

We are also engaged in several projects that study the diversity and activity of bacterial and archaeal communities involved in carbon and sulfur cycles in both planktonic and sedimentary compartments of continental water systems. The application of different molecular techniques such as CARD-FISH, qPCR, and massively parallel sequencing has permitted the study of the key microbial groups that resist cultivation. One project examines the contribution of uncultured archaea to organic carbon recycling in anoxic freshwater sediments and was recently funded by the Spanish government through project ARCOS (CGL2012-33033). This investigation is providing new data on the diversity and abundance of a specific lineage of *Crenarchaeota*, the Miscellaneous Crenarchaeotic Group (MCG), in sediment layers of lakes and reservoirs differing in their trophic status. Our current investigations point to a clear habitat segregation of MCG groups specifically adapted to lacustrine habitats. In addition, the combination of DNA/RNA-based molecular techniques and lipidomics has allowed the identification of active MCG subgroups at different sediment horizons as well as in biofilms developed on leaf litter. These results suggest the capacity of lacustrine MCG crenarchaeota to degrade humic compounds. In another project, we are investigating the activity of autotrophic sulfide-oxidizing bacteria of the class *Epsilonproteobacteria* in oxic-anoxic interfaces of stratified karstic lakes. Molecular data obtained from phylogenetic and functional gene markers as well as in situ incubation experiments using radiolabeled bicarbonate have provided direct evidences that a freshwater member of the genus *Arcobacter* is capable of an active dark carbon fixation coupled to sulfide oxidation at the redoxcline of studied lakes. Finally, and in close collaboration with colleagues from the Water Technology and Assessment Area (see below) we are studying the activity of biofilms colonizing anaerobic sewer systems, by combining molecular techniques and process engineering. Our main goal is to resolve the dynamics of biofilm formation by sulfate-reducing bacteria and methanogenic archaea and to elucidate their contribution to sulfide and methane emissions, respectively. Current investigations are focused on determining how sulfide and methane production rates vary in relation to physico-chemical variables and how the activity of methanogens is affected by sulfate-reducers.

Water technologies and assessment

This area develops and evaluates methodologies and technologies for optimizing resources, energy efficiency, cost reduction, and impact minimization of processes related to the urban water system. In the recent years, the focus has been on all aspects of urban wastewater systems, including wastewater transport (sewers) and wastewater treatment technologies and its economical and environmental impact. Recently, the expertise has been extended to include water reuse and drinking water technologies, thus expanding our area of research to the whole urban water system.

Assessing and minimizing detrimental emissions from sewer networks. Sewer systems are an integrated and very important component of urban wastewater and storm water management. By design their primary function is the conveyance of wastewaters and or/storm water to treatment plants or directly to the receiving waters. The nature of the wastewater, however, leads to a range of complex chemical and biochemical transformations, resulting in the release of detrimental compounds such as sulfide (toxic at high concentrations and causing offensive odors at low quantities; and responsible for corrosion in sewers) and methane (one of the main greenhouse gases, with a global warming potential 23 times higher than that of CO_2). The aim is the development and optimization of strategies to minimize the occurrence of these uncontrolled emissions. A three-way experimental approach has been implemented, including: 1) real-scale sewer monitoring to identify where these compounds are produced and emitted; 2) sewer pilot-plant studies to assess the effectiveness of several mitigation strategies and to understand the microbial ecology in these environments; and 3) predicting the potential emissions of any sewer installation. This research is currently being funded by a MINECO research project (GEISTTAR) and a Marie Curie Reintegration Grant (SGHGEMS) from the EU 7th Framework Programme.

Assessing and minimizing direct greenhouse gas emissions from wastewater treatment systems. Direct greenhouse gas emissions have been mainly overlooked when performing carbon footprint analyses in wastewater treatment systems. However, the general growing concern about climate change and the implementation of carbon taxes for CO_2-eq emissions in some countries have encouraged the water industry to account for these emissions. This research line focuses on the study of nitrous oxide (N_2O) production and subsequent emissions during biological wastewater treatment. N_2O is a powerful greenhouse gas, 300 times stronger than CO_2, and in some cases responsible for the majority of the carbon footprint on the planet. In recent years we have investigated some of the operational factors leading to N_2O emissions in nitrifying systems and proposed mitigation strategies for their minimization [24,25]. Also, we have conducted the first monitoring campaign in a full-scale domestic wastewater treatment installation in Spain [14], including the quantification of direct emissions of N_2O and CH_4. This research is currently being funded by a MINECO research project (GEISTTAR) and a Marie Curie Career Integration Grant (NITRI-GHG) from the EU 7th Framework Programme.

Occurrence of micropollutants across the whole urban water system and their biotransformations. The presence of micropollutants, including pharmaceutical, personal care, and endocrine disrupting compounds, in water bodies have been the subject of increasing concern in recent decades. An important fraction of these compounds arrives to the environment with the discharge of WWTP effluents into the receiving waters since most of our current wastewater treatment infrastructure is not designed for the removal of these micropollutants. This research line focuses on: (i) exploring the occurrence of these compounds across sewer networks [17], wastewater treatment facilities [6], and receiving water bodies [4]; (ii) studying different treatment processes for the removal of these compounds via advanced oxidation processes or biological degradation; and (iii) assessing the utility of proteomics to study the biodegradation of specific micropollutants [5].

Alternative water supply and advanced water treatment. In this line of research, the goal is to develop innovative technologies and to achieve the optimized integration of the different treatment steps in conventional and advanced treatments. Technologies to treat traditional (surface and ground water) and alternative water sources (saline, recycled and urban surface run-off) are assessed in an integrative manner directed at increasing resource efficiency, sustainability outcomes, and water quality. Research activities targets are: (i) innovation in membrane processes for desalination, water treatment, and reuse; (ii) monitoring and managing disinfection by-products in conventional and advanced treatments, thus generating drinking and recycled water; (iii) innovative technologies integrating the urban and industrial water cycle; and (iv) developing decision support tools for water management and distribution systems to reduce costs while increasing robustness. As an example, a recently awarded demonstration project from the European Commission, demEAUmed, is focusing on the implementation and promotion of innovative technologies for closed-loop optimal and safe water in Mediterranean Euro-tourist facilities. The project demEAUmed addresses two fundamental challenges: the importance of the tourism economy and the scarcity of water, characteristic features of the area. The project aims to provide a key platform to promote the use of sustainable technologies and innovative tourist facilities not only within the Mediterranean context but also under the global tourism market. The MINECO research project WATERFATE will explore basic aspects of the removal of priority and emerging micropollutants (metals and pharmaceuticals) and disinfection by-products (DBPs) in all the steps involved in the transformation of municipal wastewaters into high quality reclaimed water. The case study selected involves a membrane bioreactor coupled to nanofiltration/reverse osmosis followed by disinfection (chemical, physical and physico-chemical).

Life cycle assessment and modeling. Life cycle assessment (LCA) is a technique to quantify the impact associated with a product, service, or process from a cradle-to-grave perspective. Within the field of wastewater treatment, LCA pursues more environmentally sustainable methods to determine the environmental impact of design and operation decisions. Nowadays, there is growing interest from utilities, practitioners, and researchers to use the LCA methodology as a diagnostic tool in their installations. Our research in this area focuses on the use and adaptation of the LCA tool to incorporate the new goals of wastewater treatment technologies that are beyond water sanitation and include minimizing the loss of resources, reducing the use of energy and water, reducing waste generation, and enabling nutrient recovery [7]. The application of this tool is not limited to wastewater technologies but also has been applied to sewer networks [13].

Our efforts are also focused on predicting the behavior of certain pollutants (nutrients, micropollutants, greenhouse gases, sulfide, etc.) across the urban water cycle under certain conditions. Specific models are, therefore, being applied to describe the reactions occurring in each part of the urban water cycle: sewer systems, WWTPs, and rivers. Intensive monitoring campaigns conducted in full-scale sewer networks, WWTPs, and receiving waters are providing invaluable data for the calibration and validation of these models.

In this line, another European demonstration project was recently funded, R3-WATER. Its overall objective is to demonstrate how innovative technology solutions from European companies will turn municipal WWTPs into production plants for safe, reused water, energy, and products with direct markets.

Facilities at the ICRA

The most singular facility is the Scientific and Technical Services (SCT), formally inaugurated in May 2011. The SCT offers transversal comprehensive support to researchers, from the design of experiment to data processing, for both public and private projects. Tasks can be accomplished taking into account investments into advanced equipment, which is 50% financed by the Ministry of Science and Innovation (MICINN) and the European Regional Development Fund (ERDF) under the ERDF Operational Program 2007–2013 in Catalonia.

Since the Institute is a multidisciplinary center, a specific organization of SCT has been established through the following units: Chemical Analysis, Mass Spectrometry, Biological and Molecular Techniques, and Microscopy (Fig. 3). Laboratories operate in compliance with quality assurance methods to ensure technical competence, sustainable management of resources and accurate results, together with a continuous improvement plan.

Chemical Analysis Unit (CAU). Water characterization is a valuable tool for making decisions about future uses, treatments, and protection policies. Consequently, the determination of a wide range of physicochemical parameters provides the information needed to assess the impact of processes that are under evaluation. In this sense, the CAU offers high-technology instrumentation combined with a professional staff for analytical measurements in different water matrices, which generally run according to standardized protocols.

Equipment available in this unit includes a spectrophotometer (UV-1800, Shimadzu), BOD (OxiTop OC100, WTW) and COD (Autosampler 814, Metrohm) analyzers, an alkalinity titrator (Titrosampler 855, Metrohm), a conductimeter (GLP31+, Crison), a pH meter (GLP21+, Crison), a dissolved oxygen sensor (ProODO Handheld, YSI), freeze dryers (Lyoalfa 6-80, Lyoalfa 10-85, Telstar), an ultrapure water purification system (Milli-Q Advantage, Millipore), a muffle furnace (AFF1100, Carbolite), a TOC and TN analyzer (TOC-V CSH, TNM-1, Shimadzu), a discrete analyzer (Smartchem 140, Alliance Instruments), a total Kjeldahl nitrogen analyzer (K-370, Buchi), Rotary Evaporator (R-215, Buchi), an ionic chromatograph (ICS5000, Dionex), and an elemental analyzer (Truspec Micro CHNS).

Mass Spectrometry Unit (MSU). The development of more sensitive and versatile mass spectrometry instrumentation allows the detection of emerging organic contaminants at very low levels, in addition to providing the necessary tools to guarantee a precise identification, in agreement with the increasingly strict criteria established by the European directives. The MSU has been endowed with the following cutting-edge gas and liquid chromatography equipment:

- An ultra-performance liquid chromatograph coupled to a quadrupole ion trap (Acquity UPLC, Waters; QTrap 5500, ABSciex). QTrap5500 is a hybrid spectrometer, with triple quadrupole and linear ion traps that quickly and easily performs multiple reaction monitoring scans (MRM) for quantitation purposes and for the characterization of metabolites.
- An EQuan MAX liquid chromatograph coupled to triple stage quadrupole mass spectrometer (TSQ Van-

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Fig. 3. View of the Scientific and Technical Services (SCT) facilities at ICRA, showing the GC/MS and LC/MS equipment (left) and the Chemical Analysis Unit (right).

tage, Thermo Scientific). This configuration stands for the improvement of traditional time-consuming sample handling since it embodies on-line solid-phase extraction, thus providing more robust and efficient methods.

- A turboflow technology (or EQuan MAX) liquid chromatograph coupled to LTQ OrbitrapVelos (Thermo Scientific). For rapid sample preparation, this equipment includes a Turboflow technology that promotes turbulence in a column bed, thereby allowing molecules to move quickly into and out of the pores and packing particles. Orbitap represents the latest generation of mass analyzers and permits high-resolution spectrum (100000 FWHM) with a mass accuracy below 5 ppm. Furthermore, this system can work together with the direct analysis in real time (DART) technology, a powerful ionization technique for the rapid, non-contact surface sampling of compounds, either in solid or liquid form.
- A gas chromatograph coupled to a mass spectrometer (TSQ Quantum, Thermo Scientific). To extend the number of applications, a wide range of sampling and preparation techniques has been integrated (split/splitless, headspace and large volume injection, solid-phase microextraction). Other, routine detectors, such as an electronic capture detector (ECD) and a flame ionization detector (FID), are also available.

Unique features are associated with each of these mass spectrometers and enable the fulfillment of all the needs of environmental analysis, both identification and quantification. As a result, several fully automated analytical procedures for the determination of trace organic compounds (pharmaceuticals, endocrine disruptors, disinfection by-products, pesticides, etc.) in different kinds of environmental samples have been successfully validated.

Biological and Molecular Techniques Unit (BMTU). The BMTU carries out molecular characterizations of microorganisms in environmental samples (DNA and RNA extraction, detection, identification and quantification of phylogenetic and functional marker genes). The BMTU offers an analytical potential to meet the current challenges in the field of molecular microbial ecology of aquatic systems. For this purpose it has the following equipment for sample preparation: homogenizer (FastPrep-24, MP Biomedical), Fluorometer (Qubit 2.0, Invitrogen), microvolume UV-Vis spectrophotometer (Nanodrop 2000, Thermo Scientific), refrigerated centrifuge (5804-R, Eppendorf), high-speed centrifuge (Avanti J-26 XPI, Beckman-Coulter), centrifuge (5424, Eppendorf), ultrafreezer (-80°C) (CVF-525/86, Ingeniería de Climas,and NU9333E, Nuaire), vertical laminar air flow cabinet (AV-100, Telstar), UV-cabinet (UVC/T-AR, Grant Bio), incubators (IPP400 and INB400, Memmert), sonifier (250CE Digital, Branson), orbital shaker (KS260 BASIC, IKA), autoclave (Presoclave-II 30L, Selecta), concentrator (Concentrator Plus, Eppendorf), acute toxicity analyzer (Microtox 500, SDI), orbital shaking (SBS40, Stuart) and Ultrasonic (USC 2600 TH, VWR) baths, ice machine (HV-T 1020, Savemah), vortex (Genius 3, IKA), and block heater (SBH200D/3, Stuart).

The molecular techniques currently available are: thermal cyclers (2720 Thermal Cycler and Veriti Thermal Cycler, Applied Biosystems), real-time thermal cycler (Mx-3005P, Stratagene), denaturing gradient gel electrophoresis (phorU-2x2, Ingeny), electrophoresis (Mini-SubCell-GT and Protean II xi Cell, BioRad) and molecular imager (GelDoc XR,+Biorad,), and UV table (ETX-F 26MX Super Bright, VilberLoumat).

The Radioactive Installation of the ICRA (IRA) is a specific research laboratory allowing work with radioactive isotopes. The radioisotope laboratory will be classified as a category 3 radioactive installations for research purposes. The IRA has been designed to operate with the main radioactive isotopes used, in particular ^{3}H, ^{14}C, and ^{35}S. The laboratory is made up of a hall, designed as the dressing and decontamination room, a laboratory for manipulation and counting with the equipment needed to measure the incorporation of radioactive compounds into samples and subsequent analysis through liquid scintillation counting, and a laboratory for the management and storage of radioactive waste, defined as an area for the segregation of liquid and solid residues, with temporary storage areas for them at specific sites until their removal as radioactive or conventional wastes. The IRA is equipped with a liquid scintillation analyzer (Tri-Carb 2910TR, Perkin Elmer), and a monitor for measuring radiation and radioactive contamination (MiniTRACE ß CSDF, Saphymo GmbH). Radioisotopes are used in a wide range of research disciplines and practical applications:

- Quantification of primary productivity estimated as the uptake and assimilation of dissolved inorganic carbon (^{14}C-labeled bicarbonate)
- Quantification of bacterial production, estimated as 3H-thymidine (3H-TdR) incorporation into bacterial DNA
- Metabolic labeling of cells, based on the quantification of protein synthesis in cultured cells through the incorporation of sulfur amino acids (^{35}S-methionine or cysteine).

Microscopy Unit (UM). The MU contains the infrastructure and technical support needed for microscopy techniques. The equipment includes a confocal microscope (CS1, Nikon), an epifluorescence microscope (Eclipse 80i, Nikon) and an inverted microscope (Eclipse Ti-S, Nikon). An optical stereoscopic microscope (SMZ1000, Nikon) allows advanced microscopy (gene expression studies, 3D analysis, cell quantifications, FRAPS FRET, etc.).

Water Science and Technologies Research Platform (PLANTEA). This is a research infrastructure that supports both research and transfer activities. PLANTEA includes pilot plants of different sizes that enable basic and applied research projects in advanced treatment processes for waste, treated, or drinkable water (Fig. 4). The PLANTEA platform includes two groups of research installations with two different objectives: (i) the study of fluvial ecosystems under different conditions in an experimental streams facility and (ii) the study of wastewater transport and treatment systems in a pilot scale installation mimicking full-scale systems.

The facility is equipped with artificial streams (Experimental Streams Facility) that allow the manipulation of different ecological variables and characterization of the ecosystem response. Specifically, the facility (Fig. 4) has four functional units with six artificial streams each, allowing experiments with 24 channels simultaneously and therefore providing broad flexibility in experimental design (for example, six treatments with four replicates per treatment). This facility enables the study of the behavior of rivers in different situations, such as drought, with respect to chemical pollutants and /or biological fluctuations and temperature, among others. It aims to play a key role in research activities devoted to stream ecosystems and ecotoxicology. This automated facility is especial in Europe, although similar facilities can be found in Vienna, Berlin and London, as well as in the Environmental Protection Agency (EPA) in Cincinnati, Ohio (USA).

So far, two experiments have been successfully performed at the Experimental Streams Facility at the ICRA. The first one examined the effects of the duration of non-flow events on functioning of stream ecosystems. Temporary streams (those experiencing non-flow events) are ubiquitous and are probably more common than perennial streams. Moreover, they are becoming more common in many regions because of global climate change. However, the effects that the increasing duration of these non-flow periods have on stream functioning has been ignored. These changes are relevant, as the functioning of streams determines to a large extent economically relevant ecosystem services such as water purification, and perhaps also organic carbon dynamics in river networks. In this first experiment, we assessed the effects of the duration on different functional variables and found that autotrophic processes are more negatively affected by non-flow events than heterotrophic processes. Therefore, leading the system to heterotrophy and possibly disrupting the longitudinal connection of temporary streams to downstream systems in terms of organic carbon fluxes. The second experiment examined the interactive effects between

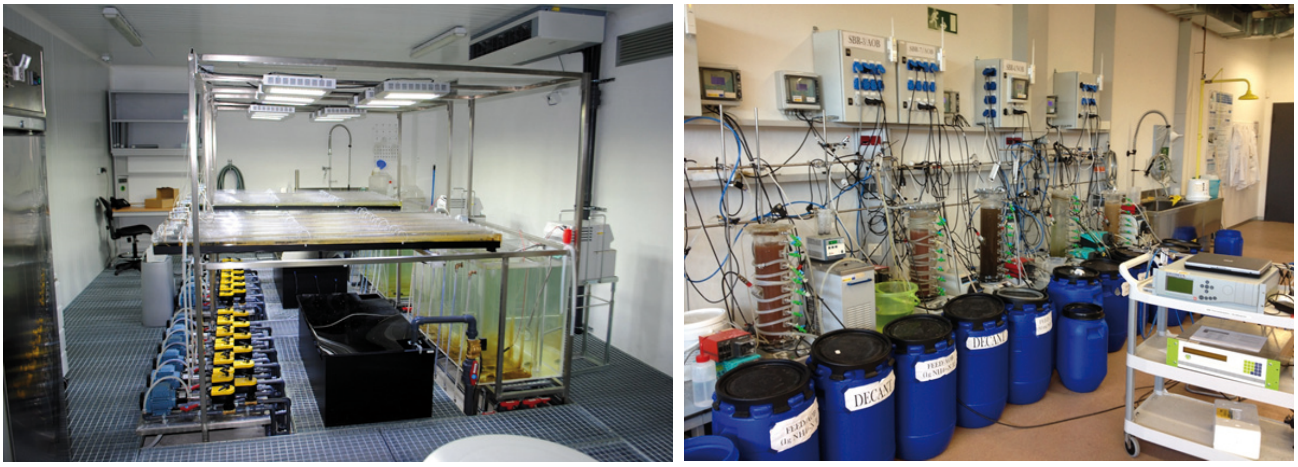

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Fig. 4. View of the Experimental Stream Facility (left) and of the Wastewater Treatment Facility (right) of PLANTEA at the ICRA.

a physical stressor, such as non-flow event, and a chemical stressor, such as chronic exposure to a mixture of pharmaceutical compounds. Most ecosystems, such as rivers, are exposed simultaneously to several stressors, in so-called multiple-stress situations. Therefore, assessing the combined effects (e.g., a non-flow event and chemical exposure) on natural communities is crucial for a better understanding of the potential effects of global change on river ecosystems. In this second experiment, we assessed, both separately and combined, the effects of a non-flow event (simulated by a dry period of 7 days) and of exposure to a mixture of ten pharmaceuticals (those more commonly found in polluted rivers) on biofilm structure, function, and tolerance induction. We determined that the effects of pharmaceuticals on biofilm communities were modulated by a non-flow event and that those effects differed between algal and bacterial communities.

PLANTEA includes several pilot plants mimicking real wastewater transport and treatment systems. To facilitate the operation of these systems, a connection to a nearby sewer network was built that provides fresh domestic sewage to the laboratory's pilot plants. Regarding the sewer systems, two pilot-scale sewer systems simulating two rising mains from a local sewer network from Catalonia are currently operating. The majority of the detrimental compounds produced during wastewater transport originate in the anaerobic zones of the sewer networks, the rising mains. The two most detrimental compounds produced are hydrogen sulfide, responsible for obnoxious odors and toxic at certain concentrations, and methane, which after CO_2 is the most important greenhouse gas. These sewer pilot plants enable studies of the chemical and microbiological transformations in these parts of the sewer networks, which are very difficult to access in real facilities. These installations, which are the first at the European level, allow researchers to investigate why and how these detrimental products form during wastewater transport and how their formation can be prevented. It also allows sampling of the microbial community present in the different parts of the sewer pilot plant, providing valuable information needed to link process performance with microbial ecology.

In investigations based on wastewater treatment systems, six sequencing batch reactors (SBRs) are also being operated in PLANTEA. These reactors, completely controlled and monitored, allow the study of different biological processes occurring in WWTPs during the removal of nutrients. Current investigations are focused on unraveling the mechanisms behind N_2O production during nitrification and denitrification, key processes in the removal of nitrogen from wastewater. N_2O is a potent greenhouse gas, 310 times more powerful than CO_2, and its uncontrolled emission from wastewater treatment processes has gained the attention of the research community. The operation of these SBRs within the PLANTEA platform has led to the identification of some of the main factors contributing to the formation and subsequent emission of N_2O during wastewater treatment. Current, research focuses on the development of mitigation strategies that could be implemented in wastewater treatment facilities with high N_2O emissions. Another controlled and monitored SBR is being operated to collect data on the efficacy of biological treatment of selected micropollutants and their transformation products. In long-term experiments, the main operative parameters needed to increase the removal of micropollutants will be evaluated, as well as their impact on activated sludge communities; at the protein level, the utility of proteomics in the study of these processes will be determined. Finally, a membrane bioreactor pilot plant will be used for purposes such as assessment of the effectiveness of using membrane technology for sludge thickening, studies of the removal of arsenic from groundwater, and techniques for membrane fouling minimization. Process data from this pilot plant are being used for the development of mechanistic and data-driven filtration models.

In the near future, PLANTEA will be extended to the facilities of a local WWTP located close to the ICRA. This will allow the installation of semi-industrial-scale pilot plants, which require large volumes of wastewater, to operate in the WWTP, thus facilitating their monitoring, sampling, and control.

Since the establishment of the PLANTEA facilities in 2011, several projects have benefited from its installations, such as Consolider-Ingenio SCARCE, MINECO project CARBONET, the SANITAS-Sustainable and Integrated Urban Water System Management (EU Initial Training Network Marie Curie), SGH-GEMS-Sulfide and Greenhouse Gas Emissions From Mediterranean Sewers (EU Marie Curie Reintegration grant), NITRI-GHG-Exploring Novel Nitrifying Pathways To Minimize Greenhouse Gas Emissions from WWTPs (EU Marie Curie Career Integration Grant), GEISTTAR-Understanding Fugitive Greenhouse Gas Emissions from Wastewater Transport and Treatment Systems (MINECO, Spanish Government), Study of the Mechanisms Involved in N_2O Production During Wastewater Treatment (MINECO, Internationalization projects, Spanish Government), VITEMESP-Feasibility Study of Membrane Technology for WAS Thickening (CDTI- Acciona), and MBR-Control-Development and Validation at Full Scale of an MBR Air-Scour Control System (CDTI-OHL MedioAmbiente INIMA SAU).

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Competing interests. None declared.

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