This project is part of the PRIMA programme supported by the European Union

PRIMA-SECTION 2-2022

"Modelling and Technological Tools to Prevent Surface and Ground-Water Bodies from Agricultural Non-Point Source Pollution Under Mediterranean Conditions"

NPP-SOL

Report on Designing CW

Deliverable number: D.2.2

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Non-Point Pollution SOLutions (NPP-SOL)

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Report on designing CW

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Date of approval	18/02/2025
Approved by project coordinator	A. Coppola
Date of approval	18/02/2025
Call	PRIMA Section 2 2022- Multi-topic Topic 2.1.1 RIA (Prevent and reduce land and water salinization and pollution due to agri-food activities).
Project URL:	https://npp-sol.iamm.ciheam.org/

1. Introduction

Environmental problems related to irrigated agriculture, such as salinization and nitrate pollution of surficial and groundwaters, are openly recognized in many areas of the Mediterranean. This is the case for example of the Ebro basin, in Spain and of the Gharb area, in Morocco.

In particular, the Arba River, one of the Ebro's tributaries and receiver of the Lerma gully waters, presented the highest increase in water salinity and nitrate pollution in the Ebro Basin during the period 1975-2004, as confirmed by the Ebro Basin Hydrological Authority (CHE, 2006, 2009). In fact, this river was the first surface water body declared as affected by nitrate pollution from agricultural sources by the Ebro Basin Authority (MMARM, 2011). Consequently, large areas of the Arba Basin were designated as Nitrate Vulnerable Zones from agricultural sources in 2008 by the Regional Government (BOA, 2009), according to the Spanish legislation and following the European Council Directive (91/676/EEC, 1991) concerning the protection of waters against pollution by nitrates from agricultural sources. The indications and implications of this directive were included in the posterior Water Framework and Groundwater Directives.

Similarly, in the Gharb plain, one of Morocco's main irrigated regions, agricultural intensification has contributed to soil degradation and the contamination of surface water and groundwater (Bendra et al., 2012). This is especially true in the Mnasra and Sidi Allal Tazi regions, interested by severe nitrate pollution in groundwater and surface water due to excessive fertilizer application, livestock waste runoff, and inefficient irrigation practices. This contamination poses significant environmental and public health risks, as nitrate concentrations in shallow wells frequently exceed the World Health Organization (WHO) threshold of 50 mg/L (Al-Qawati et al., 2018; Nouzha et al., 2016; Kanga et al., 2020). Rice cultivation is also practiced on grey alluvial soils. The fertilisers applied for rice growth are considered responsible for seasonal water nitrate peaks frequently observed in the area.

In the scope of NPP-SOL project, in both the area, Constructed Wetlands, CW, have been identified as potential bioremediation tools for removing nitrates from surface water. Constructed wetlands (CW) are promising low cost, low energy-requiring and simply to operate nature-based solution systems specifically designed to remove diverse pollutants from water by using mostly the processes that occur in natural wetlands, but within a more controlled environment. Specifically, a surface flow CWs consists of free surface water flowing horizontally through an artificial pond containing floating and/or emergent rooted vegetation and a high diversity of microorganisms (Ilyas and Masih, 2017; Sirivedhin and Gray, 2006; Vymazal, 2007). CWs offer a sustainable, cost-effective, and nature-based solution for mitigating nitrate pollution by utilizing sedimentation, microbial biodegradation, phytoremediation, and chemical precipitation to remove excess nutrients from agricultural runoff. A simplified scheme of a surface flow CW is given in **Error! Reference source not found.**.

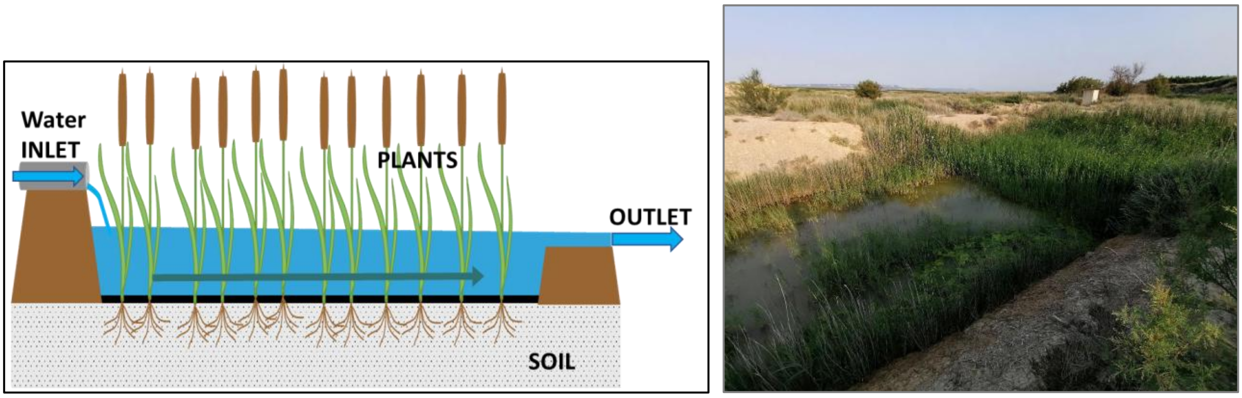

Figure 1. Schematic representation (left image) and a picture (right image) of a surface flow constructed wetland with emergent vegetation.

Among the diverse applications of this systems, directing agricultural runoff waters through a CW could be useful for removing nitrates and mitigate pollution in agricultural area. Apart from nitrate attenuation, CW can be useful for the removal of other contaminants, such as pesticides, pharmaceuticals, etc.

If a CW cannot provide enough organic carbon to support complete denitrification (e.g., from inlet water, soil, plant root exudates, and decomposed vegetal material), the addition of an external organic carbon source as an electron donor could enhance the heterotrophic denitrification efficiency (Lu et al., 2009; Si et al., 2018). Details on the main pollutant removal mechanisms occurring in a Constructed Wetland are given later in Section 3.

Within the scope of **NPP-SOL, the Pollution-Preventing Technology (PPT) operating in the Spanish Case Study of Lerma basin is a CW**, in which a **bio-stimulation strategy** is being adopted to increase the denitrification efficiency of the system. Its aim is to intercept and remove nutrients (and pesticides) from the Lerma Gully, polluted by runoff water from the agricultural fields of the basin and by seepage of shallow perched aquifers draining the agricultural returns flow, and to diminish thus the release of nitrates to the Arba River. The measured nitrate concentration in the Lerma Gully for the period 2015-2024 ranges from 30 to 140 mg/L depending on the irrigation and/or agricultural year.

The CW operating in the Moroccan case study is a natural drainage canal modified to be a multi-stage treatment system integrating wetlands into drainage networks. This system has been developed at the Sidi Allal Tazi Experimental Farm, managed by INRA-Morocco, situated north of Kenitra city (**Error! Reference source not found.**). The CW is proposed to receive drainage water from a 6.7-ha area mainly cultivated with rice. This area is irrigated by a flood irrigation system, with water coming from the river Sebou.

Details on the design, construction, operation, maintenance and monitoring of the CW in both sites are given in Sections 4, 5 and 6.

2. Description of the study areas

The Spanish study area description is inspired from Merchàn (2015). The Arba River Basin is located on the left bank of the middle Ebro River Valley, Spain. In the 2000s, approximately 20,000 ha of rainfed croplands of the Basin were transformed from dryland into irrigated agricultural land through a network of channels and ditches carrying water from the Aragón River (affluent of Ebro River) and its Yesa Reservoir (Pyrenees Mountain range, Figure 2), leading to a progressive increase in the use of fertilizers in the area (compound and liquid NPK fertilizers). The Lerma basin (zoom in Figure 2), a small watershed drained by the Lerma gully

toward the Arba river and located in the municipality of Ejea de los Caballeros (Zaragoza province), has been monitored since 2004 to assess the effects of this transformation on the water balance as well as on salt and nitrate (NO_3^-) exports (Merchàn et al., 2015; Causapé et al., 2023). The area of the basin is 7.38 km^2, with about 4.5 km long (from East-Southeast to West-Northwest) and around 2 km wide. Altitudes range from 335 to 495 m above sea level. About 48% of the basin, i.e. 352 ha, was included in the irrigation project. According to several studies carried out in the area (Merchán, PhD Thesis, 2015), the implementation of irrigation implied a three-fold increase in N export to the receiving Arba River, which was the first surface water body in the Ebro River Basin to be declared affected by NO_3^- pollution from agricultural sources according to the Nitrates Directive 91/676/EEC.
Within the NPP-SOL project, the Lerma basin – Spain - has been selected as Case Study for the implementation and validation of one of the selected Pollution Preventing Technology, aiming to intercept and remove agricultural pollutants before they reach water bodies.

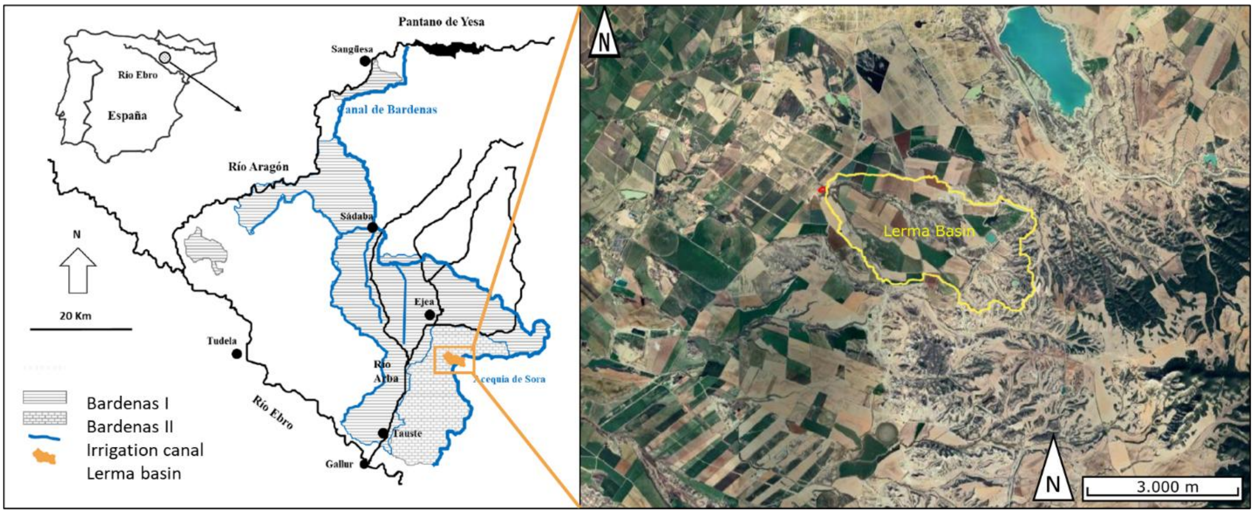

Figure 2. NPP-SOL Case Study area location in Ejea de los Caballeros, Zaragoza, Spain (42°03'35.4"N 1°08'02.4"W). In the left image, "Bardenas I" indicates the land sector in which both the transformation to irrigated-land and modernization of the irrigation systems (from open channels to pressurized systems) has been already completed, and "Bardenas II" indicates the land sector where transformation to irrigation is completed but its modernization is still ongoing.

The Moroccan case study is conducted at the Centre Régional de la Recherche Agronomique de Kénitra, specifically at the Domaine Expérimental de Sidi Allal Tazi. It lies in the Gharb-Chrarda-Beni Hssen region and is confined between the coastal zone (the Mnasra area) and Oued (river) Sebou in the northwestern part of Morocco. Its geographical coordinates are approximately 34°32'36" N latitude and 6°20'12" W longitude (Figure 3). The Mnasra region characterized by a Mediterranean climate with a pronounced oceanic influence, the study area experiences an average annual precipitation of approximately 551 mm. The rainy season generally extends from October to the end of April, peaking during November, December, and January. Temperature variations range from 12 °C during winter to 23 °C in summer. Notably, the potential evaporation exceeds 150 mm per month during the dry months from June to September, whereas it remains below 80 mm from December to February. Elevation levels fluctuate across the region, with the highest point reaching approximately 70 m above sea level.
The aquifer system in the Gharb Plain is intricate, comprising two primary superimposed aquifers. The first is the phreatic aquifer located in the central portion of the plain, while the second is the deep aquifer emerging

in the southern and western perimeters, extending beneath the shallow aquifer in the central area. Characterized by a thickness ranging from 60 to 200 meters, the deep aquifer is situated within a heterogeneous Plio-Villafranchian complex consisting of sands, sandstones, calcarenites, and conglomerates (Amharref et al., 2007). Conversely, the upper aquifer, with a thickness ranging from 20 to 100 meters, comprises heterogeneous Quaternary materials such as clays, silts, and sandy and/or silty sandy layers. Aquifer recharge primarily occurs through rainwater infiltration and the return of irrigation water.

The Mnasra region is distinguished by the prevalence of sandy-clay and silty-clay textures. It should also be noted that the soil of the region is occupied by heavy soils (vertisols and fluvisols).

Agriculture and livestock (sheep and cattle) are the main economic activities in Gharb area. Beyond nitrate contamination, the region also experiences groundwater pollution from pesticides, particularly organochlorines, with concentrations ranging from 0.03 to 0.3 µg/L (Al-Qawati et al., 2018). Pesticides are widely used without tailored application strategies, leading to the accumulation of active substances in the soil and their subsequent infiltration into water resources (Marouane et al., 2014)..

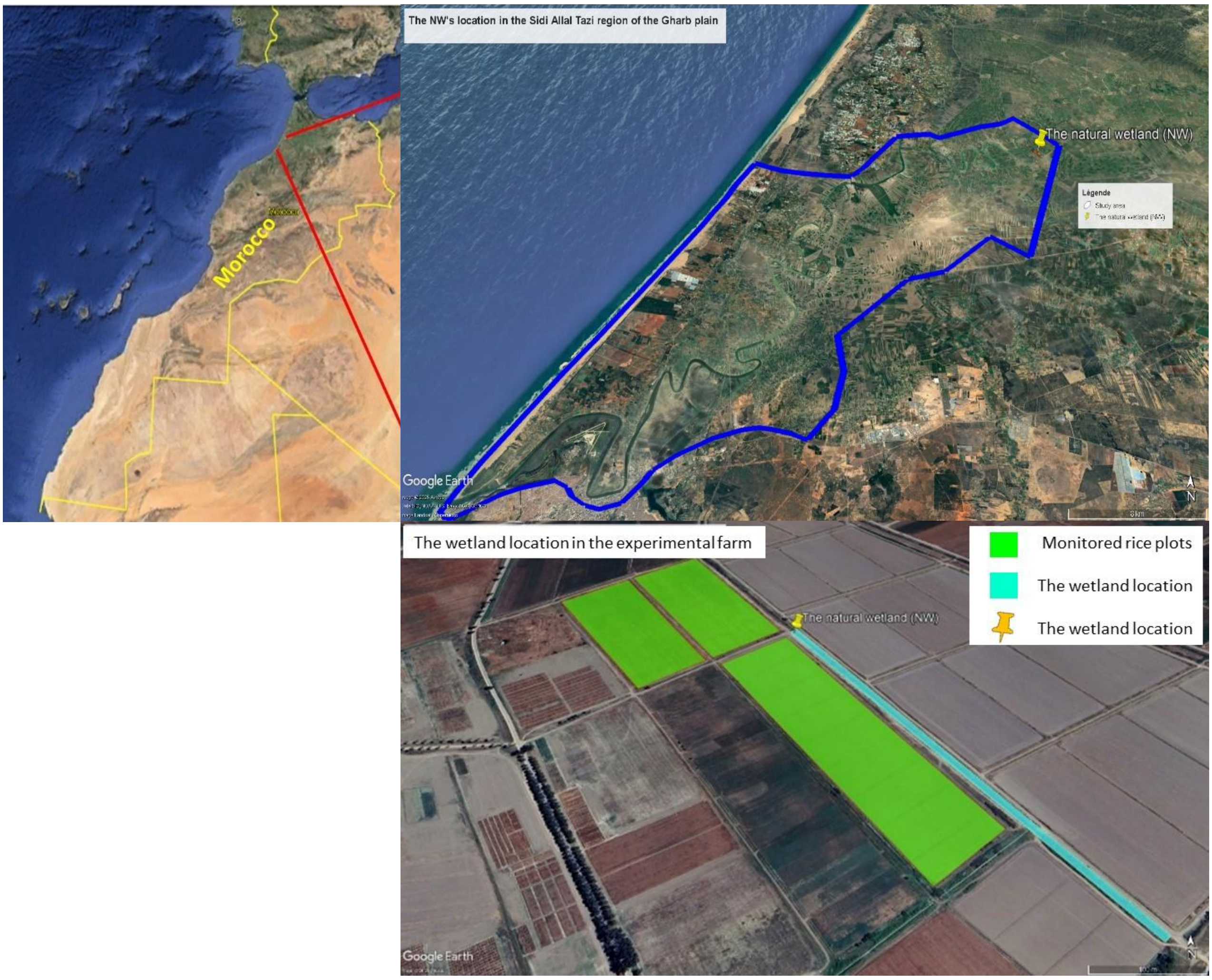

Figure 3. NPP-SOL Case Study area location of Morocco (blue border) and the fields (in green), cultivated with rice, draining into the CW (in light blue).

3. Pollutant removal mechanism

The main processes that might contribute to NO_3^- pollution mitigation in surface flow CWs are plant uptake, assimilation by microorganisms, and denitrification processes (Rogers et al., 1991). The latter refers to the reduction of NO_3^- by microorganisms through a series of enzymatic reactions involving the intermediates NO_2, NO, and N_2O, before finally being reduced to N_2 (Knowles, 1982). Parameters such as temperature, dissolved oxygen (O_2), NO_3^- loading, the source and amount of organic carbon (C), microbial species, the type and density of macrophytes, wetland age, and hydraulic conditions play key roles in the NO_3^- removal efficiency (Bachand and Horne, 1999; Beutel et al., 2009; Kong et al., 2009; Sirivedhin and Gray, 2006). Different approaches can be implemented to enhance water remediation, but strategies directed toward the induction of bacterial NO_3^- respiration are preferred since denitrification is an authentic N sink in water, unlike biomass sequestration (Scott et al., 2008). N storage by plants is generally considered temporary, because organic N returns to the system after the death and decay of plants if they are not harvested (Cooper and Cooke, 1984; Gumbricht, 1993).

In CWs, macrophytes are able not only to assimilate NO_3^-, but also to promote denitrification efficiency. Plants exert an influence on the diversity of microbial species and their enzymatic activities by releasing exudates and oxygen to the rhizosphere (Kong et al., 2009 and references therein), and decomposed plant material can be used by microbes as a source of organic carbon. For this reason, increased NO_3^- removal is usually found in vegetated CWs relative to that in non-vegetated systems (Jacobs and Harrison, 2014; Soana et al., 2017).

If the constructed wetland (CW) does not supply adequate organic carbon to fully support denitrification—whether from inlet water, soil, plant root exudates, or decomposed vegetation—supplementing with an external organic carbon source as an electron donor can improve the efficiency of heterotrophic denitrification (Lu et al., 2009; Si et al., 2018). Since the use of pure reagents such as glucose, acetate, or ethanol may be expensive in long-term treatments, the use of industrial or agricultural residues that are rich in organic carbon could represent a more sustainable solution for biostimulation. Solid products such as animal or vegetal waste (Grau-Martínez et al., 2017; Si et al., 2018; Trois et al., 2010), as well as industrial liquid by-products (Carrey et al., 2018; Margalef-Marti et al., 2019b), have already been reported as being useful for promoting denitrification. Liquid by-products are usually advantageous for its easier applications.

4. Designing a CW system

The CW at the Lerma Basin

It was constructed in October 2013 as nature-based solution to treat surface waters contaminated by agricultural activities. The surface water of the Lerma gully, which presents a variable water flow between 15 and 60 L/s and is contaminated by the agricultural return flows, can be totally or partially diverted towards the CW through a barrier installed at its inlet, which therefore allows to control the flow rate within the CW. The water, is afterwards given back to the Lerma gully after the treatment.

The system, initially covering an area of about 1500 m^2, was then enlarged in June 2017 to a final area of ~2500 m^2 with a water depth of ~40 cm. Emergent macrophytes started growing since its construction and reflect the old and new parts.

Within the NPP-SOL Project, at the beginning of year 2024 a gravel bed has been added to a small part of the CW located close to the discharging point, corresponding to approximately 20% of the CW surface. Four short piezometers have been installed inside the gravel bed during its filling, aiming to sample the subsurface water circulating through it during the CW operation (Figure 4). Therefore, currently the CW system relies on:

- Surface flow CW: aerobic zone with emergent macrophytes (Typha and Phragmites) growing since the CW construction, that works as natural green filter;
- Subsurface flow CW: area filled with gravel that, by minimizing the equilibrium with the atmosphere, might induce anoxic conditions. In this area, thus, a higher nutrients removal efficiency is expected.

Figure summarizes the different phases of construction, enlargement, vegetation growths and gravel disposal characterizing the CW facilities at Lerma basin.

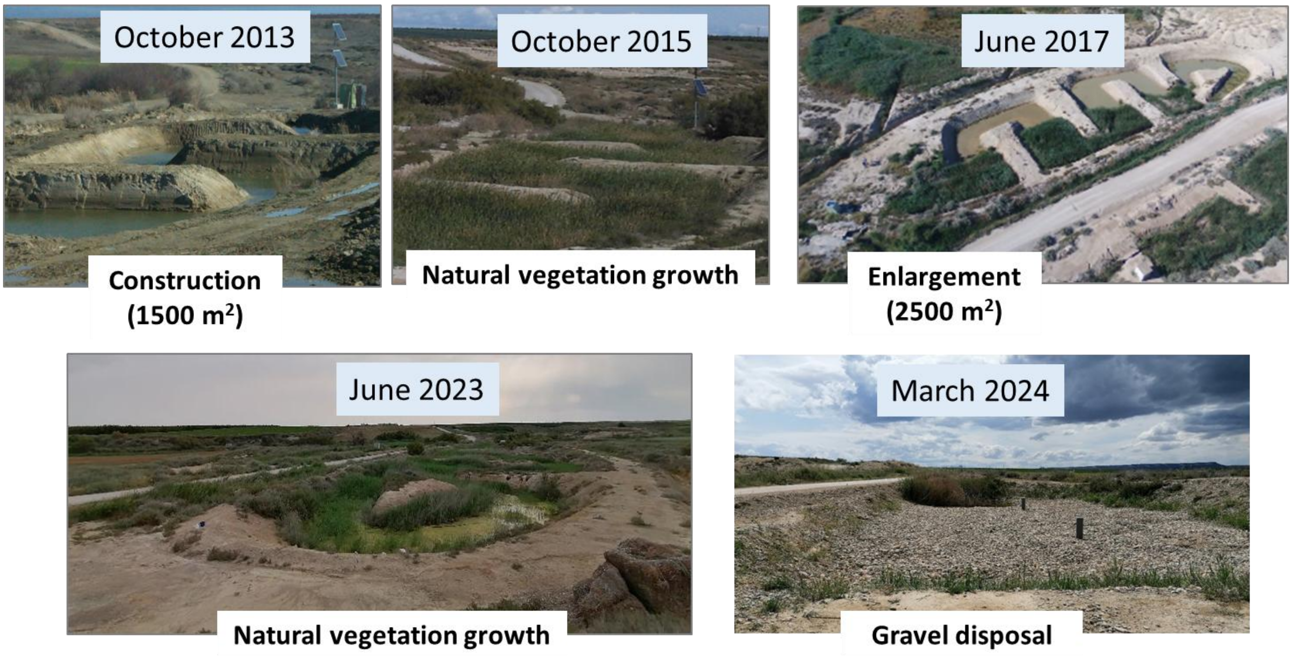

Figure 4. Construction, enlargement, vegetation growth and gravel disposal at the CW in Lerma.

New conductivity and temperature sensors have been installed at the beginning of year 2024 at the inlet and outlet of the CW for continuous monitor to complement already existing sensors for nitrate concentrations and water flow rate measurements. More details on the monitoring system are provided in section 6.

Also, aiming at injecting the selected organic carbon rich solution to the CW for the biostimulation experiments, a tank was installed close to its inlet (Figure), where an electrovalve allows to control the injection periodicity and flow.

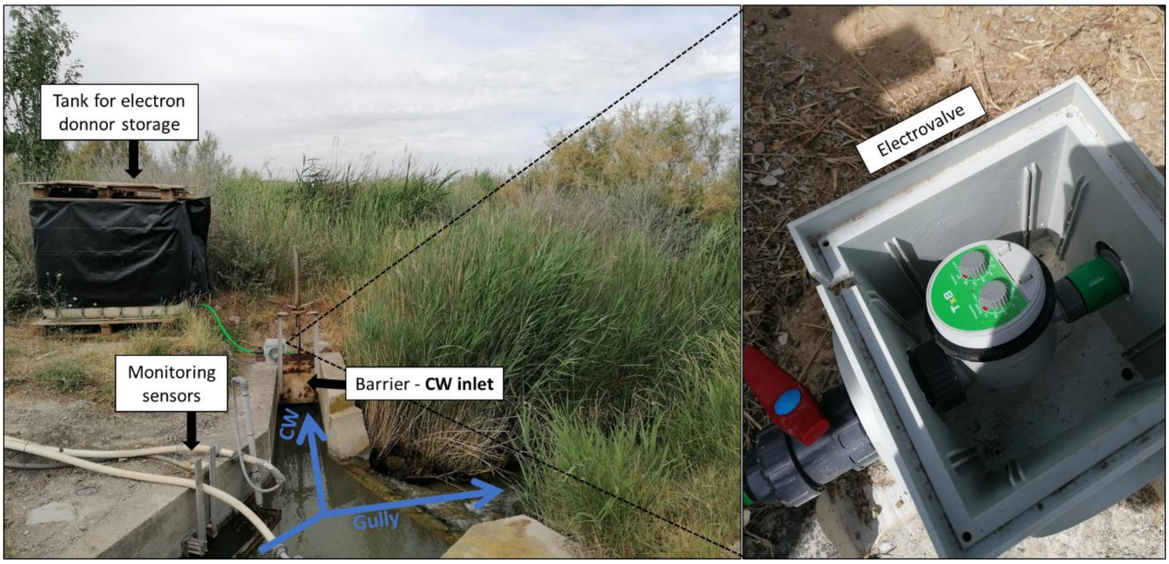

Figure 5. Detail of the CW inlet: on the left, the tank containing the organic C rich solution for the biostimulation, from which the liquid product is injected to the CW through the green plastic tube, controlled by an electrovalve (inside the grey box); and the monitoring sensors submerged in the inlet water; on the center, the barrier for regulating the flow of water from the Lerma gully to the CW; on the right, the Lerma gully natural riverbed (downstream).

Initial construction in 2013 and maintenance works at the CW facilities until 2023 (start of NPP-SOL project) have been carried on within the frame of different projects and funding: EcoRegadío (CGL2012-32395), REMEDIATION (CGL2014-57215-C4), AGRO-SOS (CGL2015-66016-R) and PACE-ISOTEC (CGL2017-87216-C4-1-R) financed by the Spanish Government and AEI/FEDER financed by the European Union; MAG (2017 SGR 1733) and MAGH (2021SGR00308) financed by the Catalan Government. Synergies and interactions are currently existing among NPP-SOL and the ongoing projects AgroSOSIII (PID2023-147588OB-I00) and Advance4Water-ISOTRACE (PID2022-139911OB-C41) financed by the Spanish Government and FEDER/EU.

The CW at the Sidi Allal Tazi Experimental Farm

In the scope of NPP-SOL project, the Constructed Wetland, CW, is a natural, vegetated drainage canal modified to be a multi-stage treatment system, exploiting the tortuosity of the drainage network. From the field, the water is firstly drained in smaller guard (secondary) canal, from where the water ends in the main drainage canal (the CW). The latter is 600 m long, 5 m in wide and 1.5 m deep and receives drainage water from a 6.7-ha area mainly cultivated with rice and irrigated by a flood irrigation system, with water coming from the river Sebou. Periodical emptying of the rice paddies, required for agronomic operations and weeding, induces periodical fluctuations of nitrates in the drainage water, which is proposed to be treated by the CW.

The main canal is a drainage canal constructed in downstream of a paddy rice plot ensuring a drainage of excess water during a paddy rice production season and winter heavy rainfall. The drainage canal has been modified to include layered substrates, such as accumulated sediment, which act as filtration media and provide a habitat for pollutant-degrading microorganisms. Vegetation zones, colonised with species such as *Phragmites australis*, *Tipha Latifolia*, and *Arundo Donax*, play a crucial role in nutrient absorption, while microbial activity within wetland substrates facilitates the breakdown of nitrates and pesticides, preventing their accumulation in groundwater (Yousaf et al., 2021). Other plant species found in the wetland, such as *Cyperus difformis*, *Saccharum spontaneum*, and *Tricholaena teneriffae*, also contribute to water filtration and ecosystem stability (Rathore et al., 2020). The Figure 6 provides some pictures of the canal and the field to be cultivated with rice.

Figure 6. The CW at the Sidi Allal Tazi Experimental Farm. The arrows indicate the path of the surface drainage water. The two figures on the right provides a detail of the vegetation mainly existing in the CW (see the text)

Water flow within these systems is naturally controlled, but structures like adjustable weirs or sluice gates can be added to regulate hydraulic retention time. This ensures that water remains in contact with the substrates and vegetation long enough for effective pollutant removal. The canal's length and gradual water flow enable a stepwise reduction in pollutant levels as water moves through the system.

For optimal integration of wetlands into drainage canals, the site has been chosen to be relatively flat to maintain even water distribution and reduce erosion risks. Proximity to sources of agricultural runoff, such as fields, ensures direct inflow into the wetland without requiring complex transport systems. Also, adequate space along the canal is also essential to accommodate wetland cells without compromising the canal's primary function of water conveyance.

To adapt the drainage canal to be a CW, the following steps were required:

– Canal Preparation: The canal was cleared of debris and reshaped to create defined zones for wetland cells. Embankments were constructed or reinforced to contain water and control erosion;

– Planting Vegetation: Species naturally grown in the drainage canals were planted, providing optimal spacing and density to enhance pollutant absorption and support habitat creation.

During operations, water control structures, such as weirs or gates, will be adjusted to regulate water levels and flow rates.

5. Operation and maintenance

Lerma Basin CW

Since its construction in 2013, the CW at Lerma basin has been operated intermittently. Its operation is uniquely regulated by the opening degree of the inlet barrier (Figure 5). Higher or lower water flow rates are achieved by opening more or less the barrier, respectively. In specific occasions the barrier is completely closed for maintenance works. Within the NPP-SOL project the CW has been operated continuously since December 2023 up to current date (December 2024) at a flow rate of approximately 5 L/s.

As observed in previous studies and reported in Margalef-Marti et al. (2019a), the CW presented a low denitrification efficiency when operating at a high flow rate (> 5 L/s) under natural conditions. Thus, a biostimulation strategy has been foreseen in the context of the NPP-SOL project to increase the denitrification efficiency at high flow rate operation. The strategy has been designed after evaluating the results of a series of laboratory experiments set-up for this purpose, which are described in detail below in the section "Laboratory experiments". As explained in the aforementioned section, a liquid by-product from the local agri-food sector has been finally selected for the biostimulation in the Lerma CW. Thus, after some preliminary tests conducted in May-June 2024, the field-scale application of biostimulation started in July 2024, consisting on periodic injections of the selected organic C rich solution at the CW inlet thanks to an automatic electrovalve (Figure 5). A weekly supply (filling the tank at the CW inlet, Figure 5) of the selected biostimulating by-product is ensured by a local factory, to fit the biostimulation strategy needs. The biostimulation has been continuously applied since July up to current date (December 2024), with the plan of continue it at least until September 2025. Different strategies including different injection periodicities and amounts are being tested. The details on the field scale application of the designed CW and results from the tested biostimulation strategies in the Lerma basin will be given further on, being object of the NPP-SOL Deliverable D3.2 (due at Month 30).

Before and within the NPP-SOL project, maintenance works are periodically carried out at the CW system. These include the periodic revision and calibration of the sensors installed at the inlet and outlet of the CW (conductivity, temperature, nitrate concentration and water flow rate. More details on the monitoring system are given in Section 6) as well as the revision of functioning and flow rate of the injecting electrovalve at the entrance of the CW. Other routine maintenance tasks at the CW during operation, involving the removal of algae or sediment accumulation at the inlet and outlet of the CW, checking the precision of the monitoring system sensors (by comparing the telematic measures with on-site measurements) as well as giving solution to potential minor damages and malfunctioning of the system, are carried out approximately once per month in strict collaboration with Dr. Jesús Causapé from the IGME-CSIC in Zaragoza, one of the main local stakeholders at the Spanish CW.

Lerma Basin CW: Laboratory experiments

Laboratory experiments has been conducted within the NPP-SOL project prior to the biostimulation in the study area, with the overall aim to test and optimize the induced denitrification strategy. One part of such experiments and their results have been published by Abu et al., 2024; another part is object of a currently ongoing PhD thesis at Group MAiMA-Universitat de Barcelona, so details will be published further on along the project.

These laboratory experiments included batch and column flow-through tests (Figure 7), both of which required water from the wetland inlet and gravel from the site to mimic field conditions. The goal of these tests was to stimulate the denitrification process, where nitrate (NO_3^-) is reduced to nitrogen gas (N_2) by microorganisms using the provided organic carbon. In the CW, denitrification is complementary to plant uptake with regards to N pollution attenuation.

The batch experiments involved the set-up of multiple microcosms (Figure 7), with each microcosm periodically sampled to monitor the denitrification process. Bottles were filled with contaminated water from the wetland inlet and gravel from the site. Various by-products rich in organic carbon, including two from the wine production and one whey from a cheese factory, were injected to determine the most efficient carbon sources to induce denitrification.

The batch experiments' results indicated that denitrification was successfully achieved within 48 to 72 hours with the two whey and only one of the wine residues, confirming that these carbon sources effectively promoted the desired microbial activity. To determine the optimal biostimulation strategy in terms of C/N ratio and injection periodicity, column experiments were set-up.

In the column flow-through experiments (Figure 7), water was forced upward through a glass column filled with gravel, and periodic injections of the organic carbon-rich by-products were conducted. This setup simulated a continuous flow system, allowing to adjust and optimize the carbon-to-nitrogen (C/N) ratio during the denitrification process to avoid an excess of dissolved organic carbon in the system after denitrification. This step is crucial to prevent potential negative effects, such as the accumulation of undesirable by-products (e.g., nitrite, ammonium, hydrogen sulphide, excessive biomass), while ensuring the system maintains effective denitrification.

Overall, both the batch and column flow-through experiments provided valuable insights into the efficiency of different organic carbon sources in promoting denitrification. These tests helped refine the biostimulation approach and allowed for better control over the C/N balance, ensuring an effective and environmentally sustainable denitrification process. Previous results from Margalef-Martí et al. (2019b) were also considered for the final selection of the substrates for the biostimulation at field scale. Finally, among the tested by-products, one of the two whey was chosen to be applied in the CW as the manufacturing industry is placed close to the study area and therefore, it decreases the transport costs.

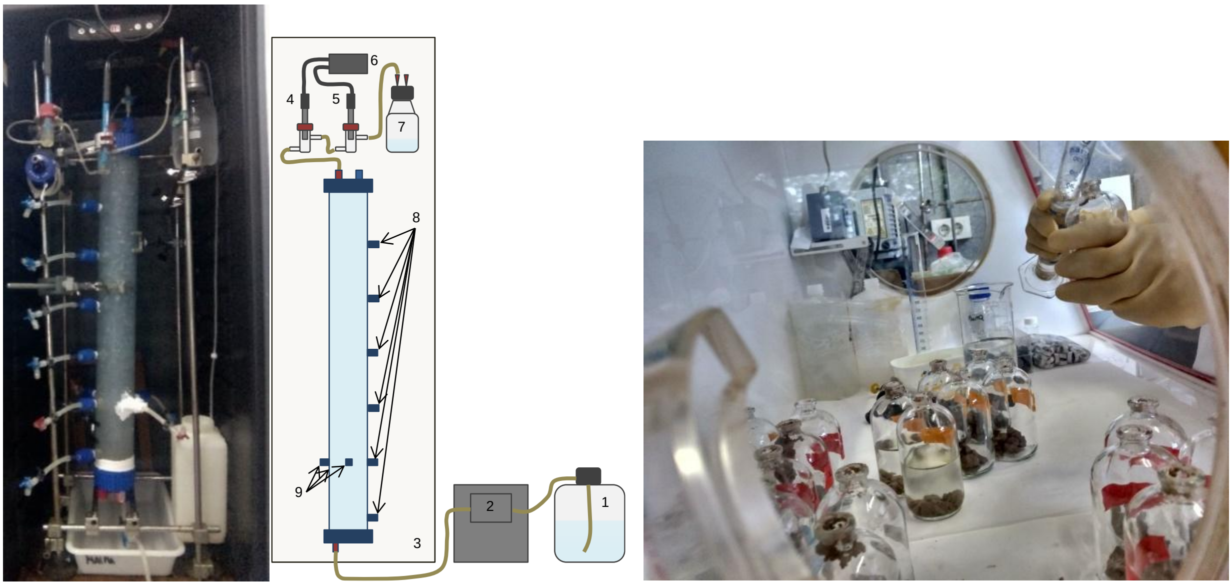

Figure 7. On the left, Scheme of the flow-through laboratory experiment: 1) inflow water, 2) peristaltic pump, 3) refrigerating chamber, 4) Eh probe, 5) pH probe, 6) multiparametric analyzer, 7) outflow water, 8) sampling points and 9) injection points. On the right: batch experiments inside the glove box

Sidi Allal Tazi CW

The following operations are considered crucial to ensure optimal operations of the CW at the Sidi Allal Tazi site.

- Managing vegetation and sediments in the canal;
- Maintaining hydraulic efficiency of the CW; and
- Regular monitoring and inspection of water quality parameters.

Periodic biomass harvesting prevents decomposing vegetation from reintroducing excess nutrients into the system, while the control of invasive species ensures ecological balance (Maranho and Gomes, 2024). Additionally, sediment and debris removal is necessary to maintain hydraulic efficiency (Adamo et al., 2021). The buildup of sediments can reduce wetland capacity, requiring periodic dredging, while clearing debris from inlet and outlet structures prevents blockages and maintains proper water flow.

Regular hydrology checks help detect flow irregularities, such as stagnant zones and channelization, which can reduce treatment effectiveness. Maintaining hydraulic structures, such as inlets, outlets, and flow regulators, is equally important. Regular inspections and cleaning help prevent uneven water distribution, which can significantly impact treatment performance. Seasonal adjustments to flow regulators accommodate fluctuations in water levels and ensure continuous functionality (Debroy et al., 2025).

Regular monitoring and inspection of water quality parameters, including nitrate, ammonia, pH and dissolved oxygen are critical for assessing pollutant removal efficiency.

Seasonal dynamics also influence wetland performance, necessitating seasonality adjustments in maintenance practices (David et al., 2023). During high-flow periods, overflow paths may need to be cleared, while drought conditions require closer monitoring to prevent vegetation desiccation (Tanner et al., 2022).

6. Monitoring

Lerma basin CW

The Spanish CW is equipped with the following monitoring systems:

- two gauging stations (inlet and outlet) including automatic sensors for the telematic measurement of water flow, nitrate concentration, conductivity and temperature (Figure 8). The on-site measurement system is configurated to obtain measures every 10 minutes. Remote sensing allows a real-time control of the CW operation conditions.
- Seven sampling points have been established for chemical and isotopic analyses of the surface/subsurface water circulating in the CW system: inlet, outlet, and 5 intermediate points (Figure 9). The sampling periodicity depends on the CW operation conditions, i.e. could be from weekly to monthly. The concentration of nitrate, nitrite, ammonium, organic and inorganic carbon, major and minor ions and metals is determined in the obtained samples as well as the isotopic composition of nitrate.

Figure 8. Gauging stations (inlet and outlet), automatic sensors and controllers for telematic measurements at the CW facilities

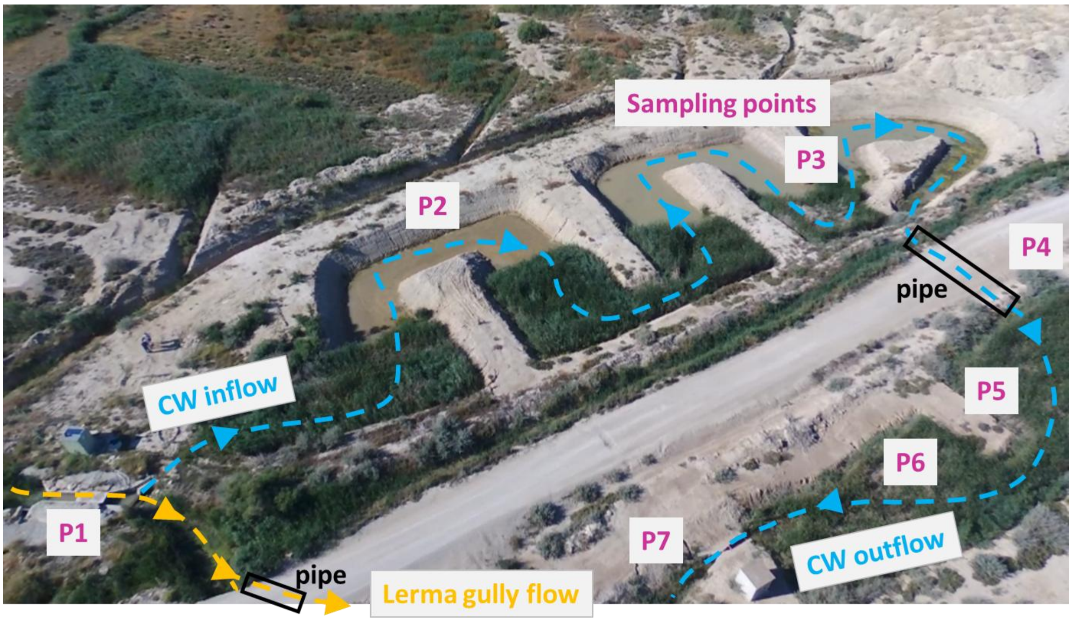

Figure 9: Schematic representation of the CW system and selected sampling points. P1: inlet; P2,P3 and P4: surface water sampling point; P5 and P6: subsurface water sampling point; P7: outlet.

The pollutant removal efficiency in CWs can be estimated by monitoring the inlet and outlet concentrations of the pollutant (Kovacic et al., 2000; Tanner et al., 2005; Uusheimo et al., 2018). However, this method does not reveal the specific processes involved in the attenuation, making it challenging to focus on the improvement of the wetland design and operation. Thus, hydrochemical characterization and multi-isotopic tools is being used at the Spanish CW for evaluating the efficiency of the on-site denitrification strategy. Namely, the NO_3^- isotopic characterization of water samples collected at the CW might improve the understanding and support the evaluation of the performance of the remediation strategy.

Actually, stable isotope analyses can provide information on the NO_3^- transformation pathways. In the course of denitrification, the unreacted residual NO_3^- becomes enriched in the heavy isotopes ^{15}N and ^{18}O, permitting the distinction between biological attenuation and other processes such as dilution which could also lead to decreases in concentration without influencing the isotopic signature (Aravena and Robertson, 1998; Böttcher et al., 1990; Fukada et al., 2003; Mariotti et al., 1981). In plants, significant enrichment in both ^{15}N and ^{18}O is observed in the NO_3^- extracted from leaves after uptake relative to the NO_3^- from water, but the changes in the NO_3^- isotopic composition in the water associated to this process are minor (Estrada et al., 2017; Spoelstra et al., 2010).

Monitoring technique and frequency at the Lerma basin CW

The remote sensing measurements are daily checked to detect any possible changes in the system. Sampling for water chemistry and isotopic composition is carried out by the Universitat de Barcelona (Figure 9) or by the Centro Tecnológico Agropecuario Cinco Villas, S.L. a technological centre settled in Ejea de los Caballeros (Zaragoza), close to the CW facilities. The frequency for sampling and corresponding analyses could vary from 1 per day, week or month, depending on the observed changes on real-time nitrate concentration measured with the monitoring sensors (more frequent in summer, where the activity of denitrifying bacteria is expected to be higher, and less frequent in autumn/winter where bacterial activity is expected to slow down).

The water samples are filtered with 0.2 µm syringe filters in less than 24h after obtention. Aliquots are prepared, conditioned and stored depending on the target analyses: samples for N compounds concentration and isotopic analyses are stored at -20ºC, samples for major and minor elements are acidified (HNO_3 1%) and stored at 4ºC, samples for organic carbon determination are acidified (pH 2-3) and stored at 4ºC, samples for inorganic carbon are stored at 4ºC filling completely the flasks to avoid free air headspace. Both chemical and isotopic analysis are carried out in the Centres Científics I Tecnològics de la Universitat de Barcelona (Scientific and Technological Centers of the University of Barcelona, CCiT-UB, https://www.ccit.ub.edu/EN/home.html).

Figure 9. Measurement of parameters, water sampling and preparation of samples before analysis at the CW facilities

Sidi Allal Tazi CW

Key monitoring activities in the Moroccan CW include:

- Water Quality at Inlet and Outlet

Regular water sampling at both the Upstream (inlet) and downstream (outlet) points is critical for evaluating the wetland's pollutant removal efficiency. Parameters such as Nitrate, phosphorus, pH and electrical conductivity (EC) are measured to ensure the system meets environmental quality standards. Sampling frequency depends on factors like seasonal variations, pollutant loads, and local regulations, with more frequent testing recommended during peak agricultural runoff periods.

- Vegetation Monitoring

The condition of wetland vegetation must be carefully monitored, as plants are critical for nutrient uptake and serve as habitats for microbial communities. Monitoring should include assessments of biomass density, species diversity, and the presence of invasive species, which can disrupt ecosystem balance.

– Seasonal and Environmental Monitoring

Seasonal changes significantly influence wetland operations, requiring adjustments to management strategies. For instance, clearing overflow paths during wet seasons and maintaining adequate water levels during dry periods are crucial to prevent vegetation desiccation and ensure continued functionality.

– Wildlife Monitoring

Wildlife interactions in wetlands are important indicators of ecosystem health. However, pest species, such as mosquitoes, should be monitored and controlled to prevent breeding in stagnant water areas, thus maintaining a balance between beneficial and detrimental species.

Monitoring technique and frequency at the Sidi Allal Tazi CW

Effective monitoring of nitrate pollution in the Moroccan CW requires a combination of sampling, in-situ measurements, and defined monitoring frequencies to assess water quality and pollutant removal efficiency. Water sampling and laboratory analysis are essential for tracking key parameters such as nitrate, ammonia, phosphate, pH, dissolved oxygen (DO) and EC. These samples will be collected at both the inlet and outlet of the wetland to evaluate changes in pollutant concentrations and treatment performance.

In addition to laboratory analysis, in-situ sensors provide real-time monitoring of critical water quality parameters, including DO, pH, and turbidity. Continuous monitoring probes enable the detection of fluctuations in water conditions, allowing for immediate corrective actions if necessary. The monitoring frequency will be adapted to seasonal variations and external factors influencing pollutant loads. Regular weekly or biweekly sampling will ensure consistent data collection, while additional samples will be taken during peak periods, such as intensive irrigation seasons or following heavy rainfall events when nitrate leaching is at its highest.

By integrating these monitoring techniques with an optimized frequency schedule, a well-designed program can provide comprehensive insights into the efficiency of wetlands in nitrate removal. Such an approach ensures that timely interventions can be implemented to maintain wetland functionality and protect water resources in the Gharb region

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