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**CONSTRUCTED WETLANDS FOR WATER MANAGEMENT AND REUSE IN  
AGRICULTURE**

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*“If we knew what it was we were doing, it would not be called research, would it?”*

*— Albert Einstein*

# Abstract

Global warming mainly induced by anthropogenic causes has already restricted the increase in agricultural productivity along with aggravating the conflict between water supply and demand. In this context, constructed wetlands (CWs) can be one of nature-based solutions for the treatment and reuse of unconventional wastewater sources. The aim of this research was to evaluate the performance of CWs in wastewater treatment (i.e., agricultural drainage water and domestic wastewater) and to explore the potential of their effluents for agricultural reuse. In order to achieve this goal, the work based on literature review and experimental approach was carried out. The findings showed that CWs can be an effective option for treating both agricultural drainage water (ADW) and domestic wastewater. It was also found that systems treating ADW can be effective even after a long period of operation. The treatment performance of CW systems is affected by the design and operational factors. For instance, the application of simple hydraulic structures and vegetation establishment can improve the pollutant removal efficiencies by increasing hydraulic retention time. Moreover, the addition of other technologies (e.g., UV treatment, anaerobic reactors) could further improve the quality of wastewater treated by single-stage CWs. In particular, it is strongly recommended to add special disinfection technologies to CW treatment systems to meet agricultural reuse standards since it was shown that the microbial loads often exceed the limits (e.g., *E. coli*).



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# **CHAPTER 1 - Introduction**



## 1.1. The impact of climate change on water resources and agriculture

Global climate is suffering unprecedented changes mainly due to the human activities, e.g. burning fossil fuels or deforestation (IPCC, 2021). The phenomena of climate change includes global warming, extreme weather events, sea level rise, etc. (Nema et al., 2012). NOAA (2021) reported that the average growth rate of the global surface temperature has reached 0.18 °C per decade since 1981. In IPCC (2014), it was shown that during the period 1951-2010, human-induced contribution almost accounted for more than half of the observed rise of earth's mean surface temperature. WMO (2021b) stated in 2020 that the global warming exceeded 1.2 °C above pre-industrial period. The findings by Lindsey (2021) reported that during 2006-2015 global average sea level went up by 3.6 mm yr<sup>-1</sup>, equal to 2.5 times the average rate (1.4 mm yr<sup>-1</sup>) observed during most of the 20th century. In addition, precipitation has been also affected. Dry areas are becoming drier while wet areas are becoming wetter. Besides, the more intense precipitation events even increase the risk of flooding (Trenberth, 2011).

The potential impacts of climate change span through many aspects, e.g., water resources, agricultural productivity, etc. (Gornall et al., 2010; Pokhrel et al., 2021). For example, the conflict between water supply and demand has worsened. It is predicted that by 2050 more than 5 billion people in the world will experience water shortages for at least one month each year (Global Commission on Adaptation, 2019). WMO (2021a) reported that in the past 20 years (2002-2021), terrestrial water storage decreased at a rate of 1 cm per year. Moreover, elevated water temperature and extreme weather events (e.g., floods and droughts) may cause water quality degradation and severe water pollution (Bates et al., 2008; WMO, 2021b). Climate change is thus considered as a primary driver of water scarcity and even further affects agriculture, energy and other sectors (Eslamian, 2016).

As the primary consumer of freshwater resources, agriculture is more sensitive than other economic sectors when confronting the threat of water scarcity (FAO, 2012). Specifically, it is greatly affected by climate change in direct and indirect ways, such as changes in precipitation and temperature, displacement of cultivation areas and soil loss, invasion by pests and invasive species. (EEA, 2019). It has decreased the yield growth of multiple crops worldwide (such as wheat, maize, etc.) and has further worsened food insecurity due to the more frequent variability and extreme conditions (Global Commission on Adaptation, 2019). In addition, it may also have negative effects

on livestock, such as water stress, food pressure, the spread of diseases through flooding, etc. (World Bank, 2013). Therefore, the use of non-conventional water resources is becoming an alternative solution for the water crisis in the conditions of climate change.

## 1.2. Wastewater reuse in agriculture

The reuse of treated wastewater, a type of non-conventional water resources, was identified as a potential contributor to water supply (Almuktar et al., 2018; Shoushtarian and Negahban-Azar, 2020). It can mitigate negative impacts on ecosystem caused by wastewater discharge and increase the availability of water resources (Gurel et al., 2007). Moreover, it is relatively reliable unlike other water resources affected by rainfall and climate conditions (Shoushtarian and Negahban-Azar, 2020).

Although the utilization of reclaimed wastewater is carried out in different sectors, agricultural reuse is the most common worldwide (Eslamian, 2016). Particularly, there is a great potential for irrigated agriculture, since it consumes 70% of world's freshwater withdrawals (Norton-Brandão et al., 2013).

Reclaimed water reuse in agriculture can not only recycle nutrients and reduce the use of fertilizer (WHO, 2006), but it can also save some freshwater resources and use them to supply other sectors (Chen et al., 2016). In general, it can have many environmental benefits if implemented and managed carefully (WHO, 2006).

However, some barriers and challenges may exist when treated wastewater is reused in agriculture, e.g., the possible environmental and human health risks, public acceptance of the reclaimed water (Kihila et al., 2014; Verlicchi et al., 2018). Specifically, irrigation with treated wastewater may affect the physicochemical properties of the soil and microbiota. Furthermore, the dissemination of contaminants can result in human health risks by direct contact and food chain (Becerra-Castro et al., 2015).

In this context, regulatory and institutional policies have been published by different regions and organizations of the world (USEPA, 2012). For instance, the United States Environmental Protection Agency (USEPA, 2012) stated different water quality requirements for irrigation based on crop types, i.e., food crops and processed food crops/non-food crops. Although the regulations adopted by different states of the USA are not the same, the regulations applied to non-food crop irrigation are generally less stringent than the ones for food crop irrigation. Furthermore, these regulations are more stringent than the water reuse requirements proposed by the WHO (2006). In 2020, European Commission (2020) approved a new regulation on minimum requirements for



water reuse. In this regulation, reclaimed water quality has been divided into four levels depending on both irrigation methods and crop types. It is expected to boost wastewater reuse in European Union once it enters into force on June 2023.

### 1.3. Water pollution

In order to achieve the goal of wastewater reuse, particular attention should be paid to the pollution causes and treatment measures of wastewaters. Water pollution is due to the release of harmful substances and energy into water environments, then resulting in changes in water nature and degradation of water quality (Von Sperling, 2007). Since water pollution is closely related to human health and economic development, it is of high importance for all the world areas (Mateo-Sagasta et al., 2017).

According to different ways in which pollutants enter receiving water bodies, water pollution can be divided into point-source pollution and non-point source (NPS) pollution. Point-source pollution, such as domestic and industrial wastewater, is defined as pollutants that enter water environments from a specific point (Von Sperling, 2007). NPS pollution (e.g., urban and agricultural runoff) refers to the release of pollutants that occurs in many different sites along water bodies (Lian et al., 2019; Von Sperling, 2007). Due to its characteristics of diffuse sources, it is generally believed that NPS pollution is more difficult to control than point-source pollution (Calijuri et al., 2011).

At present, NPS pollution has become the most serious threat to the water quality of aquatic ecosystems (Scholz and McIntyre, 2015). For example, it was reported by Jabbar and Grote (2019) that agricultural NPS pollution is the leading source of impairments to surface water in the USA. Similarly, Zhang (2010) found that in 2010, agricultural NPS pollution was responsible for 50% of China's total water pollution. Furthermore, it contributed 57 % of the total nitrogen (TN) and 67 % of the total phosphorus (TP). In Europe, agricultural sector is responsible for 50-80% of N and P freshwater pollution (Lankoski and Ollikainen, 2013).

### 1.4. Constructed wetlands for wastewater treatment

There are different technologies that can be used for treatment of polluted waters, but nature-based solutions (NBS) gained a lot of attention in the recent years since they have many benefits (e.g., reducing public health costs and disaster risks). Among the different types of NBS, constructed wetlands (CWs) are the most commonly used type (Oral et al., 2020). CWs are engineered systems that have been designed and built to treat different types of water, especially for domestic

wastewater, agricultural drainage water (ADW) and industrial wastewater (Arden and Ma, 2018; Vymazal, 2005). The processes of wastewater treatment include a series of physical, chemical and biological mechanisms, e.g., sedimentation, plant uptake and bacterial activities (Menon and Holland, 2013; Morato et al., 2014). Moreover, they involve the interactions among different components of the system, e.g., wetland plants, substrates and microorganisms (Sandoval-Herazo et al., 2018; Wirasnita et al., 2018; Zhang et al., 2012). On the other hand, the treatment performance of CW systems can also be affected by different factors, such as water flow regime, hydraulic retention time (HRT), etc. (Malyan et al., 2021).

The first application of a full-scale CW was in the late 1960s for treating wastewater from a Dutch camping site (Gregoire et al., 2009; Liu et al., 2015). Owing to the growth of financial incentive and public acceptance of this “green” technique, CWs for wastewater treatment started to develop intensively in the 1990s, which was largely driven by severe climate change (Lee et al., 2009).

CWs can be classified into free water surface (FWS) and subsurface flow (SSF) systems in terms of water hydrology (Vymazal, 2010). Specifically, one of the most obvious characteristics of FWS CWs is the open water surface (Kadlec and Wallace, 2008), which implies that water flows above the substrate layer. Unlike FWS CWs, water surface of SSF CWs is invisible and is beneath the surface of substrates, which thus minimizes the risk of pollutant exposure (Kadlec and Wallace, 2008; White, 2013). On the other hand, according to types of dominant plants in CWs, the classification comprises CWs with emergent, submerged, floating leaved and free-floating macrophytes (Brix and Schierup, 1989). Generally, emergent macrophytes are frequently used by a majority of CWs for wastewater treatment (Vymazal, 2005), especially the species such as *Phragmites australis* and *Typha latifolia* (Liu et al., 2009), since they cannot only tolerate wastewater environment, but can also reach good removal efficiency of pollutants (Fountoulakis et al., 2017).

In addition to single CWs, the application of hybrid CWs is also widely documented (Nan et al., 2020). The combination of different CWs is potential for a greater removal efficiency of pollutants (e.g., nutrients, metals and pesticides) and further improve in wastewater quality (Kadlec and Wallace, 2008; Malyan et al., 2021).

To date, the treatment and reuse of domestic wastewater (i.e., greywater and blackwater) has showed a large potential for freshwater saving (Ramprasad et al., 2017). Especially, greywater has gained special attention due to its higher availability and low pollutant strengthen (Patil and Munavalli, 2016). On the other hand, it is reported that ADW coming from farm land contributes to the direct transport of pollutants to surface water bodies, especially for nutrients and pesticides, threatening the water quality of ecosystems in different regions (Lavrić et al., 2020; Vymazal and

Březinová, 2015). In this context, increasing research on the treatment of ADW or domestic wastewater by using CWs have been carried out.

## 1.5. Objective of this research

The intention of this thesis is to highlight the effectiveness of CWs for treating ADW or domestic wastewater. Based on the literature findings and a specific CW case study, the author would like to provide some suggestions on CW design and operation in order to help CWs to achieve their full potential and even prolong their lifespan.

The specific objectives of the research are to:

- Assess the performance of CWs for ADW treatment after long-term operation
- Analyze the possible impact of main CW design and operational factors
- Analyze the features and treatment efficacy of treatment systems based on CWs (i.e., single CWs, hybrid CWs and the combination of CWs and other technologies)
- Assess the potential of water treated by CWs for agricultural reuse

## 1.6. Thesis structure

This thesis consists of six chapters, respectively focusing on the two research points (i.e., ADW treatment and domestic wastewater treatment).

**Chapter 1** provides a brief introduction on the use and application of CWs for polluted water treatment (ADW and domestic wastewater) and their effluent reuse in agriculture.

**Chapter 2** analyzes the design and operational factors of CWs for treating ADW. In order to verify their effects on full-scale CWs after long-term operation, a search of related literature was conducted. A total of 15 case studies were selected and discussed based on treatment performance, advantages and possible improvements of the studied CWs. The study explored the effects of design and operation factors and to further understand the actual performance of the CWs which operated for a long period of time. The findings may thus help CW systems to maximize their potential for ADW treatment.

**Chapter 3** introduces a case study conducted in the Emilia-Romagna region (Northern Italy). It explored the overall performance of a full-scale FWS CW, which has already treated ADW for 20 years. The findings showed that this CW still maintained a good treatment efficiency after two decades, particularly for the removal of pollutants total suspended solids (TSS) (up to 82%), TN (up to 78%) and  $\text{NO}_3^-$ -N (up to 78%). This study can provide evidence for the effectiveness of CWs for

the long-term treatment of ADW.

**Chapter 4** explored the potential of CWs for domestic wastewater treatment and their effluents reuse in agriculture. It reviewed 39 experimental case studies from 2008 to 2019. The CW treatment systems included single-stage CWs, hybrid CWs and the combination of CWs and other technologies. The purpose of this review was to 1) analyze pollutant removal efficiencies of the systems with different configurations and characteristics, and 2) assess the potential of treated wastewater for agricultural reuse based on the EU regulations. The findings demonstrated that the combination of CWs with additional technologies can further increase their performance and provide better pollutant removal efficiencies. However, it is strongly recommended to take disinfection measures before using treated wastewater for agricultural irrigation.

**Chapter 5** introduces an ongoing case study and provides a clear overview of the design, test and start-up of the CW units for domestic wastewater treatment conducted at the pilot plant within a wastewater treatment plant near the city of Bologna, in Italy. This experimental facility is part of the FIT4REUSE project and it aims to optimize CWs and to enable them to treat domestic wastewater up to the standards for reuse in agriculture. So far, the experimental activities and data analysis are still in progress.

**Chapter 6** provides a conclusion based on the main findings of the previous chapters. It highlights the effectiveness of CWs for treating ADW and domestic wastewater and provides the suggestions for maximizing the potential of CW treatment systems.

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# CHAPTER 2 - Effects of design and operational conditions on long-term performance of constructed wetlands for agricultural pollution control

This chapter is based on a manuscript in preparation.

*Key words: Constructed wetland; Agricultural drainage water; Design and operational factors; Long-term operation*

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# Abstract

Constructed wetlands (CWs) can be considered as an efficient nature-based solution for treatment of agricultural drainage water and consequently for the mitigation of the non-point source pollution. Aiming to provide suggestions for the construction and implementation of CWs, this paper proposes and discusses key parameters of CW design and operation. In order to verify the effect of these features, a total of 15 long-term field studies were reviewed, focusing on the performance of full-scale CWs that are treating agricultural drainage water. The findings showed that design and operational factors (e.g., the application of simple hydraulic structures and vegetation establishment) can improve the pollutant removal efficiencies by increasing hydraulic retention time. Hydraulic efficiency of CWs can also be enhanced through certain shape characteristics (e.g., adoption of a high aspect ratio, creation a long and narrow CW shape). The careful consideration of these parameters before and during CW implementation can therefore help these systems to achieve their full potential. However, further study is recommended for uncertain effects of some parameters (e.g., flow direction and the application of deep zones).

## 2.1. Introduction

Agricultural drainage water (ADW) is one of the leading non-point sources (NPS) of pollution (Budd et al., 2011; Karpuzcu and Stringfellow, 2012). The transportation of agricultural pollutants causes water quality impairment on the side of receiving water bodies (e.g. eutrophication) (Budd et al., 2011; Carstensen et al., 2019; Johannesson et al., 2017), and increases human health risks (D áz et al., 2012).

As a countermeasure, CWs are capable of managing NPS pollution and attenuating the loads of agricultural contaminants through intercepting water flow (D áz et al., 2010; Imfeld et al., 2013; Tournebize et al., 2015; Tournebize et al., 2017). Apart from the advantages of simple and low-cost operation (Beutel et al., 2013; Margalef-Marti et al., 2019), these ecological treatment systems can also provide diverse services, e.g., flood protection, wildlife habitat, groundwater recharge, aesthetic and recreational values (Lavrić et al., 2018; Lenhart et al., 2016; Maynard et al., 2011; McLaughlin and Cohen, 2013). Therefore, CW systems are an object of a growing interest and have been increasingly applied in agricultural landscapes worldwide (Abbassi et al., 2011; Dal Ferro et al., 2018).

To date, many studies have documented CWs as an efficient and promising method for reducing nutrients and pesticides from agricultural runoff and drainage water (Brauer et al., 2015; Budd et al., 2011; Calvo-Cubero et al., 2014). However, wide variability of pollutant removal efficiencies was observed in these systems (Crumpton et al., 2020; D áz et al., 2012; O'Geen et al., 2010). For example, the reported removal efficiency of pesticides was in the range 0-100% (O'Geen et al., 2010), while a similar ratio was found for total phosphorus (TP) (e.g., 3-80% measured in free water surface CWs (FWS CWs)) (Kill et al., 2018; Reinhardt et al., 2005). Most studies stated that N removal efficiency was generally within the range from 35% to 55% (Brauer et al., 2015), although some extreme values of nitrate removal efficiency (e.g., from negative values up to 98%) were also detected (O'Geen et al., 2010). The wide range of these pollutant retention efficiencies can be attributed to diverse factors, such as CW design and operation (e.g., location, hydraulic retention time (HRT) and vegetation characteristics), meteorological condition (e.g., climate, temperature), pollutant loading, seasonality, annual variations in water flow and dissolved oxygen concentration (Brauer et al., 2015; Kynk äänniemi et al., 2013). Furthermore, it is emphasized that some processes regarding nutrient removal may be temporary, such as nutrient uptake by plants and retention of Fe-bound P in the sediment (Margalef-Marti et al., 2019; Mendes et al., 2018a; Mendes et al., 2018b), which can also contribute to the variation of removal efficiencies. Moreover, it was

suggested that the treatment capacity of CWs was associated with their age (White, 2018).

Given the potential benefits of CWs for treating ADW and a lack of specific guidelines and related research dedicated to design and operational factors (Ioannidou and Pearson, 2018; Soana et al., 2020), further information on how these parameters affect the performance of CWs is needed in order to maximize the removal efficiency of pollutants in agricultural water. With this aim, an increasing number of studies have been carried out worldwide on the implementation of CWs, even if these systems were observed to be often not capable to have long-term operation period and so far relatively little is known about their sustainable performance (Groh et al., 2015; Nilsson et al., 2020). Therefore, the effectiveness of these systems after long-term operation is also something that needs further investigation.

In this context, in the present work a review of a series of field studies on the treatment of ADW was carried out, aiming to give better insight to the above-mentioned problems. The general objectives of the present review were to: (i) identify the main design and operational parameters that can influence the performance of CWs and (ii) evaluate the pollutant retention efficiency of long-lifespan CW systems, analyzing the influence of design and operational factors.

## 2.2. Materials and methods

In order to discuss and assess design and operational conditions for CWs treating ADW, full-scale studies published in the period 2010-2020 were considered since it was reported that only the implementation of field-scale CW treatment systems can precisely verify the actual systematic performance impacted by certain design parameters (White, 2018). Moreover, only the systems that were constructed at least 5 years before the reported monitoring period were taken into account in order to avoid the possible biases due to the specific conditions of start-up and transitional periods. The literature retrieval was conducted in November 2020 by using two main scientific databases Scopus (<https://www.scopus.com/>) and Web of Science (<http://apps.webofknowledge.com/>). The combination of keywords “constructed wetland”, “treatment wetland”, “tile drainage”, “agricultural drainage” and “agricultural runoff” was used to perform the search. The flow diagram of Fig. 2.1 presents the detailed information on the literature screening processes.

Therefore, while the section 2.3 provided primary considerations for design and operation of CWs treating ADW, section 2.4 overviewed a total number of 15 case studies selected to better understand the long-term effectiveness and sustainability of fully established and long-lifespan full-scale CW systems. As long-lifespan systems were considered those that were at least 5 years old at the time of monitoring. Most of the 15 studies focused on the investigation of FWS CWs, since

these systems are most designed and implemented for treating water from agricultural sectors (Tournebize et al., 2017; Vymazal and Březinová, 2018). The age of these reported CW systems was ranging from 5 to 18 years. Moreover, referring to the design and operational recommendations proposed in section 2.3, the potential relationship between the systematic functionality and specific design and operational factors of these CW systems was analyzed.

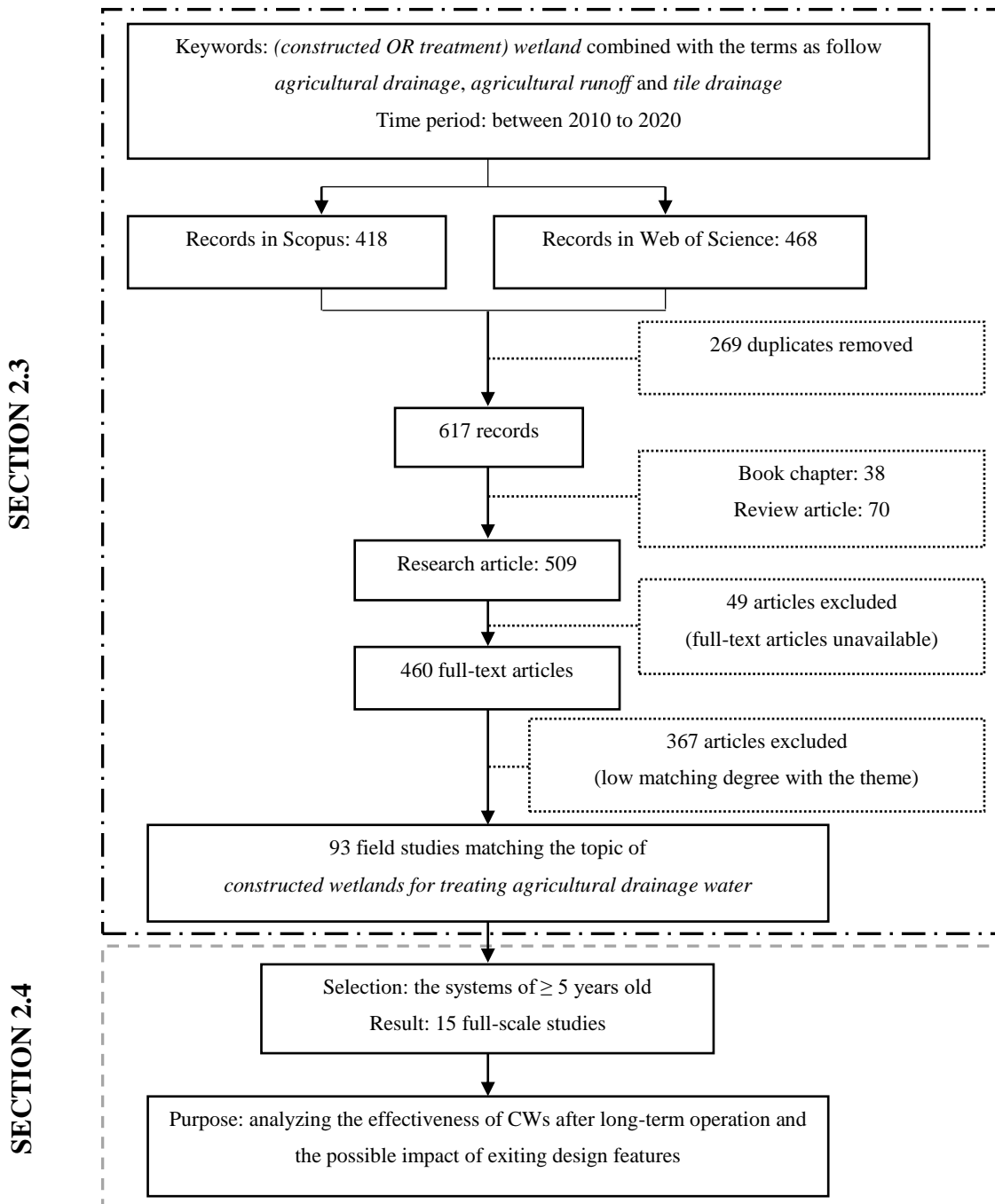


Fig. 2.1 - Outline of the review methodology.

## 2.3. The impact of design and operational factors

Below are listed the key points for CW design and operation that were frequently documented in literature. The importance of these characteristics is highlighted and discussed in detail in the following sections.

- Local climate condition and seasonal variation (section 2.3.1)
- CW shape (section 2.3.1)
- Wetland-to-catchment and length-to-width ratios of CWs (section 2.3.1)
- Flow direction (section 2.3.2)
- Application of simple hydraulic structures (e.g. gated spillways, baffle curtains) (section 2.3.2)
- Configuration of inlet and outlet points (section 2.3.2)
- Vegetation establishment (section 2.3.3)
- Regular harvesting of CW vegetation (section 2.3.3)

### 2.3.1. Location and size

Location of a CW and local conditions were reported as the prerequisites for design by Koskiaho and Puustinen (2019). Before determining a site where CWs should be built, detailed investigation and assessment is generally recommended to be taken related to the features such as geology, topography, and soil layers, which were suggested as important for pollutant removal efficiencies (Lenhart et al., 2016; Tournebize et al., 2017). However, also site characteristics as hydrology and climate conditions that influence it were proven to be of particular importance (Tanner and Kadlec, 2013).

For example, Steidl et al. (2019) assessed a FWS CW in north-eastern Germany that served for mitigating nitrogen loads from agricultural drainage before discharging it into the river. From October 2013 to May 2017, they monitored the treatment performance of the system on nitrogen removal. The results showed that total nitrogen (TN) retention efficiency ranged from 0.2% to 8.9%, much lower than the values of other CWs reported in other studies (Dal Ferro et al., 2018; Kadlec et al., 2010). It could be attributed to the influence of inner-annual distribution of outflows from the local agricultural field. Specifically, the CW received a large proportion (>70%) of the annual nitrogen loads in winter, when sediment temperature lower than 10 °C limited plant uptake and microbial activities, resulting in low N reduction.

Similarly, in the study of Koskiaho and Puustinen (2019), a CW located in Finland was strongly affected by seasonal variations. Particularly, performance of the CW from summer to autumn

dominated water purification processes throughout the year, whilst winter efficiencies were extremely low. The author attributed this difference to the severe Finnish weather condition (e.g. mean winter temperature below 0 °C), and further proposed that if a CW of similar dimension were constructed in other regions in southern Europe, the effectiveness would likely be enhanced.

It was found that the ratio between the areas covered by a CW and the catchment area (“wetland-to-catchment ratio”) also affected the removal efficiencies. As expected, CWs with larger surface area and consequently larger wetland-to-catchment ratio can achieve greater treatment performance by having longer HRT (Moreno-Mateos et al., 2010; Steidl et al., 2019; Tournebize et al., 2017). The comparison on two Finish CWs done by Koskiaho and Puustinen (2019) showed that the one with wetland-to-catchment ratio of 5% had much better annual removal efficiencies (74% for total suspended solids (TSS), 58% for TP and 54% for TN) than the one with wetland-to-catchment ratio of 1.3% (7% for TSS, 12% for TP and 9% for TN). The authors argued that these results can be largely attributed to the effect of wetland-to-catchment ratios.

In the case study of Steidl et al. (2019) aforementioned, the wetland-to-catchment ratio was much smaller and in the range of 0.4% - 0.5%, although it was the largest possible ratio according to the local conditions. Indeed, the TN removals were considerably low, less than 5% in most of the monitoring time. The optimal range of wetland-to-catchment ratios recommended by several studies was within 1-8% to reach 50% N removal (Garnier et al., 2014; Ligi et al., 2015; Tanner and Kadlec, 2013), or within 1.5-4% suggested by Moreno-Mateos et al. (2010) for ideal nitrate removal.

However, Moreno-Mateos et al. (2010) indicated a reasonable wetland-to-catchment ratio alone was not yet adequate for a satisfactory effect of water purification. The wetlands studied by Moreno-Mateos et al. (2010) located in the Ebro River basin, Spain, equivalent to 5.8% of the catchment, achieved significant nitrate removal rates ranging from 76 g N m<sup>-2</sup> per year to 227 g N m<sup>-2</sup> per year, but these values were not greater than the ones of Norwegian CWs in cold climates (50-285 g N m<sup>-2</sup>) (Braskerud, 2002). Therefore, the authors stressed it was still necessary to consider other requirements (e.g., morphological design).

Tournebize et al. (2017) and Kynkäänniemi et al. (2013) pointed out that the shape of CWs should be designed in order to be suitable for the landscape. For instance, different shapes were applied such as the widely used rectangular (Ioannidou and Pearson, 2018; Song et al., 2019; Tournebize et al., 2017), trapezoidal (Dal Ferro et al., 2018), ellipsoidal (Maynard et al., 2014), but also irregular shape (Ioannidou and Pearson, 2019; Johannesson et al., 2017). Especially, the long and narrow shape was shown to be advantageous to hydraulic efficiency (Kynkäänniemi et al., 2013). That is to say, the aspect (length-to-width) ratio could affect the hydraulic performance of CW systems

(Kadlec and Wallace, 2008; Su et al., 2009). For example, Lavrnić et al. (2020a) suggested that the creation of successive meanders in the studied FWS CW of a rectangular shape led to a high aspect ratio (52:1), thus the system had less dead zones and reached effective flow distribution. An Irish study reported that only when the aspect ratio of the integrated CW they investigated is lower than 2.2 can the effluent P concentration be reduced to the desirable value of below 1 mg/L (Scholz et al., 2007). Furthermore, the authors argued that the greater treatment effect could be achieved with the ratio closer to 1. However, there were different opinions on effective aspect ratios. For example, to reach laminar flow the recommended aspect ratio by Tao et al. (2014) ranged from 3 to 6, whilst Su et al. (2009) suggested the optimal ratio would be larger than 5, or at least 1.88 for maintaining uniform flow. Despite different aspect ratio values proposed, Kadlec and Wallace (2008) argued that there is no specific requirements for deciding aspect ratios as long as it can be within a reasonable range (e.g. from 2 to 10).

White (2018) investigated the effect of deep zones in two FWS CWs in USA. CW1 was made up of both shallow cells (average water depth of 20.3 cm) and deep cells (average water depth of 76.2 cm), while CW2 had successive deep zones with an average water depth of 80 cm. It was found that in CW1, deep zones contributed to the most of N removal in the systems. The overall removal efficiency in CW1 (65.1%) was not better than the one measured in CW2 (74.4%), which was attributed to the combined effect of several factors (e.g., water depth, HRT, inlet concentration and plant species richness) explained by the authors. Regarding water depth, there is still no clear conclusion on benefits of the application of deep cells in FWS CWs (Kadlec, 2007).

In summary, it is encouraged to consider seasonal discharge regime before CW site selection and define both the wetland-to-catchment ratio and the shape of CW systems based on local conditions (Steidl et al., 2019; Tournebize et al., 2017). Particularly, the lowest wetland-to-catchment ratio was suggested to be 1% for CWs with an average water depth of 0.8 m (Tournebize et al., 2017). The length to width aspect ratio should be within a reasonable range, e.g.  $2 < L:W < 10$  (Kadlec and Wallace, 2008).

### 2.3.2. Hydraulic design and CW configuration

The hydraulics of CW systems can not only determine the distribution of contaminants, but can also affect removal efficiencies (Pugliese et al., 2020). Regarding subsurface flow CWs (SSF CWs), Hoffmann et al. (2019) applied three types of flow direction (i.e., horizontal flow, up-flow, down-flow) in a total of six woodchip-based CWs with the same dimension ( $L \times W \times D$ :  $10 \times 10 \times 1$  m). After a two-years monitoring period, they observed a difference in the range of 12-15% for N removal efficiency between the most and least performing CWs, under similar hydraulic loading rates (HLRs) and water temperatures. It was found that the horizontal CWs showed the best N



removal efficiency, followed by down-flow CWs and up-flow ones. Furthermore, the impact of flow designs on N removal was in accordance with the hydraulic efficiencies (the best achieved by horizontal CWs and the worst achieved by vertical upward CWs). It thus demonstrated flow directions could affect the treatment capacity of CW systems due to the variation of hydraulic efficiency within the systems. In contrast, it was not the case in Bruun et al. (2016), which investigated the same systems. The authors reported that the vertical downward SSF CW achieved the highest removal rate of  $3.64 \text{ g N m}^{-2} \text{ d}^{-1}$  under the low flow rate applied ( $0.49 \text{ L s}^{-1}$ ), while the horizontal CW had the highest removal rate of  $10.5 \text{ g N m}^{-2} \text{ d}^{-1}$  among all the systems (horizontal, vertical upward and vertical downward CWs) under the high flow rate of  $1.83 \text{ L s}^{-1}$ . Such difference of findings could be attributed to the different conditions applied in the two studies. For instance, the study of Bruun et al. (2016) was under two fixed flow rates ( $0.49$  and  $1.83 \text{ L s}^{-1}$ ), whilst Hoffmann et al. (2019) was conducted under natural conditions with daily fluctuation of HLRs. Therefore, for the optimum performance, flow direction and HLR should be considered together.

As documented by Bruun et al. (2017), the direction of flow path was also notably affecting the export ratio of the greenhouse gas methane. The most effective mitigation of  $\text{CH}_4$  emission was observed in the vertical down-flow systems as a result of offsetting the upward diffusional transport of dissolved  $\text{CH}_4$ . Another case study for treating drainage water from a hydroponic farm in Jordan, by Abbassi et al. (2011), presented that the vertical CW had overall better effectiveness on pollutant removals (e.g.  $\text{BOD}_5$ , COD, nutrients) than the horizontal one, under a series of HRTs applied. It can be explained by the fact that distribution of water flow within the vertical system led to greater contact with the substrates and plants, therefore promoting treatment effects.

In view of the limited aforementioned findings regarding SSF CWs, it is advisable to carry out more experiments under different conditions in order to gain a better understanding of the effect of flow directions on the system treatment performance. However, it was reported that FWS CWs are the type that is usually used for treatment of ADW, owing to their adaptability to flow rate variations and capacity to store and treat larger volumes of water (Land et al., 2016; Lavrnić et al., 2018).

The advantages of the application of simple hydraulic structures (e.g. gated spillways, baffle curtains) within a FWS wetland system were highlighted. Their presence generally leads to an increase of HRT, one of the key parameters that determines the overall performance of CW treatment systems (Pugliese et al., 2020).

Carrer et al. (2011) studied seven-year (2003-2009) variation of a CW system located in Northern Italy before and after the construction of a gated spillway and a storage basin. The results reflected the greater purification efficacy of the system was achieved during the period of 2006-2009, after the construction of these hydraulic structures, rather than during 2003-2005, period before their

construction. Specifically, in comparison with the previous performance, annual discharged nutrient loads of the system equipped with the new hydraulic structures reduced 71%, 57%, 55% and 15% of N-NO<sub>x</sub>, P-PO<sub>4</sub>, TN, TP, respectively. It can be attributed to the presence of the gated spillway and larger water storage, both resulting in a higher HRT in the CW system and promoting removal efficiencies. Similarly, in Tournebize et al. (2017), small dikes (i.e., vegetated embankments) applied in the investigated CW lengthened the flow path and decreased the flow rates, leading to increase of HRT in systems. These findings were also confirmed by Ioannidou and Pearson (2018), who analyzed the performance of 6 full-scale treatment systems with different design parameters in the UK (including 4 FWS CWs and 2 lagoons). It was concluded that the implementation of obstacles (e.g. baffle curtains) can effectively decrease short-circuiting levels and optimize hydraulic performance and treatment efficiency of systems.

In terms of the configuration of inlet and outlet points, Ioannidou and Pearson (2018) indicated that the banded, i.e., closed pipe, outlet was preferred. It can be explained by the fact that the CW equipped with banded outlet discharged effluents through an elevated exit pipe, therefore leading to higher water depth, better pollutant spread and higher HRT. On the other hand, Tournebize et al. (2017) recommended the proper locations should be taken into consideration, e.g., positioning them at the edges of the flow pathway, to limit the occurrence of hydraulic dead zones.

Hydraulic efficiency index ( $\lambda$ ) is an important reference index when determining the application of aforementioned hydraulic design (e.g., configuration of inlet and outlet points, implementation of obstacles) (Su et al., 2009). It is usually considered to be good hydraulic efficiency when  $\lambda$  exceeds 0.75, though the satisfactory range may be within  $0.5 < \lambda \leq 0.75$  (Persson et al., 1999). Hydraulic efficiency not only reflects the flow distribution of influents, but also reflects the amount of mixing or recirculation within CW systems (Persson et al., 1999). Its calculation can result advanced understanding of the processes occurring within FWS CWs and can shed better light on the removal rates.

Accordingly, it is noted that the reasonable setup of CW configurations would be conducive to enhance the overall treatment capacity of systems, e.g. the proper positioning of inlet and outlet points and the implementation of simple hydraulic structures (e.g. gated spillways, baffle curtains) (Carrer et al., 2011; Ioannidou and Pearson, 2018; Tournebize et al., 2017). In term of the effect of the flow direction, it still remains to be investigated.

### 2.3.3. Vegetation management

The treatment processes of CW systems are dominated by the interaction of water with vegetation cover (Ioannidou and Pearson, 2019). The presence of vegetation within a CW system facilitates

multiple treatment processes and enhances HRT by decreasing flow rates (Allred et al., 2014; Lee et al., 2017). Moreover, plant uptake and biomass accumulation could contribute to removal of nutrients and pesticides (Nan et al., 2020; Picard et al., 2005; Vymazal and Březinová, 2015). They enhance oxygen availability of CW systems through root oxygen release while the decaying plant residues could be a source of carbon, satisfying treatment process needs to some extent (e.g., microbial decomposition, nitrification) (Álvarez-Rogel et al., 2020; Barbera et al., 2009; Lavrnić et al., 2020b; Vymazal, 2017). In addition, vegetation can promote water column-sediment interactions (e.g., the soluble transport of nitrogen species due to macrophyte water uptake) (Martin et al., 2003), and provide surface area for biofilm growth and microbial attachment (Barco and Borin, 2020; Brix, 1997; Kumwimba et al., 2017).

Due to its uptake capacity, biomass harvesting can effectively remove pollutants from CWs (Vymazal, 2007). Furthermore, harvested biomass might also be recycled for different use (Mancuso et al., 2021) and at the time create economic benefits (Kumwimba et al., 2018). Regular harvesting could not only avoid the release of pollutants accumulated by plant uptake back into CW treatment systems, but could alter the original plant cover and facilitate the new plant growth (D'Áz et al., 2012; Giannini et al., 2018; Kumwimba et al., 2018; Vymazal, 2007). To some extent, small-scale harvest can be also seen as an economical way to select the species most suitable for water purification, while at the same time reducing ecosystem disturbances compared with the full-scale harvesting (Carty et al., 2008).

Lenhart et al. (2016) carried out a 3-year field study on a 0.1 ha treatment wetland for treating subsurface tile drainage water in Minnesota, USA. They pointed out that planting vegetation on the edge of CWs or selecting native species well adapted to CW soil conditions can assist plant harvesting and further achieve the goal of permanent phosphorus removal. In Lenhart et al. (2016), the establishment of vegetation was slow during the first monitoring year due to the effect of surface water flooding and was completed by second and third year of monitoring. Consequently, the initial plant uptake was negatively affected by the delay of plant development. In contrast, Steidl et al. (2019) attributed the good potential for nitrogen removal to the quick vegetation development in the CW they investigated. This was in agreement with the findings of Nilsson et al. (2020), which pointed out the importance of emergent vegetation existence in young CWs.

Accordingly, the application of vegetation related management techniques (e.g., vegetation establishment and routine harvesting) can be crucial for long-term and effective nutrient removal (Hoffmann et al., 2012; Margalef-Martí et al., 2019; Nilsson et al., 2020; O'Geen et al., 2010). Plants were recommended to be developed before a start-up period for the system, avoiding the possible delay of their establishment process due to uncontrolled water level during operation

period, which would further affect the potential of pollutant removal in systems (Izadmehr and Rockne, 2018; Lenhart et al., 2016; Steidl et al., 2019). Vegetation species, density and harvest regimes, which are closely associated with removal efficiencies, are the important factors to be taken into account (Lenhart et al., 2016; Wu et al., 2013). In addition, it is not advisable to plant seedlings in CW systems, since they generally need longer time to develop and are relatively more sensitive to pollutants and change of water level (Carty et al., 2008).

## 2.4. Long-term performance of full-scale CWs

As Table 2.1 listed, a total of 15 field studies were considered, which investigated full-scale CW systems for treating ADW. Based on the performance of these CWs after long-term operation, this section discusses the design and operational advantages and possible improvements with reference to the CW design and operational considerations proposed in section 2.3. Meanwhile, a summary of the main findings is provided in Table 2.2.

Allred et al. (2014) operated a wetland reservoir subirrigation system (WRSIS) in Ohio, USA for the goal of agricultural water recycling. It was built in 2003 and consisted of a CW and reservoir connected with subsurface pipes, achieving water purification, storage and farmland irrigation via transportation of water from the reservoir to plant roots. In 2009, four field tests on nitrogen removal have been performed under different conditions (i.e., inflow volume, HRT, nitrogen input load), occurring in different time periods from May to November. The most effective was Test 3 with reductions of 44% for  $\text{NO}_3^-$ -N, 87.5% for  $\text{NH}_4^+$ -N and 44.9% for TN. The authors attributed such a result to a longer HRT (5.3 days) of Test 3 if compared to Test 1 (1.8 days) and Test 2 (1.7 days). Nevertheless, Test 4 had a HRT of 11.1 days, the highest one, but the removal of  $\text{NO}_3^-$ -N and TN was still as low as 15.6% and 16.1%, respectively. Unlike the other tests carried out during warm months (May and June), Test 4 occurred between October and November, thus the cold temperatures had a negative influence on activities of denitrifying bacteria. In this study, the application of hydraulic structures (i.e. an adjustable height weir, a peninsula) should be highlighted. The outlet weir regulated water discharge and the peninsula functioned as a baffle, both advantageous to optimization of the treatment efficiency. In terms of wetland-to-catchment ratio, it was reported to be 2%, which is within the range recommended in section 2.3.1.

Groh et al. (2015) reported the CWs investigated in Illinois, USA. They continued to function well even after 18 years of operation, achieving a nitrate removal of 56%, providing a similar treatment capacity as for the initial period of operation. Moreover, it was stressed that the addition of riparian buffer strip allowed the total nitrate removal to rise to 62%, which was attributed to the presence of

seepage losses.

In Denmark, two restored riparian wetlands (Egeskov and Stor Å) have been investigated for two successive discharge seasons, 5 years after their re-establishment, to avoid the possibly unrealistically high rates of nutrient uptake measured in start-up and transitional periods (Hoffmann et al., 2012). The wetland to upland ratios were reported to be 13.7% for Egeskov wetland and 2.4% for Stor Å wetland. In the first and second monitoring year the load removal rates of TN were 43% and 75% at Egeskov and 32% and 26% at Stor Å, respectively. Similarly, nitrate was reduced by 41% and 90% at Egeskov, while it was by 32% and 26% at Stor Å, respectively in year 1 and year 2. Accordingly, the authors ascribed the higher N values obtained by Egeskov wetland to its greater wetland-to-upland ratio. It is also reported that P accumulation in the aboveground biomass reached to 10.3 kg P ha<sup>-1</sup> yr<sup>-1</sup> (Egeskov) and 16.5 kg P ha<sup>-1</sup> yr<sup>-1</sup> (Stor Å), namely 8-11 times more than the annual P load input, implying the good potential for plant uptake of phosphorus. In order to retain the great capacity, therefore, annual harvesting was recommended by the authors.

Another study, taking place in France, reported the fate of herbicide glyphosate and its main degradation product aminomethylphosphonic acid (AMPA) in a stormwater wetland during three consecutive seasons of applying this product (Imfeld et al., 2013). The authors aimed to quantify the variation of the total glyphosate loadings (derived from both glyphosate and AMPA). The data reflected a gradual growth of removal over three monitoring years, i.e., 75% in 2009, 90% in 2010 and 99% in 2011. In this study, it was explained by the fact that (i) increasing plant cover (from less than 1% to about 100%) in the period 2009-2011 led to more sorption of glyphosate and AMPA, (ii) there was a better adaptation of microorganisms over time. Similarly, Koskiaho and Puustinen (2019), mentioned in section 2.3, reported the stable and even better treatment performance of a CW (already serving for 15 years) compared to the capacity during early years of establishment. Similar removals of TSS (68% and 74%) and TP (62% and 58%), more effective removals of NO<sub>3</sub>-N (35% and 69%) and dissolved reactive P (DRP) (27% and 76%) were measured during 1999-2000 and 2007-2014, respectively. The authors attributed it to vegetation growth, increased biological activity and unexhausted soil adsorption capacity.

In Northern Italy, an 18-year-old FWS CW studied by Lavrnić et al. (2020b) showed positive overall treatment capacity. The maximum mass load retention was 82% for TSS and 78% for TN and NO<sub>3</sub>-N, during the monitoring years 2018-2019. Interestingly, it is also found that plant uptake of pollutants TN, TP and total organic carbon (TOC) was in accordance with coverage of the dominant species *Phragmites australis* in the CW. The good capacity of the CW system for pollutant treatment was also confirmed by another study of the same system. Lavrnić et al. (2020a) focused on the hydrological and hydraulic performance. Although there were certain unsatisfactory

phenomena observed after long-term operation, i.e., clogging, dead zones and preferential flow paths, the function of the CW was not remarkably restricted. For example, the FWS CW had a limited number of dead zones due to the presence of four meanders within the system and a long water course indicated a higher value of aspect ratio, the design parameters that are considered important as explained in the section 2.3.

Regarding metal mitigation that is not often reported in literature, Lebrun et al. (2019) studied a 5-year-old French CW and proved its effectiveness on treating ADW with low metallic pollutant levels of influents, which was subjected to temporal fluctuations. The authors sampled both suspended sediments and free inorganic metallic contaminations and weak organic complexes in water. The results revealed that the metals present in trapped sediments were reduced by 11-23% from the inlet to the outlet (i.e. As, Cd, Cr, Co, Cu, Ni, Pb, Sb, Se and Zn). On the other hand, among all the metals measured in water column, five of them (i.e., Cd, Cr, Co, Mn and Ni) indicated significant abatements ranging from 13% to 51%. Notably, the configuration of the wetland studied was divided into several sub-basins by bunds, thereby lengthening HRT. Furthermore, the positive relationship between removal efficiency and HRT has been demonstrated during this three-month observation.

Pugliese et al. (2020) performed tracer experiments on a 6-year-old Danish CW mainly composed by alternated deep and shallow zones. It was found that the shallow zones functioned as barriers when the flow from completely-mixed deep zones reached them at high velocity. The length-to-width ratio of this system was reported to be 7:2, within the range ( $2 < L:W < 10$ ) aforementioned in section 2.3.1 for achieving sufficient hydraulic efficiency.

In Tolomio et al. (2019), the authors confirmed the reliable treatment performance regarding nutrient removal of an Italian FWS CW system, which already had a 10-year long operation lifespan. The yearly TN removal remained relatively stable and was on average 79% during the monitoring period (2007-2013), slightly lower than 90% provided in the previous study Borin and Tocchetto (2007), measured in earlier period (1998-2002) of the same CW system establishment. On the other hand, the mass load removal efficiency substantially varied from 3% to 93% and from 25% to 94%, for  $PO_4\text{-P}$  and TP, respectively. The fluctuation was considered as rational and expected, especially for short-term investigations (Mitsch et al., 2012). In addition, the role of structural modifications should not be ignored. For example, the construction of additional banks in 2007, benefited the hydraulic efficiency of the system and probably caused the increase in the removal efficiencies observed after 2007.

The study by White (2018), already mentioned in section 2.3.1, started 12 years after CW1 construction. The system consisted of parallel deep cells followed by shallow ones. It was observed

that the average nitrogen removal efficiency ranged from 56.9% to 65.3% for different sampling sites within CW1. Not only the sustainable and stable performance, but the economic benefit of the system design should be noted. Specifically, the creation of deep cells probably contributed to saving land resources needed for purification processes and at the same time enhanced the HRT.

Maynard et al. (2014) pointed out that carbon retention potential of CW systems could be negatively affected by algal growth. In their study, actual C removal of the investigated CW (located in California, USA) was weakened due to the production of algal C, especially during years when the system had a lower plant cover. Thus, the authors suggested to enhance CW vegetation development and efficient vegetation management to reduce the productivity of algae, which is consistent with section 2.3.3.

Li et al. (2018) carried out three-year seasonal (i.e., spring, summer and autumn of each year) monitoring of a Chinese wetland restored from cropland. The average removal efficiencies reached 43.84% and 48.44% in this study, consistent with the common range of 40-55% and 40-60% reported by Vymazal (2007), for TN and TP, respectively. It was found that the pollutant concentrations from influent to effluent showed a reduction of 7.54-84.36% for TN and a reduction of up to 70.83% for TP. The lowest values of both pollutants were measured in the summer of 2016, which can probably be explained by the inadequate capacity of the wetland for purifying the excessive input loads that occurred in those months. According to the recommendations mentioned in section 2.3.1, the effect of seasonal variability on water purification (e.g., seasonal rainfall) is thus advised to be taken into consideration when designing a CW. However, in certain cases it is also important to consider seasonal discharge regimes that are dependent on the farming activities in the area.

In addition to the frequently mentioned retention capacity of CWs on nutrient and pesticide pollutants, the potential of some CW systems to be a CO<sub>2</sub> sink was highlighted. For example, Maucieri et al. (2014) reported abundant organic carbon (OC) storage in CW soil during the monitoring period. In particular, OC sequestration of 32.6 Mg ha<sup>-1</sup> for the 0-20 cm soil layer (measured in 2007-2012) and 78.1 Mg ha<sup>-1</sup> for the 20–50 cm layer (measured in 2009-2012) was observed. It was also found that the soil OC concentration in the top 20 cm layer had a marginal increase during the research period 2007-2012, varying from 12.3 g kg<sup>-1</sup> to 13.1 g kg<sup>-1</sup>. However, it increased considerably compared to 7.3 g kg<sup>-1</sup>, the value of OC concentration measured in the construction year of the CW (1996). The findings of C storage capacity were also supported by Maynard et al. (2011a), who studied the spatial and temporal variation on carbon sources of a 13-year-old CW.

### 2.4.1. The relationship between nitrogen removals and related factors

Although not all 15 field studies provided the necessary data, we used those available to assess the relationship between nitrogen removals (TN and  $\text{NO}_3^-$ -N) and main design or operational parameters. Wetland-to-catchment ratio and average water depth were the two factors for which enough data was available and for which the strongest relationship was obtained. In general, the treatment performance of full-scale CW systems reflected connections with these factors to some extent.

Fig. 2.2 presents the relationship between load removals and wetland-to-catchment ratio. Overall similar trend was found for parameters  $\text{NO}_3^-$ -N and TN, which can be explained by the fact that  $\text{NO}_3^-$ -N was usually considered as the primary N form in ADW (Hoffmann et al., 2012; Lavrnić et al., 2020b). However, the goodness of fit ( $R^2$ ) of both parameters was not high. The reasons might be that (i) the data quantity was not sufficient to precisely describe the relationship between load removal and wetland-to-catchment ratio, or (ii) wetland-to-catchment ratio was not the only factor that dominated the removals, as already suggested by Moreno-Mateos et al. (2010). Based on these results, the recommended wetland-to-catchment ratio should be at least 5% to achieve  $\text{NO}_3^-$ -N and TN removals higher than 50%. This finding was not consistent with Vymazal (2017), who suggested 1% was sufficient to reach 40% TN removal and who also reported that the ratio larger than 1% did not significantly increase TN removal. This difference was probably caused by the fact that this study considers only the full-scale systems that functioned over a longer time period.

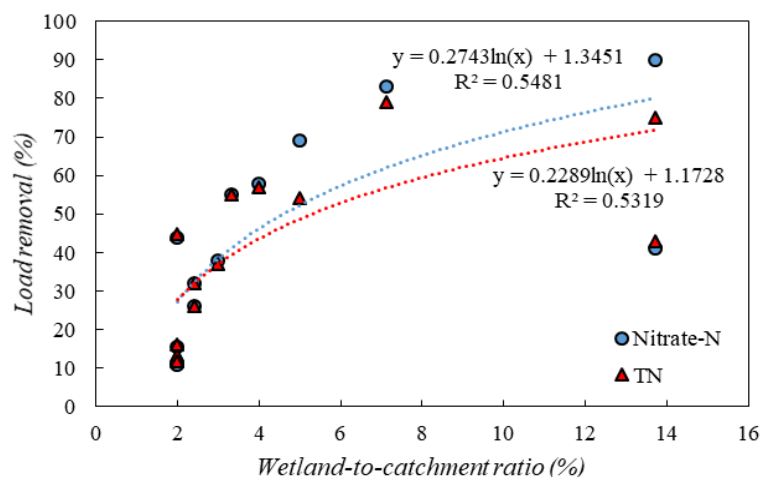


Fig. 2.2 - Relationship between load removals and wetland-to-catchment ratio.

Fig. 2.3 shows that the load removal of  $\text{NO}_3^-$ -N and TN may be positively affected by average water depth. The increased water depth could yield a relatively higher removal, which was in agreement with the findings of White (2018). However, it was noted that the goodness of fit was only 0.48 and 0.26 for TN and  $\text{NO}_3^-$ -N, respectively. It indicated that the relationship remained uncertain, which



was supported by Kadlec (2007), who suggested that there were not enough data to make a clear conclusion on the advantage of FWS CWs with different water depth.

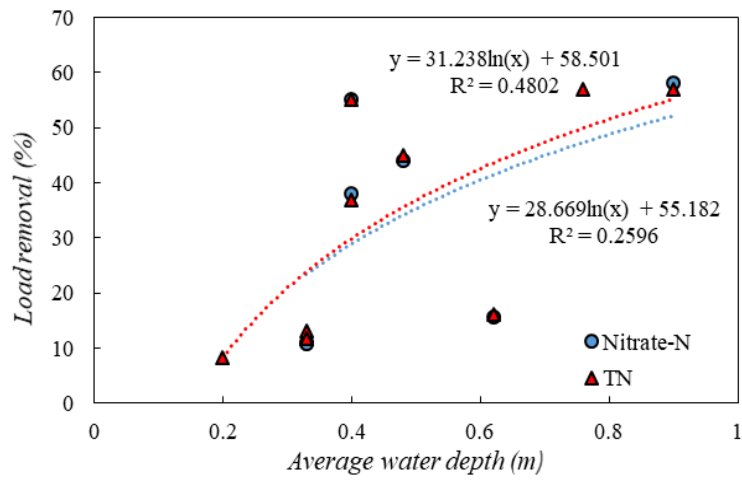


Fig. 2.3 - Relationship between load removals and average water depth.

Table 2.1 - General information on multi-year-old full-scale constructed wetland systems extracted from 15 literature studies.

Location	Construction year	Monitoring period	Surface area	Wetland/Catchment area ratio	Reference
USA	2003	May to November 2009	0.34 ha	2%	Allred et al. (2014)
USA	1994	2012-2013	0.60 ha	4%	Groh et al. (2015)
USA	1994	2012-2013	0.30 ha	3.33%	Groh et al. (2015)
Denmark	2001 <sup>a</sup>	2007-2009	0.62 ha	13.7%	Hoffmann et al. (2012)
Denmark	1990 <sup>a</sup>	1996-1998	0.59 ha	2.4%	Hoffmann et al. (2012)
France	2002	2009-2011	0.03 ha	0.07%	Imfeld et al. (2013)
Finland	1998	2007-2014	0.60 ha	5%	Koskiaho and Puustinen (2019)
Italy	2000	2017, 2018-2019	0.40 ha	3%	Lavrnić et al. (2020a), Lavrnić et al. (2020b)
France	2010	March to May 2015	0.53 ha	0.15%	Lebrun et al. (2019)
China	2005 <sup>b</sup>	2014-2016	-	-	Li et al. (2018)
Italy	1996	2007-2012	0.32 ha	5.82%	Maucieri et al. (2014)
USA	1993	2004-2005	7.30 ha	0.32%	Maynard et al. (2011a)
USA	1993	April to September 2007	4.50 ha	0.20%	Maynard et al. (2014)
Denmark	2010	March to April 2016	0.30 ha	0.79%	Pugliese et al. (2020)
Italy	1996	2007-2013	0.32 ha	7.11%	Tolomio et al. (2019)
USA	1997	2009-2013	3.80 ha	7.82%	White (2018)

<sup>a</sup> It indicates the year when the semi-natural wetland was redesigned for agricultural drainage water treatment.

<sup>b</sup> It indicates the year when the wetland area was restored from farmland.

Table 2.2 - Performance of CW systems after long-term operation and related information on CW design and operation.

Reference	Main results	Advantage of CW design and operation	Possible improvement of CWs
Allred et al. (2014)	The highest removals were 44% for NO <sub>3</sub> <sup>-</sup> -N, 87.5% for NH <sub>4</sub> <sup>+</sup> -N and 44.9% for TN in Test 3 due to a longer HRT (5.3 days)	The application of hydraulic structures (i.e. an adjustable height weir, a peninsula); the wetland-to-catchment ratio is within the recommended range	-
Groh et al. (2015)	Nitrate removal of 56%	the wetland-to-catchment ratios are within the recommended range	-
Hoffmann et al. (2012)	The higher N values obtained by Egeskov wetland due to its greater wetland-to-catchment ratio	Wetland-to-catchment ratios of both Egeskov wetland (13.7%) and Stor Å wetland (2.4%) are within the recommended range	Annual harvesting could increase nutrient removal
Imfeld et al. (2013)	A gradual increase of total glyphosate removal over three years due to increased plant cover and biodegradation	Increasing plant cover led to more sorption of herbicide glyphosate and its main degradation product	The wetland-to-catchment ratio (0.07%) could be enhanced
Koskiaho and Puustinen (2019)	The pollutant removal efficiencies were similar or better than the previous removals during early years of establishment	Increased vegetation improved N and P retention, the wetland-to-catchment ratio is within the recommended range	-
Lavrić et al. (2020a)	A limited number of dead zones due to the presence of four meanders and high length-to-width ratio	The application of hydraulic structure (i.e. the creation of four meanders)	-
Lavrić et al. (2020b)	A positive overall treatment capacity	Nutrients and heavy metals were stored by plants and decaying plant residues provided organic matter and attachment surfaces for bacteria; the wetland-to-catchment ratio is within the recommended range	-
Lebrun et al. (2019)	The efficient removal of metallic pollutants in agricultural drainage water	The application of hydraulic structure (i.e. the wetland was divided into several sub-basins by bunds)	The wetland-to-catchment ratio (0.15%) could be enhanced

Li et al. (2018)	The average removal efficiencies reached 43.84% for TN and 48.44% for TP	-	The effect of seasonal variation (i.e. seasonal rainfall and discharge regimes dependent on the farming activities) should have been considered
Maucieri et al. (2014)	The CW can function as a CO <sub>2</sub> sink	The wetland-to-catchment ratio is within the recommended range	-
Maynard et al. (2011a)	The CW can function as a sink for eroded C	-	The wetland-to-catchment ratio (0.32%) could be enhanced
Maynard et al. (2014)	Actual C removal of the CW was negatively affected by algal growth	-	Efficient vegetation management should be enhanced in order to reduce the productivity of algae; the wetland-to-catchment ratio (0.2%) could be enhanced
Pugliese et al. (2020)	The shallow zones functioned as barriers when the flow from completely-mixed deep zones reached them at high velocity.	The length-to-width ratio of CW is within the recommended range 2-10; the creation of alternated deep and shallow cells	The wetland-to-catchment ratio (0.79%) could be enhanced
Tolomio et al. (2019)	The annual TN removal was relatively stable and was on average 79%	The application of hydraulic structure (i.e. banks); the wetland-to-catchment ratio is within the recommended range	-
White (2018)	The average N removal efficiency was in the range of 56.9-65.3% for different sampling sites within CW1	The creation of deep cells saved land resources needed for treatment processes and increased the HRT; the wetland-to-catchment ratio is within the recommended range	-

## 2.5. Conclusions

The goal of this study was to explore design and operational recommendations for CWs treating ADW, and to link them to the removal efficiencies of already established CW systems. Through the literature review, and especially overview of 16 CWs with long lifespan (from 15 literature), it can be concluded that a longer HRT, being one of the most important parameters affecting the treatment, can promote pollutant removal efficiencies of CW systems. In addition, a large wetland-to-catchment ratio can result in a high HRT, but it can also be lengthen by (1) the application of simple hydraulic structures within CW systems (e.g. baffle curtains, small dikes), (2) the alternated distribution of deep and shallow zones, (3) the application of banded outlets and (4) the presence of vegetation. These changes were shown to be beneficial for treatment efficiencies in different systems. Moreover, in order to reduce the occurrence of hydraulic dead zones and promote hydraulic efficiency of CWs, it is advised to (1) position inlet and outlet points at the edges of flow pathway, (2) adopt a high aspect ratio and (3) create a long and narrow CW shape. Vegetation establishment and regular harvesting can help CW systems to achieve greater pollutant removal potential. Besides, the effect of local climate condition and seasonal variation should be taken into consideration prior to CW design.

As shown by the authors that studied the removal efficiencies of established and mature CWs, these systems can be considered as a practical solution for ADW treatment even a long time after their establishment. Nevertheless, certain modifications and design and operational recommendations reported in this research can prolong their lifespan and ensure efficient pollutant removal.

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# CHAPTER 3 - Performance of a full scale constructed wetland as ecological practice for agricultural drainage water treatment in Northern Italy

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## Abstract

Non-point sources of pollution, primarily agricultural drainage waters, can cause eutrophication and deterioration of water bodies. Surface flow constructed wetlands (SFCWs) are an ecological solution that can represent an efficient barrier and prevent agricultural pollutants from reaching other ecosystems. However, to better manage them and to understand removal processes occurring, it is important to study SFCWs that are functioning for longer periods of time and assess their efficiencies. This study concentrates on a full-scale SFCW in the Northern Italy that has been treating agricultural drainage water for past 20 years. An in-deep monitoring done for two years (2018 and 2019) showed that the system achieved satisfactory retention of up to 82% for TSS and up to 78% for TN and  $\text{NO}_3^-$ -N. TP retention seemed to be poor, but further analysis showed that the SFCW performed well in this aspect as well, and that it is important to include precipitation loads in the overall balance. Soil content of nutrients and different trace elements did not show considerable differences in respect to the beginning of the monitoring period, and the uptake rates of TN and TP by above-ground vegetation were in the range 19.0-26.3 and 1.6-2.1  $\text{g m}^{-2}$ , respectively.

### 3.1. Introduction

Constructed wetlands (CWs) are generally regarded as an economical, easy operation and effective alternative (Lavrnić and Mancini, 2016; Liu et al. 2015) for treating wastewater of different sources, including domestic and industrial wastewater (Calheiros et al. 2009; Arden and Ma, 2018; Lavrnić et al. 2019; Russo et al. 2019a) and agricultural drainage water (Lavrnić et al. 2018). Reed and cattails are the most common vegetation planted in these ecosystems (Rousseau et al. 2008). The application of CWs can not only reduce pressure on conventional treatment plants (Ghaitidak and Yadav, 2013), but it can also provide habitat for wildlife, aesthetic and recreational values for public (ElZein et al. 2016; Rousseau et al. 2008). Furthermore, treated effluents can be reused for various purposes, such as agricultural irrigation, domestic purposes and gardening (Toscano et al. 2013; ElZein et al. 2016; Dou et al. 2017; Russo et al. 2019b).

CWs are classified into surface flow constructed wetlands (SFCWs) and subsurface flow ones, based on their hydraulic functioning. The latter one can be further divided into horizontal and vertical flow CWs. In comparison with subsurface systems, SFCWs are the type of CW that is most commonly used for agricultural drainage water treatment since they provide favorable environment for it (Tanner and Kadlec, 2013; Tournebize et al. 2017; Dal Ferro et al. 2018). SFCW systems are designed using parameters like water depth, size, substrate, plant, etc., and implemented under various requirements (e.g. flow rate, feeding mode) (Headley et al. 2013; Morató et al. 2014; Herrera-Melián et al. 2018; Song et al. 2019). However, factors like seasonal and annual variation, the aging of CW systems or other internal/external conditions have a possibility to negatively affect the treatment performance of CWs, particularly over the longer time periods. Besides, the research on long-term experiments taking into account local conditions is limited in quantity (Dal Ferro et al. 2018).

Agricultural drainage systems shorten the retention time of water in soil, leading to nutrient losses from farmland (Steidl et al. 2019). Agricultural watersheds and their network of ditches and canals may have a high capacity for TN removal due to their heterogeneous hydraulic, ecological and biological parameters (Castaldelli et al. 2018). Nevertheless, the agricultural drainage water can be a source of diffuse pollution in aquatic ecosystems due to high concentrations of nitrate, certain salts, phosphorus, organic nitrogen, pesticides and sediments (Woltemade, 2000; Haverstock et al. 2017), as confirmed by the authors from different parts of the world (Tanner and Kadlec, 2013; Darwiche-Criado et al. 2017; Mendes et al. 2018a; Song et al. 2019). Particularly, P enrichment in

runoff leads to eutrophication harmful to plants growth (Johannesson et al. 2017; Lavrnić et al. 2018), and it can even result in toxic algae blooms and loss of biodiversity (Reinhardt et al. 2005). Therefore, different international agreements (e.g. European Union Water Framework Directive) were achieved in order to reduce nutrient load from agricultural land and consequently improve water quality in the environment (Ulén et al. 2019).

It was reported that SFCWs can be an inexpensive and efficient nature-based solution for the reduction of non-point source pollution, especially for nitrogen and phosphorus (Tolomio et al. 2019, Pugliese et al. 2020), and they were used for that purpose in different countries (Song et al. 2019). Nitrogen removal/retention in SFCWs mainly depends on the biological and physico-chemical mechanisms (e.g. nitrification/denitrification, plant uptake, biomass assimilation and volatilization) (Billy et al. 2013; Song et al. 2019), while the mechanisms for phosphorus removal include soil accretion, adsorption, microbial/plant uptake and precipitation (Vymazal, 2007). On the other hand, scientific research on the effect that CWs can have on agricultural pollution abatement is still limited, especially the one considering seasonal and long-term hydro-meteorological variations (Ulén et al. 2019). Moreover, since the expansion of SFCWs for agricultural drainage water treatment started a few decades ago (Song et al. 2019), it is important to understand their behavior and performance once they reach the mature stage.

With these considerations in mind, the present paper studies a full-scale SFCW located in Northern Italy, which was built and is operating since 2000. In the beginning of 2017 the above-ground vegetation was harvested and in-depth monitoring of the system operation started in 2018. After having assessed its historical performance (Lavrnić et al. 2018), the capacity for pesticide removal (Braschi et al. manuscript in preparation) and having evaluated its hydrological and hydraulic behavior (Lavrnić et al. 2020), the main objective of this study was to assess the overall performance for agricultural drainage water treatment of this particular SFCW after two decades of operation.

## 3.2. Materials and methods

### 3.2.1. Experimental set-up and condition of the constructed wetland

The research was conducted at an experimental agricultural farm of Canale Emiliano Romagnolo land reclamation consortium (CER) in the Emilia-Romagna region (Italy), from January 2018 to December 2019. The site has a sub-humid climate, with the mean annual temperature of 13.7 °C and the average annual rainfall of 771 mm (Lavrnić et al. 2018). During the monitoring period discussed



in this study, the annual climatic conditions were similar to the average, as in 2018 the mean yearly temperature and yearly rainfall were 14.4 °C and 752 mm, respectively, while in 2019 they were 14.5 °C and 751 mm, respectively.

The SFCW (Fig. 3.1), constructed in 2000 and operating since, is a part of the experimental farm, occupying about 3% of the total surface area (12.5 ha) that is inside the recommended range of 0.5-5% (Tanner and Kadlec, 2013). It has a surface of approximately 0.4 ha and it is an off-stream measure meaning that it is located outside of the main water stream. The farm drainage water flows to the main ditch from where it is abstracted with two pumps and conveyed to the SFCW. The pumps activation depends on the water level inside the ditch. In case it surpasses a certain level, an overflow activates and excess water bypasses the wetland.

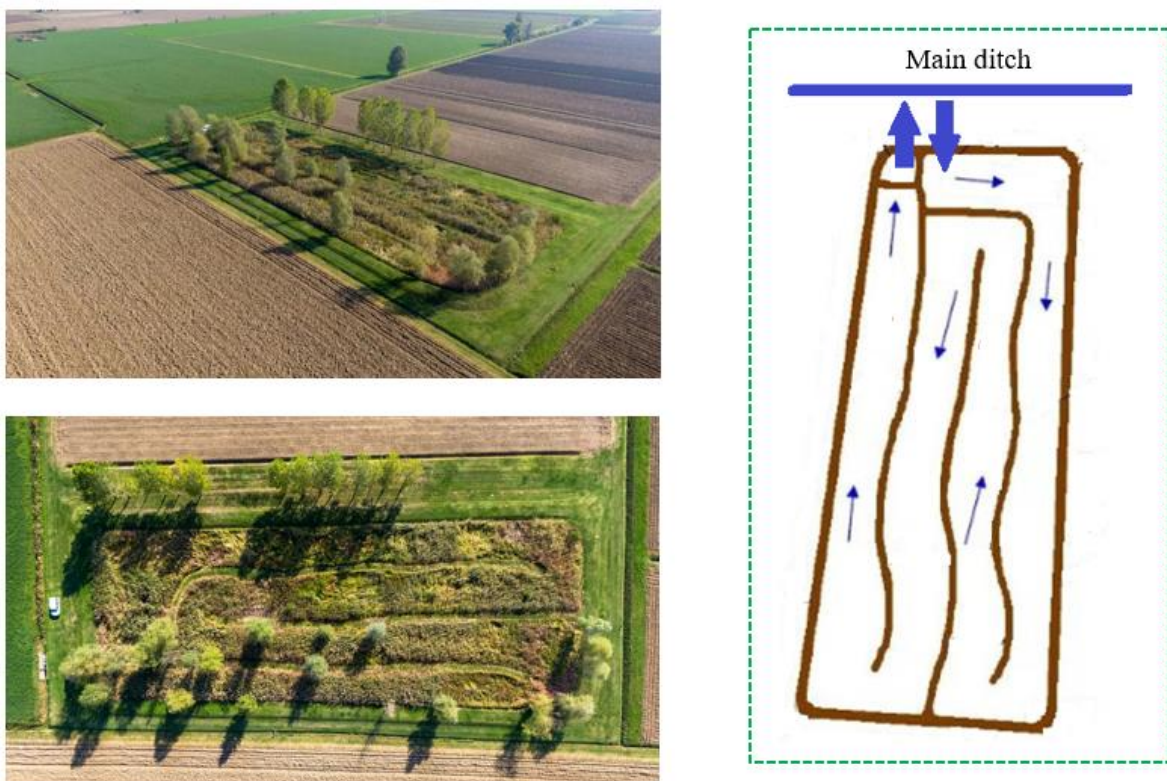


Fig. 3.1 - Areal views and scheme of the monitored SFCW and the surroundings.

The surface of the SFCW was partitioned with a few barriers, effectively dividing it into four 8-10 m wide meanders and creating a 470 m long water course. The total volume of the system is close to 1,500 m<sup>3</sup> and the outlet is set on 0.4 m above the bed surface level. The most dominant plant species in the SFCW were *Phragmites australis*, *Typha latifolia* and *Carex* spp. Some additional information and the complete description of the experimental system can be found in Lavrnić et al. (2018; 2020).

The system is equipped with two mechanical flow meters that record influent and effluent volumes

every hour, and two automatic samplers that take influent and effluent water samples on the basis of the inlet water volume and time, respectively. Since the functioning of the system depends mostly on the presence of precipitation, no general sampling schedule could be established and followed. Hence, due to the lack of drainage water, longer or shorter periods of time passed without sampling. The water level inside the CW is measured by a specific sensor. All the collected data were managed and recorded by a central control system. The precipitation height data were taken from the farm weather station, equipped with a precipitation sampling unit.

### 3.2.2. Water balance

The hydrological year was considered to begin on 1<sup>st</sup> January and finish on 31<sup>st</sup> December. For the analysis purposes, a year was further divided into four seasons: winter (January-March), spring (April-June), summer (July-September) and autumn (October-December). The SFCW dynamic water budget (Kadlec and Wallace, 2009) can be expressed as:

$$Q_{in} + (P \times A) - Q_{out} - I - (ET \times A) = \frac{dV}{dt} \quad (1)$$

Where:

$Q_{in}$  = inflow rate ( $\text{m}^3 \text{d}^{-1}$ );

$P$  = precipitation rate ( $\text{m d}^{-1}$ );

$A$  = wetland top surface area ( $\text{m}^2$ );

$Q_{out}$  = outflow rate ( $\text{m}^3 \text{d}^{-1}$ );

$I$  = infiltration flow rate ( $\text{m}^3 \text{d}^{-1}$ );

$ET$  = evapotranspiration rate ( $\text{m d}^{-1}$ );

$V$  = water storage inside the SFCW ( $\text{m}^3$ );

$t$  = time (d)

Over long averaging periods ( $\Delta t$ ), the change in storage ( $\Delta V$ ) can be considered negligible (Kadlec and Wallace, 2009). Even though Lavrnić et al. (2020) had measured punctual infiltration, an overall estimate could not be done. Moreover, it was not possible to measure infiltration and evapotranspiration rates separately. Therefore, a simplified water balance over each hydrological year was calculated as:

$$Q_{in} + (P \times A) - Q_{out} = I + (ET \times A) \quad (2)$$

In Equation (2), the term  $I + (ET \times A)$  was considered as the overall water loss from the SFCW, and it represents the water retained by the SFCW, i.e. not released directly to surface water bodies.

Percentage of time that the SFCW was submersed was also estimated, counting the days when the average water level in the system was at least 2 cm. That limit was taken to prevent that measurement errors or accumulation of sediments affect the assessment.

Furthermore, in order to analyze different inflow episodes that occurred during the monitoring period, the data set was divided into periods when influent into the system was continuous. In order to be considered as an inflow event, an episode had to have a permanent inflow for at least 5 consecutive days and a total volume of at least 200 m<sup>3</sup>. Moreover, to differentiate single events and not confuse them with different parts of the same one, at least 7 consecutive days without inflow before and after the event were taken as a condition. For every inflow event, nominal hydraulic retention time (HRT<sub>N</sub>) was calculated like in Lavrnić et al. (2020):

$$\text{HRT}_N = 0.9 * \frac{V}{\frac{Q_{in} + Q_{out}}{2}} \quad (3)$$

### 3.2.3. Water quality

Water samples (i.e., precipitation and CW influent and effluent) were analyzed for chemical oxygen demand (COD), total suspended solids (TSS), total organic carbon (TOC), total nitrogen (TN), nitrate-nitrogen (NO<sub>3</sub><sup>-</sup>), ammonium-nitrogen (NH<sub>4</sub><sup>+</sup>-N), nitrite-nitrogen (NO<sub>2</sub><sup>-</sup>-N) and total phosphorus (TP).

COD was analyzed spectrophotometrically with a COD Digestion Vials kit (Hach Lange) and TSS by the gravimetric method. TN and TOC were measured by the elemental analyzer Shimadzu TNM-1 (Shimadzu, Kyoto, Japan). Before analysis, all samples were filtered through Watman 42 filters (Merck KGaA, Darmstadt, Germany). Moreover, NO<sub>2</sub><sup>-</sup>, NO<sub>3</sub><sup>-</sup> and NH<sub>4</sub><sup>+</sup> concentrations were determined by using a flow analyzer (AA3, Bran Luebbe, Norderstedt, Germany). TP analysis was performed by using an inductively coupled plasma optical emission spectrometer ICP-OES which was equipped with a plasma source and an optical detector with a charge-coupled device CCD (SPECTRO Analytical Instruments GmbH & Co., Kleve, Germany). Before analysis, in filtered water samples was added 1% of HNO<sub>3</sub> (> 69% v/v, for trace analysis, Fluka, Sigma-Aldrich, St. Louis, MO, USA).

The concentration of different parameters in influent ( $C_{in}$ , mg L<sup>-1</sup>) and effluent ( $C_{out}$ , mg L<sup>-1</sup>) were

multiplied by the corresponding water volume that flowed into ( $V_{in}$ , m<sup>3</sup>) or out ( $V_{out}$ , m<sup>3</sup>) of the system, respectively. Afterwards, all the inflow and outflow loads during one hydrological year/inflow event were summed to calculate the mass of nutrients (kg year<sup>-1</sup>) entering and exiting the wetland. In general, mass load retention rate ( $RR$ , %) during each period of time, was calculated as:

$$RR = \frac{\sum V_{in} \times C_{in} - \sum V_{out} \times C_{out}}{\sum V_{in} \times C_{in}} \times 100 \quad (4)$$

### 3.2.4. Soil

At the end of the monitoring period, in November 2019, the soil was sampled at four points along the water course (at the middle of each meander) in order to allow comparison with results from 2017 (Lavrnić et al. 2018), that were taken as a background condition for the present study. At each position, 60 cm soil core samples were taken. First 5 cm were removed from the sample since they were mostly made of litter and mud, and therefore they were not representative of the real composition of the SFCW soil layer. After manual removal of plant roots up to a diameter of ca. 1–2 mm, the samples were air-dried and sieved to 2 mm. Afterwards, they were tested for TP and trace elements content, using the methods given in section 3.2.4, as well as for TOC and TN.

TOC and TN in the soil samples were determined by using a thermo-electron CHNS-O elemental analyzer (Thermo Fisher Scientific, Waltham, MA, USA). TP and metal concentrations were analyzed by using ICP-OES, after dissolution of soil samples in a mixture of HCl (37% v/v for trace analysis, Sigma-Aldrich, St. Louis, MO, USA), HNO<sub>3</sub> (>69% v/v, for trace analysis, Fluka, Sigma-Aldrich, St. Louis, MO, USA), and H<sub>2</sub>O<sub>2</sub> (30% v/v, for trace analysis, VWR Prolabo Chemicals, Radnor, PA, USA) in the ratio of 4:1:0.25 (v:v:v) by microwave-assisted digestion (Start D, Microwave Digestion System, Milestone, MD, USA).

### 3.2.5. Vegetation

The plants were sampled in a representative 1 m<sup>2</sup> area at the middle of each of the four meanders, once a year (end of October or beginning of November). They were tested and analyzed for nutrient and trace elements content.

Biomass dry weight and average height were measured. The above-ground biomass was harvested as close to the soil as possible while the below-ground biomass was dug up using the mechanic tools and rinsed with water to remove the soil, then dried and ground. All biomass samples were

tested for TOC, TN and TP content, as well as for the presence of the semi-metal B and different heavy metals (Cd, Cr, Cu, Fe, Pb, Mn, Ni and Zn).

TN and TOC analysis of vegetation was performed by using a thermo-electron CHNS-O elemental analyzer (Thermo Fisher Scientific, Waltham, MA, USA). TP and trace elements were measured using ICP-OES. Before elemental analysis, biomass samples were dissolved in a mixture of HNO<sub>3</sub> (>69% v/v, for trace analysis, Fluka, Sigma-Aldrich, St. Louis, MO, USA) and H<sub>2</sub>O<sub>2</sub> (30% v/v, for trace analysis, VWR Prolabo Chemicals, Radnor, PA, USA) in the ratio of 4:1 (v:v) by microwave-assisted digestion (Start D, Micro-wave Digestion System, Milestone srl, Bergamo, Italy).

### 3.3. Results and discussion

#### 3.3.1. Simplified overall water balance

The irregular hydrological nature of the system can be best seen through inflow (considering the direct precipitation onto the system) and outflow volume comparison (Table 3.1). It is important to note that the inflow is originally rainwater, since the farm uses a modern irrigation system that applies water in quantities that are necessary to maintain the soil humidity at a certain level. The major inflow, and consequently outflow, in the system occurred during the two 2018 winter months (February and March), with values that were several times higher comparing to the rest of the period considered. The inflow to the SFCW, together with outflow, varied considerably throughout the monitoring period since it depended on the presence of precipitation, but it was also strongly connected to season, temperature and crop water needs.

For example, in 2018, the overall inflow (including the direct precipitation input) and outflow were 22,389 m<sup>3</sup> and 12,294 m<sup>3</sup>, respectively, while in 2019 those values were much lower - 9,983 m<sup>3</sup> and 1,944 m<sup>3</sup>, respectively (Table 3.1). Like other types of nature-based solutions, SFCWs used for agricultural drainage water treatment are being used at different scales, for different catchments and in different climates, and that makes comparison among them rather difficult. In this study, the average water inflow (61 m<sup>3</sup> day<sup>-1</sup> in 2018 and 27 m<sup>3</sup> day<sup>-1</sup> in 2019) was much lower than the one given by Dal Ferro et al. (2018) for a SFCW in the Veneto region (Italy) that approximately amounted to 5,480 m<sup>3</sup> day<sup>-1</sup>, but was also much higher than 17 m<sup>3</sup> day<sup>-1</sup> for a Canadian SFCW studied by Haverstock et al. (2017).

Table 3.1 - Seasonal hydrology of the monitored SFCW.

Season	Inflow (m <sup>3</sup> )	Outflow (m <sup>3</sup> )	Submersion (% of time)	Water retention/loss (%)	Mean air temperature ( °C)	Seasonal precipitation (mm)
Winter 2018	17,102	12,294	100	28	5.1	261
Spring 2018	2,584	0	95	100	18.7	190
Summer 2018	1,904	0	32	100	24.0	155
Autumn 2018	799	0	0	100	10.0	147
Winter 2019	1,036	0	44	100	5.7	69
Spring 2019	1,646	0	31	100	17.2	229
Summer 2019	614	0	0	100	23.5	155
Autumn 2019	6,688	1,944	49	71	11.2	199
<b>Overall period</b>	<b>26,847</b>	<b>14,265</b>	<b>44</b>	<b>56</b>	<b>-</b>	<b>1,504</b>

In 2018 and 2019, water retention/loss, the difference between inflow and outflow, were 45% and 81%, respectively. Considering the whole monitoring period, the loss was higher than 55% (Table 3.1), and, apart from evapotranspiration and accumulation in the system itself, the important part of it was infiltration into the ground, as concluded by a previous study done on the same SFCW (Lavrnić et al. 2020). Actually, the authors presume that the biggest part of the water loss was exactly due to infiltration processes. For example, during the winter period 2018, when the temperatures were quite low and the vegetation was in senescence (with minimal evapotranspiration), water retention/loss was 28% (Table 3.1), indicating that most of it infiltrated to the ground. Similar conclusion could be taken also when analyzing winter 2019. In addition,

similar results were given by Kovacic et al. (2006), who stated that evapotranspiration represented 8-29% of the total water loss for two SFCW treating agricultural drainage water in USA, while the rest was infiltration.

If the SFCW seasonal hydrology is analyzed, it can be noted that the biggest part (53%) of the two-year inflow into the system occurred during one single season (winter 2018). The inflow during the 18 months period between spring 2018 and summer 2019 (8,582 m<sup>3</sup>) was comparable to the autumn 2019 inflow (6,688 m<sup>3</sup>) (Table 3.1), even more highlighting the hydrological unpredictability of the SFCW discussed. The system was able to accumulate to a large extent the small inflows that occurred between spring 2018 and summer 2019 owing to its total volume of about 1,500 m<sup>3</sup>. Aided by other types of water loss, it limited or completely eliminated outflow. Similar conditions and occasional dry out of a SFCW was also reported by Ulén et al. (2019). However, that study, done in Sweden, did not report 100% water losses for either of the 8 seasons considered, probably because the SFCW represented only 0.3% of the contributing catchment (compared to 3% in this study) and therefore it had a rather high inflow of water.

The smallest water retention/loss occurred during winter 2018, mostly due to the high water inflow and consequently high water level that reduced HRT. In SFCWs that are receiving fluctuating inflow, it is important to consider the percentage of time during which the system is submersed, or, in other words, when the bottom is covered with water. The submersion period was a difficult parameter to estimate since it would often happen that water is present only near the inlet while the second part of the system is dry. For example, dry periods were certainly autumn 2018 and summer 2019, when the inflow was small and dispersed throughout the season. In those conditions, inflow was never large enough to reach the outflow part and, therefore, to submerge the whole system. On the other hand, in winter 2018, as already mentioned, the SFCW received a high inflow and the system was submersed all the time (Table 3.1).

### 3.3.2. Hydrological analysis of single inflow events

Table 3.2 gives characteristics of single inflow events that occurred during the monitoring period. Comparable intensive rain episodes occurred in May-July 2018 (Event 2, Table 3.2) and in November-December 2019 (Event 7, Table 3.2), but they did not cause a similar response in water flow. The reason can be found in the atmospheric conditions during these two events.

In particular, the Event 7 occurred in winter time, when the vegetation was in senescence and the average daily temperature was 7.2 °C, that for a few days was even below 1 °C. Those conditions minimized evapotranspiration from the agricultural fields, caused a faster runoff from the farm area and increased water inflow to the SFCW. On the contrary, Event 2 occurred during the growth

phase of the farm crops and when the average temperature was 23.1 °C. Therefore, it can be assumed that crop water needs and evapotranspiration caused a much smaller water flow.

The Events 3 and 5 were events with a much smaller intensity and consequently they did not produce high inflow. However, the Event 6, although it was a result of three times bigger rain episode, produced a similar inflow as the Event 4. The answer can be again found in the overall conditions. After August, most of the crops grown at the CER farm area are harvested and the bare soil certainly facilitates runoff. On the other hand, spring period is when crops are growing and require constant humidity and therefore, similarly to the Event 2, they could have retained most of the precipitation that occurred in April and May 2019.

Table 3.2 - Single inflow events during the two-year long monitoring period.

Event	Period	Duration (d)	Inflow (m <sup>3</sup> )	Outflow (m <sup>3</sup> )	Initial water level (cm)	HRT (d)	Precipitation (mm)	Temperature (°C)
1	Feb-Apr 2018	82	17,373	12,296	7.0	6.6	264	7.8
2	May-Jul 2018	59	2,425	0	0.6	-	204	23.1
3	Aug 2018	8	283	0	1.6	-	11	25.6
4	Sep-Oct 2018	36	1,227	0	1.5	-	55	19.5
5	Feb 2019	7	930	0	0.7	-	44	3.3
6	Apr-May 2019	34	1,179	0	0.2	-	158	14.6
7	Nov-Dec 2019	42	6,100	1,943	0.1	11.6	190	7.2

The elevated flows that occurred between February and April 2018 (Event 1, Table 3.2) are, as already said, a result of precipitation that was really high during that period (264 mm). An inflow event such as the Event 1 has never been recorded since 2000, the year when the SFCW started functioning. It can be seen that inflow into the system and its frequency depended on the presence and intensity of rain. The inflow affected water level inside the SFCW, that in turn, regulated the outflow (Fig. 3.2). The most intensive rainfall occurred during the first 3 weeks (until the 23<sup>rd</sup> of



February) and again in the period 5<sup>th</sup>-11<sup>th</sup> of March, and it certainly reflected on the influent pattern. The most important outflow happened until the 25<sup>th</sup> of March, and after that date it slowed down due to the absence of intensive rainfall. Although the inflow activity continued beyond that date, its average value was 15 m<sup>3</sup> d<sup>-1</sup> and therefore it did not produce substantial response in terms of outflow or water level increase.

Only two out of the identified seven single events had a considerable outflow and therefore it was possible to calculate their HRT. Nevertheless, it is an important parameter that sheds more light onto the processes occurring inside the system and leading to the retention of pollutants (Song et al. 2019; Pugliese et al. 2020). For example, it has been suggested that the minimum HRT needed for TN reduction on a catchment scale is 2 days (Song et al. 2019). All the events considered in this study had a much higher HRT than that limit, most probably contributing to the substantial TN retention that will be discussed in the next section. In order for HRT to be lower than 2 days, the average daily flow of the SFCW studied would have to be higher than 680 m<sup>3</sup> d<sup>-1</sup> for at least 2 days. However, none of the inflow events that occurred in 2018 and 2019 reached that limit, and the maximum daily flow recorded was 618 m<sup>3</sup> d<sup>-1</sup>.

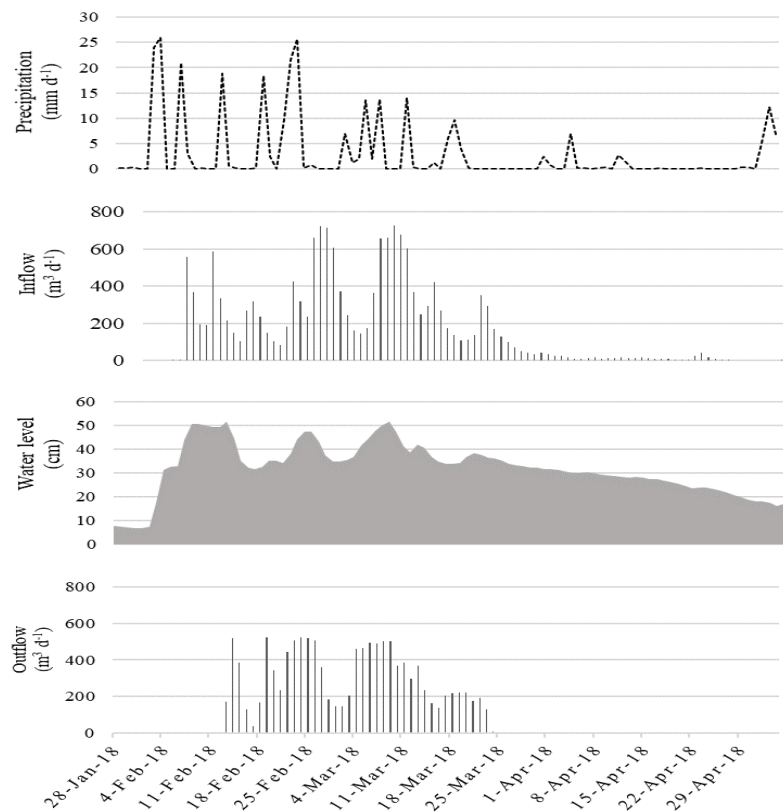


Fig. 3.2 - Hydrological conditions during the Event 1 (Feb-Apr 2018).

### 3.3.3. Water quality

The mean concentrations and mass loads of different parameters in influent and effluent water of the SFCW monitored can be found in Table 3.3. The similar concentrations in influent and effluent are a consequence of the relatively high water retention/loss (Table 3.1). In general, the system achieved satisfactory overall retention of pollutants. Inflow events with no outflow have certainly had an important contribution since the whole pollution load remained inside the SFCW. A part of it was infiltrated and reached surface water bodies via ground water. However, during the percolation process the pollution load has undoubtedly undergone additional treatment (Bali et al. 2010), and thus the SFCW provided an additional positive effect.

TSS was the parameter with the highest incoming load to the SFCW. Yearly TSS load in the SFCW was 2,622 and 949 kg in 2018 and 2019, respectively. If divided by the surface area of the contributing catchment, the load was in the range 75-210 kg ha<sup>-2</sup>. These values were lower than the ones reported by Ulén et al. (2019) for a SFCW in Sweden, probably due to the different type of catchment. However, in this study, TSS retention was satisfactory: 65 and 82% in 2018 and 2019, respectively.

Agricultural drainage waters usually do not contain elevated concentrations of organics (Vymazal and Dvořáková Březinová, 2018a) but they can still contribute to eutrophication and dissolved oxygen depletion (He et al. 2011). Average COD influent concentration was 24.3 mg L<sup>-1</sup>, with minimum and maximum value of 0.0 mg L<sup>-1</sup> and 113.0 mg L<sup>-1</sup>. The effluent one was slightly lower (23.7 mg L<sup>-1</sup>, Table 3.3). However, the system managed to remove 347 kg of COD over two years of monitoring, or 779 kg COD ha<sup>-1</sup> year<sup>-1</sup> in 2018 and 230 kg COD ha<sup>-1</sup> year<sup>-1</sup> in 2019. This retention was comparable to 287 kg COD ha<sup>-1</sup> year<sup>-1</sup> reported by Maniquiz et al. (2012) for a SFCW in South Korea.

The average concentration of TOC increased from 8.0 mg L<sup>-1</sup> in influent to 10.4 mg L<sup>-1</sup> in effluent. On the other hand, the mass load retention of this element was 21% and 62% in 2018 and 2019, respectively. That was better performance than 2-17% range that was found by Kovacic et al. (2006) for two SFCWs treating agricultural drainage water in USA.

The average TN influent concentration was 12.6 mg L<sup>-1</sup>, with the maximum and minimum value of 37.9 mg L<sup>-1</sup> and 1.4 mg L<sup>-1</sup>. NO<sub>3</sub><sup>-</sup>-N is usually the biggest component of TN in agricultural drainage water (Borin and Tocchetto, 2007; Song et al. 2019), and it was also the case in this study where its load represented more than 75% of TN entering the SFCW (Table 3.3). The average influent concentration of NO<sub>3</sub><sup>-</sup>-N was 9.1 mg L<sup>-1</sup>, but, similar to other parameters, it fluctuated a lot in the range 0.3-24.1 mg L<sup>-1</sup>. Similar finding was reported by Tanner and Kadlec (2013) who stated that

nitrate concentrations often vary between events, seasons and locations.

Table 3.3 – Average concentrations (mean±std. error (sample size)) and mass loads of the water quality parameters during the monitoring period (2018-2019).

		Influent		Effluent		Retention
		Concentration (mg L <sup>-1</sup> )	Mass load (kg)	Concentration (mg L <sup>-1</sup> )	Mass load (kg)	Mass load (%)
COD	Overall	24.3±28.6 (20)	622	23.7±9.3 (11)	304	51
	2018	26.8±34.2 (14)	495	18.5±9.3 (6)	227	54
	2019	18.7±3.4 (6)	139	29.9±4.5 (5)	60	57
TOC	Overall	8.0±3.0 (49)	197	10.4±2.7 (28)	135	33
	2018	8.0±2.1 (26)	148	10.1±1.8 (20)	116	21
	2019	7.9±3.9 (23)	48	11.0±4.2 (8)	19	62
TSS	Overall	162.4±172.3 (76)	3,579	78.0±35.2 (30)	1,101	69
	2018	186.4±218.9 (42)	2,622	76.9±33.5 (22)	928	65
	2019	132.7±79.2 (34)	949	80.7±42.0 (8)	173	82
TN	Overall	12.6±7.9 (76)	428	12.3±4.9 (30)	204	52
	2018	11.4±6.4 (42)	289	11.8±4.4 (22)	174	40
	2019	14.1±9.3 (34)	138	13.8±6.3 (8)	30	78
NH <sub>4</sub> <sup>+</sup> -N	Overall	0.43±0.57 (60)	5.3	0.10±0.30 (22)	1.8	66
	2018	0.44±0.61 (42)	2.5	0.10±0.31 (19)	1.6	35
	2019	0.40±0.46 (18)	2.6	0.09±0.15 (3)	0.2	91
NO <sub>3</sub> <sup>-</sup> -N	Overall	9.1±6.5 (76)	326	9.1±4.2 (30)	155	52
	2018	8.4±5.5 (42)	221	8.7±3.7 (22)	132	40
	2019	10.0±7.8 (34)	105	10.4±5.4 (8)	23	78
NO <sub>2</sub> <sup>-</sup> -N	Overall	0.05±0.08 (40)	1	0.13±0.12 (22)	1.8	-
	2018	0.04±0.08 (35)	0.5	0.13±0.12 (19)	1.6	-
	2019	0.06±0.09 (5)	0.5	0.11±0.14 (3)	0.2	29
TP	Overall	0.05±0.14 (76)	0.5	0.02±0.09 (30)	0.6	-
	2018	0.05±0.12 (42)	0.4	0.03±0.10 (22)	0.6	-
	2019	0.06±0.15 (34)	0.1	0.01±0.01 (8)	0.0	100

NO<sub>3</sub><sup>-</sup>-N influent concentration was higher, but in line with the one reported by Haverstock et al. (2017) (6.7 mg L<sup>-1</sup>), who also reported a much smaller effluent concentration (2.2 mg L<sup>-1</sup> vs 9.1 mg L<sup>-1</sup> in this study). A likely reason for such difference is that Haverstock et al. studied a waterproofed SFCW, that did not lose water to seepage, and to the fact that it was located in Canada, in a colder climatic zone where evapotranspiration was not that high. Therefore, their effluent was not as concentrated as the one presented in this study.

Moreover, the nutrient concentrations were much higher than those reported by Dal Ferro et al. (2018) for a SFCW also located in the Northern Italy, with a much bigger catchment area. There, due to the complexity of its drainage network, considerable part of TN and its components could have been removed (Castaldelli et al. 2018) before reaching the SFCW. On the other hand, in this study, the CER experimental farm is small and its drainage water collection ditch is no longer than 500 m, thus lower capacity for nutrient retention.

Both TN and NO<sub>3</sub><sup>-</sup>-N retention over the two-year period was 52% (Table 3.2). If single years are analyzed, the retention was only 40% in 2018. This value was lower than 68% reported by Haverstock et al. (2017), most probably due to the lower HRT. In fact, the average HRT during the Event 1, that represented 76% of the total yearly inflow, was 6.6 days (Table 3.3), while for the system studied by Haverstock et al. it was 15 days. Moreover, wetlands that receive steady flows of diffuse nitrate-rich run-off can achieve higher removals than those with pulse and inconsistent inflows (Tanner and Kadlec, 2013), such as the SFCW reported in this study. In 2019, the second year of monitoring, with a much lower inflow and consequently longer HRT than in 2018, TN and NO<sub>3</sub><sup>-</sup>-N retention was almost 80%. For example, the Event 7 accounted for 75% of the 2019 total inflow and had a HRT of 11.6 days.

The retention of TN mass load in 2018 was higher than in 2019, likely as a result of a much higher influent load of TN (Table 3.3). Expressed in surface terms, the SFCW retained 334 kg TN ha<sup>-1</sup> year<sup>-1</sup> in 2018 and 314 TN ha<sup>-1</sup> year<sup>-1</sup> in 2019. This is in line with Vymazal (2017) who found a median retention of 426 kg TN ha<sup>-1</sup> year<sup>-1</sup> for 41 CWs treating agricultural drainage water.

Influent TP concentration (0.05 mg L<sup>-1</sup>) was in line, but generally lower than those given by Johannesson et al. (2017) for several Swedish SFCWs treating runoff from arable land. Effluent concentration was slightly lower (0.02 mg L<sup>-1</sup>), but the load of this nutrient in the outlet was higher than in the inlet (Table 3.2). Since phosphorus removal occurs mainly by physical settling (Tanner and Kadlec, 2013), its negative removal, or, in other words, highest effluent than influent load, can be explained by flush out of sediments containing this element during a high flow event

(Kynkaanniemi et al. 2013). Moreover, the excess phosphorus might be a consequence of the vegetation decay and translocation and algal and microbial activity (Dal Ferro et al. 2018; Mendes et al. 2018b).

### 3.3.3.1. Retention efficiency during the Event 1 (Feb-Apr 2018)

Influent and effluent concentration trends for selected parameters during the Event 1 are given in Fig. 3.3. Influent concentration is generally higher than the effluent one, but they both followed a similar pattern. The average TSS, TN and  $\text{NO}_3^-$ -N influent concentration was 111.0, 15.6 and 12.0  $\text{mg L}^{-1}$ , respectively, while for effluent they were 73.2, 12.5 and 9.3  $\text{mg L}^{-1}$ , respectively (Fig. 3.3).

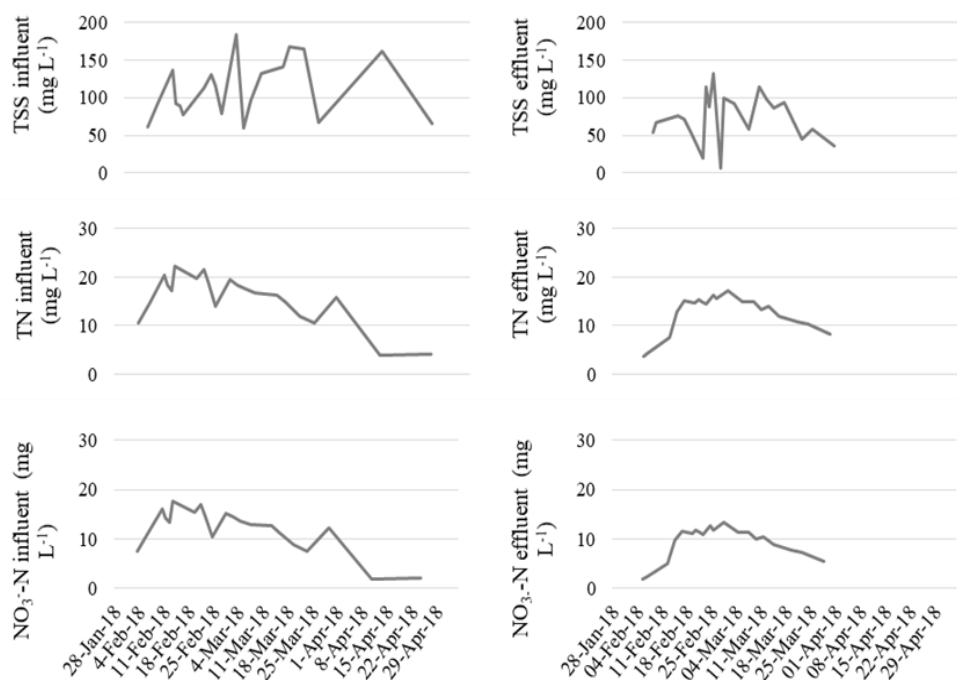


Fig. 3.3 - Influent and effluent concentrations of total suspended solids (TSS), total nitrogen (TN) and nitrate-nitrogen ( $\text{NO}_3^-$ -N) during the Event 1 (Feb-Apr 2018)

Being such a high inflow event that occurred in relatively short time (82 days), it is especially interesting to see how the SFCW reacted to the increased influent load and how it performed under constant stress. The retention efficiencies were generally lower comparing to the whole monitoring period (Table 3.4). Especially low retention of TOC (10%) was recorded, together with a negative retention of  $\text{NH}_4^+$ -N,  $\text{NO}_2^-$ -N and TP. That was probably caused by the fact that the Event 1 mostly happened during the winter season when low temperatures do not favor removal/retention processes. Moreover, the big flow of water inevitably lowered HRT in the system and therefore further reduced its treatment capacity. However, one of the main reasons could be the presence of

litter from the previous year. The above-ground vegetation usually starts its senescence in November and by February, when the Event 1 started, plant residue decomposition provides additional organic material on the bed surface. Therefore, although the litter/decomposition effect cannot be quantified, it might be considered as an additional load and pressure on the system during this specific inflow event.

Table 3.4 - Retention rates during the Event 1 (Feb-Apr 2018).

	Inflow load (kg)	Outflow load (kg)	Retention (%)
COD	418.3	226.7	46
TOC	129.7	116.4	10
TSS	2,030.4	926.7	54
TN	275.5	173.6	37
NH <sub>4</sub> <sup>+</sup> -N	1.12	1.59	-
NO <sub>3</sub> <sup>-</sup> -N	213.3	131.8	38
NO <sub>2</sub> <sup>-</sup> -N	1.08	1.58	-
TP	0.3	0.62	-

### 3.3.3.2. Nutrient input through precipitation

TP retention in other SFCWs treating agricultural drainage water was higher than in the present study. For example, Ulén et al. (2019) reported TP retention between 16 and 56% for a SFCW in Sweden, while Dal Ferro et al. (2018) concluded that a SFCW in the Italian region of Veneto removed 38% of influent PO<sub>4</sub>-P load. Although examples of negative retention of TP or its components were recorded (Kynkaanniemi et al. 2013; Dal Ferro et al. 2018), the authors of this study were intrigued that neither of the two years considered had a positive TP retention, while historical monitoring data of the same SFCW (Lavrnić et al. 2018), where nutrient atmospheric input has been included, showed that the system was able to remove TP.

Nutrients are present in rainwater in different concentrations (Vant and Gibbs, 2006, Hoffman et al. 2019), but their input through rainfall is usually not considered in the overall balance. However, it could represent a considerable input when nutrient load through influent is small, as is the case with TP in this study. Therefore, it was decided to monitor also rainwater quality during one inflow event (Event 6) and compare the precipitation and inflow nutrient loads to SCFW. Table 3.5 reports the concentration and load of nitrogen and its forms as well as phosphorus in both influent and rainwater.

The data suggest that, although NO<sub>3</sub><sup>-</sup>-N input through precipitation might be insignificant, rainwater

load can represent an important component of the overall balance of TN and especially for  $\text{NH}_4^+\text{-N}$  and TP. In fact, if precipitation is not considered in the overall balance, the retention of TP by the SFCW might seem negative (Table 3.3). However, when input by precipitation during only one inflow event is taken into account, the situation can change significantly, increasing TP input by about 25% and consequently changing TP retention from negative to positive.

Table 3.5 - Comparison of nutrient input through influent and precipitation in the Event 6 (Apr-May 2019).

		Influent	Precipitation
TN	Concentration ( $\text{mg L}^{-1}$ )	22.09	5.01
	Load (kg)	14.55	2.44
$\text{NO}_3^-\text{-N}$	Concentration ( $\text{mg L}^{-1}$ )	14.67	0.54
	Load (kg)	10.11	0.28
$\text{NH}_4^+\text{-N}$	Concentration ( $\text{mg L}^{-1}$ )	0.34	2.09
	Load (kg)	0.10	1.03
TP	Concentration ( $\text{mg L}^{-1}$ )	0.01	0.35
	Load (kg)	0.00	0.13

### 3.3.3.3. Comparison of two inflow events 15 years apart

In order to get a better insight on how the system performance changed over the years, two similar events, almost 15 years apart were compared for their nutrient retention efficiency (Table 3.6). One event occurred in 2005/06 and another in 2019 (Table 3.2). Both occurred in autumn and they were a result of similar rain episodes, 162 and 190 mm, respectively. It can be noticed that the 2005/06 event had a much lower water retention in respect to the 2019 one. While before the 2005/06 event the SFCW water level was at 31 cm, the 2019 one started with the empty SFCW. Therefore, such a big difference in effluent volume can be explained by water storage of water inside the system.

Although a comparison of two single events without analyzing the overall conditions might not be the best approach, some conclusions can still be drawn. The relatively big dissimilarity in performance of the SFCW during these two events could be connected to the big difference in effluent volume. However, the retention of nitrogen and its components during the 2019 event can be considered as high, suggesting that the system efficiency have not deteriorated over the years and that after 2 decades of constant operation it is still functioning properly.

Table 3.6 - Nutrient balance during two inflow events.

	30 <sup>th</sup> Nov 2005 - 23 <sup>rd</sup> Jan 2006			12 <sup>th</sup> Nov 2019 - 23 <sup>rd</sup> Dec 2019		
	Inflow	Outflow	Retention	Inflow	Outflow	Retention
Water volume (m <sup>3</sup> )	5,220	4,139	-	6,100	1,943	-
TN (kg)	110.7	60.3	46%	99.7	30.4	70%
NO <sub>3</sub> <sup>-</sup> -N (kg)	94.7	59.3	37%	78.1	23.3	70%
NH <sub>4</sub> <sup>+</sup> -N (kg)	0.22	0.25	-	2.32	0.19	92%
TP (kg)	0.09	0.21	-	0.00	0.00	-

### 3.3.4. Soil nutrient and trace elements content

Fig. 3.4 reports concentration of different elements, including nutrients and trace elements, in the soil layer. A comparison between the 2017 and 2019 results is given. The change in content between two years was minimal and the values were within the same order of magnitude. Therefore, 2 years might not be enough to notice any substantial change up to 60 cm soil depth and longer time gaps should be considered.

In particular, TOC concentration was 10.0 and 8.8 g kg<sup>-1</sup> in 2017 and 2019, respectively, and it was similar to the values reported by Maucieri et al. (2014) for a SFCW located near Padua (Italy). TN content was in line with the values found by Passoni et al. (2009), whereas TP concentration was much smaller (0.5 vs. 6.5 g kg<sup>-1</sup>). This difference might be a result of a higher TP load or a different soil type. However, both TN and TP content of the SFCW studied here were comparable to the values of the surface soil layer obtained for the same system between 2004 and 2009 (Lavrnić et al. 2018).



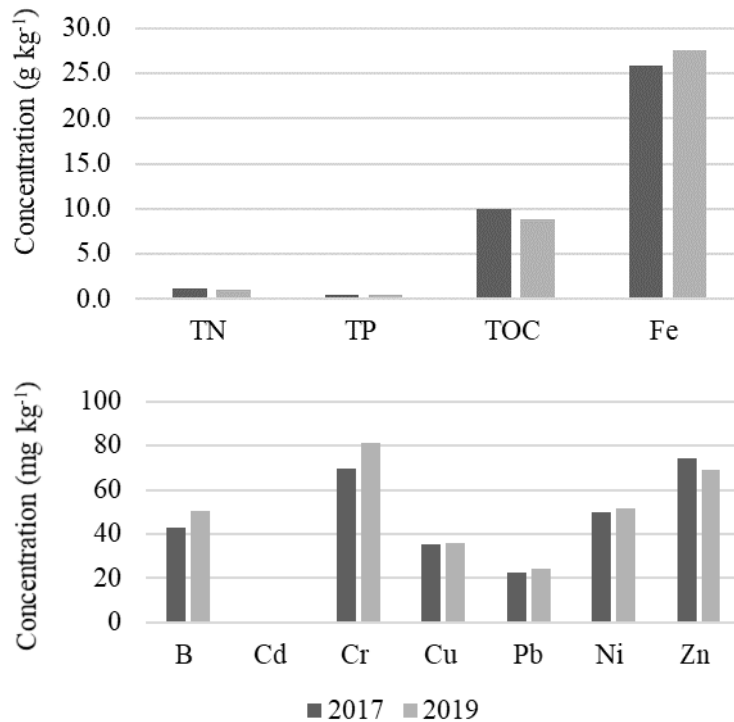


Fig. 3.4 - Concentration of nutrients and trace elements in the SFCW soil.

Iron showed the highest concentration ranging from 25 to 28 g kg<sup>-1</sup>. The average concentration of chrome increased from 69 mg kg<sup>-1</sup> in 2017 to 81 mg kg<sup>-1</sup> in 2019. Nevertheless, the content of all trace elements in 2019 was comparable to the background data (RER, 2020), and they were within the limits for green areas, private and residential use that were established in 2006 by the Italian law (D.Lgs, 2006).

### 3.3.5. Vegetation development and its nutrient and trace elements accumulation

Plants can play a positive role on wastewater treatment of CWs, such as by providing a habitat for microbial communities and favorable oxygen transfer (Abou-Elela and Hellal, 2012), uptake of nutrient and heavy metals (Fountoulakis et al. 2017) and facilitating some physical mechanisms (e.g. filtration and sedimentation) (Vymazal, 2011). Also, it is reported that heterogeneity of plant species may provide more ecological and aesthetic values due to the presence of structurally and floristically diverse plants patterns (Tanner, 1996). Vegetation used in CWs can be also used for energy purposes due to their high biomass production with considerable heating values (Molari et al. 2014).

Table 3.7 shows the main results of the plant sampling that was done yearly: before the experimental period in 2017, during it in 2018 and at the end of the experimental period in 2019.

Unfortunately, it was not possible to identify all the plant species present in the SFCW, but the three dominant ones were *Phragmites australis*, *Typha latifolia* and *Carex* spp.

Table 3.7 - Agronomic characteristics of the main plant species in the SFCW.

		Surface area covered (%)	Dry weight (kg m <sup>-2</sup> )	Above/below ground biomass ratio (%)	Average height (cm)
2017	<i>Phragmites australis</i>	41	7.43	0.13	194
	<i>Typha latifolia</i>	14	4.95	0.36	168
	<i>Carex</i> spp.	45	3.69	0.21	87
2018	<i>Phragmites australis</i>	58	6.24	0.25	251
	<i>Typha latifolia</i>	14	9.19	0.15	231
	<i>Carex</i> spp.	28	3.87	0.70	184
2019	<i>Phragmites australis</i>	73	9.43	0.27	266
	<i>Typha latifolia</i>	11	5.14	0.12	125
	<i>Carex</i> spp.	16	10.63	0.14	178

If the surfaces inhabited by the species are compared, it can be seen that *Phragmites australis* gradually increased its surface, occupying in 2019 more than 70% of the SFCW area. *Phragmites australis* mostly spread on the account of *Carex* spp., while the surface coverage by *Typha latifolia* was generally constant. Together with increasing its surface area, *Phragmites australis* also increased its above/below ground biomass ratio and average height. On the other hand, the other two dominant species did not follow the same trend. In general, the total biomass of the SFCW was increasing over the years: from 16 kg m<sup>-2</sup> in 2017, it rose to 19 kg m<sup>-2</sup> in 2018 and to 25 kg m<sup>-2</sup> in 2019. Interestingly, the biomass of *Carex* spp. had a constant increase, while the other two species showed a certain fluctuation from 2017 to 2019.

Fig. 3.5 displays the concentration of boron, some heavy metals and nutrients contained in plants between 2017 and 2019. In agreement with the results reported by Yadav et al. (2012), the content of B, Cu, Fe, Ni, Pb and Zn in the below-ground biomass was much higher than the observed in the above-ground biomass. Constant decrease of B and Fe concentration in the biomass (Fig. 3.5) was

probably related to the slight increase of their concentrations in the SFCW soil that observed in the same period (Fig. 3.4).

The total annual accumulation and distribution of nutrients and carbon in plants are shown in Fig. 3.6. Clearly, the below-ground biomass accumulated more TN, TP and TOC than the above-ground one. That feature was in line with the study of Borin and Tocchetto (2007), who emphasized that more TN was retained in the below-ground biomass of *Typha latifolia* and *Phragmites australis* because of their transfer from above to below-ground organs during the dormant periods. Chanc et al. (2019) found high level of TN and TP stored in above-ground biomass of *Juncus effusus* and *Pontederia cordata*, sampled before their senescence, when nutrient transfer still had not taken place.

The uptake rates of TN and TP by above-ground vegetation were in the range of 19.0-26.3 and 1.6-2.1 g m<sup>-2</sup>, respectively. These results were comparable with the ranges of 22.3-41.1 g TN m<sup>-2</sup> and 1.4-3.8 g TP m<sup>-2</sup> obtained by Vymazal and Dvořáková Březinová (2018b). The TP in the biomass was 14.1, 11.6 and 21.9 kg in 2017, 2018 and 2019, respectively. The total TN content varied from 113.8 to 208.5 kg over the three years, while the TOC accumulation was in the range of 5,574.1-11,311.9 kg (Fig. 3.6). As a general observation, TP, TN and TOC showed similar values in the 2017-2018 period, followed by a significant increase in 2019. This was consistent with the occupation of SFCW surface by the main plant species, *Phragmites australis*. In fact, among the others, the species in 2019 reached the maximal extension in term of average height and surface area covered (Table 3.7).

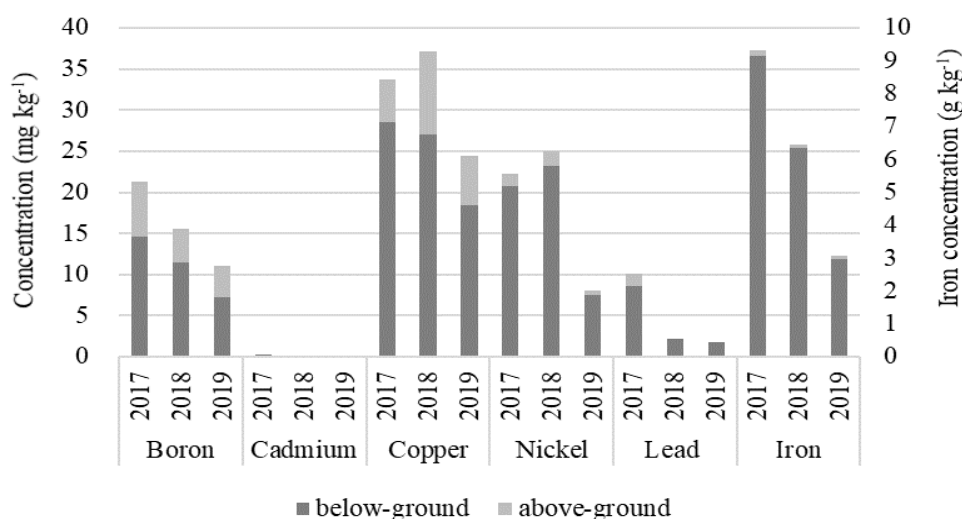


Fig. 3.5 - Distribution of trace elements between above and below-ground biomass.

If compared to the overall balance of the SFCW, the vegetation stored more of these elements than the amount that was removed annually based on the difference between influent and effluent loads (Table 3.3). However, it should be noted that the above-ground vegetation was not harvested after spring 2017 and therefore it was not possible to do the overall nutrient balance. Although biomass harvesting is recommended to achieve the full nutrient removal potential of vegetation, plant litter can also provide organic matter and suitable attachment surfaces for bacteria inside the system, and thus facilitate removal processes, primarily denitrification (Song et al. 2019). An in-depth analysis should be done in order to have a better insight into nutrient circulation inside the SFCW.

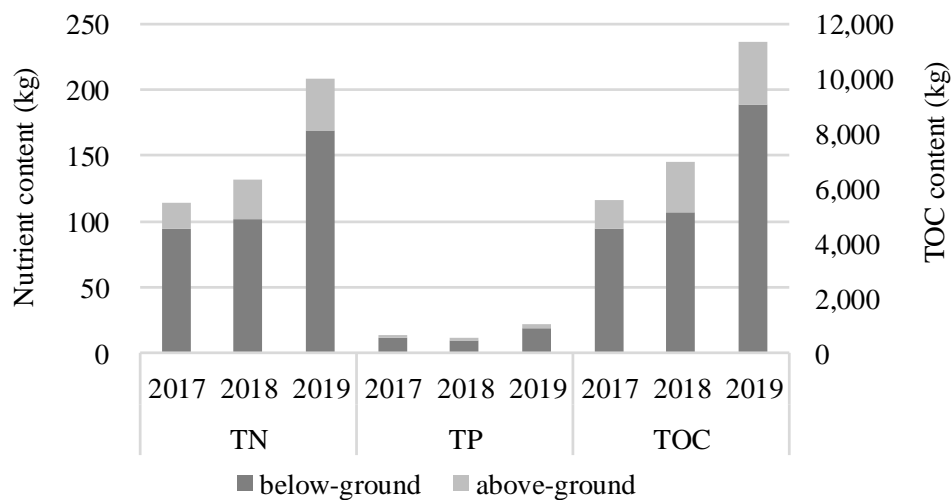


Fig. 3.6 - Total content of total nitrogen (TN), total phosphorus (TP) and total organic carbon (TOC) in below and above-ground biomass.

### 3.4. Conclusions

Calculation of retention efficiencies based on concentrations might not be the best choice when assessing performance of a non-waterproofed SFCWs. These systems, especially the ones receiving a varying flow like agricultural drainage water, can experience increased water losses and therefore effluent can have a higher concentration of pollutants than influent. To overcome misinterpretation of data, mass load retention can be considered much more appropriate. In order to obtain correct retention efficiencies, it is also important to consider input of nutrients directly through precipitation, since it can represent a sizeable part of the total incoming load. In the SFCW studied, although retention of TP seemed to be poor, additional analysis showed that the result can considerable change when precipitation input is taken into account.

The soil structure and concentration of nutrients and trace elements did not considerably change

over 2 years of monitoring, and, for this reason an assessment of soil over longer time periods is recommended. In addition, the increase of plant total biomass over the years, together with TN, TP and TOC content was indicative of the presence of favorable conditions for the macrophytes development. Further study is needed to close the nutrient balance of the system since it was not possible to estimate the loss of TN and TP through infiltration. That would certainly contribute to the better understanding of dynamics and retention processes occurring inside SFCWs.

Overall, the SFCW studied showed a high efficiency for agricultural drainage water treatment (e.g. up to 82% TSS retention and up to 78% for TN and  $\text{NO}_3^-$ -N retention), even if it has been in use for two decades. These results indicate that SFCWs can be considered as a cost-effective and long-term ecological engineering solutions for the reduction of non-point source pollution and prevention of eutrophication and deterioration of surface water bodies.

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# CHAPTER 4 - Potential of constructed wetland treatment systems for agricultural wastewater reuse under the EU framework

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*Key words: Wastewater treatment; Constructed wetland; Agricultural wastewater reuse*

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## Abstract

Wastewater reuse was recognized as one of the solutions for the problems regarding increasing water scarcity and pollution of water resources. Constructed wetlands (CWs) are a sustainable and cost-effective technology for wastewater treatment. If able to produce effluent of a needed quality, they can be a valuable addition for wastewater reuse schemes. This review studied 39 treatment systems based on CWs, and it assessed their characteristics and performance on pollutants removal. Moreover, their potential to reach the future European Union standards for agricultural wastewater reuse was evaluated. The results showed that the combination of CWs with additional technologies (e.g. UV treatment, anaerobic reactors) can further increase their performance and provide better removal efficiencies in comparison with conventional horizontal and vertical subsurface flow CWs. Particularly, hybrid systems showed a better removal of organic matter and bacterial indicators than single-stage CWs. Most of the systems considered could reach some of the limits for agricultural reuse in the terms of biochemical oxygen demand and total suspended solids, although improved single-stage CWs and hybrid systems were able to meet stricter requirements. However, that was often not the case with *Escherichia coli* and therefore it is recommended to combine them with disinfection technologies in order to reach the levels required for agricultural reuse.

## 4.1. Introduction

Currently, water scarcity is becoming a worldwide risk (Mekonnen and Hoekstra, 2016). The severe pressure on water resources is mainly attributed to global population growth, expansion of irrigation agriculture, economic development and climate change (Gosling and Arnell, 2016; Hamadeh et al., 2014; Mekonnen and Hoekstra, 2016; Tao et al., 2017). In fact, existing natural freshwater resources seem to be inadequate to satisfy various ever-increasing demands (Almukhtar et al., 2018) and therefore imbalance between water demand and water supply (Ghaitidak and Yadav, 2013). Moreover, discharging wastewater without previous treatment may not only lead to a certain “waste” of water resources, but can also harm different ecosystems (Lavrnić et al., 2018). On the other hand, treated wastewater is regarded as an alternative resource for water supply, and also has the potential to be used for different purposes (Barbagallo et al., 2012; Hamadeh et al., 2014; Tao et al., 2017). Under such circumstances, research on wastewater reclamation strategies is being carried out in order to address increasing demand for water resources and to prevent further deterioration of water quality (Almukhtar et al., 2018; NAS, 2016).

Apart from conventional wastewater treatment technologies, nature-based solutions, especially constructed wetlands (CWs), have been used worldwide for treating wastewater and improving its quality (Hamadeh et al., 2014). CWs are engineered ecological system that can treat wastewater through different natural processes which are under the influence of the combined action of aquatic plants, soils and microorganisms (Hamadeh et al., 2014; Tao et al., 2017).

CWs are viewed as a cost-effective and sustainable option for wastewater treatment (Arden and Ma, 2018; ElZein et al., 2016; Lee et al., 2009). Their main advantages are good removal efficiency, simple and low cost construction, running and maintenance, nutrients recycle, energy biomass production and esthetic values (Brix et al., 2011; Chen, 2011; Hamadeh et al., 2014; Liu et al., 2015; Molari et al., 2014; Rousseau et al., 2008). However, CWs also have certain shortcomings. For example, there is a certain risk of bed clogging (Lavrnić et al., 2020), especially under high loading rates of organic and suspended solids (SS). It can later cause hydraulic malfunction and decrease overall treatment performance, even further shortening the lifespan of systems (Aiello et al., 2016; Barbagallo et al., 2011; Józwiakowski et al., 2018; Kim et al., 2016; Ruiz et al., 2010). Besides, nitrogen removal efficiency of CWs is sometimes limited due to insufficient conditions for denitrification and nitrification (Józwiakowski et al., 2018; Liu et al., 2015; Wu et al., 2014). Also, pathogen removals could be low due to the lack of disinfection treatment or other chemical agents (Andreo-Martínez et al., 2017), and unsatisfactory performance of single-stage CW (Toscano et al.,

2015; Zurita and White, 2014), while antibiotic resistance in CW is a new challenge (Russo et al., 2019b). In order to overcome these problems, it can be a good solution to integrate CWs with right disinfection measures (Arden and Ma, 2018), artificial aeration technologies, anaerobic baffled reactor (ABR), even combining different types of CWs, etc.

CWs effluents can be reused in irrigation, gardening, flushing toilet, groundwater replenishment and other public and industrial utilizations (Angelakis and Snyder, 2015; Barbagallo et al., 2014; Dou et al., 2017; Rousseau et al., 2008). Being one of the biggest consumers of freshwater resources in the world, agriculture is under a constant threat of climate change and water scarcity, and in order to ensure sufficient crop production, additional water resources (e.g. treated wastewater) need to be used. Nevertheless, although wastewater reuse can be beneficial for agricultural irrigation, negative effects of this practice (e.g. on soil or plants) should be considered. Thus, it is important to regulate this area and prevent negative consequences on the environment. Several countries (e.g. Italy, Spain, U.S.) have already implemented regulations and limits for wastewater reuse in agriculture (Andreo-Martínez et al., 2017; Jokerst et al., 2011; Licciardello et al., 2018). Some of these countries provide detailed classification of irrigation water quality according to crop categories, irrigation methods and areas (Ayaz et al., 2015; Russo et al., 2019a). Also, the European commission proposed a regulation on minimum requirements for water reuse in agriculture (European Commission, 2018). However, wastewater treated by CWs cannot always satisfy these standards related to safe reuse (Arden and Ma, 2018; García et al., 2013; Jokerst et al., 2011; Lavrić et al., 2017).

Therefore, the main objective of this paper is to provide an overview on the design, characteristics, as well as performance of CWs treating domestic wastewater in order to assess their potential for wastewater reuse within the new EU framework (European Commission, 2018).

## 4.2. Materials and methods

This review mainly focuses on domestic wastewater treatment systems based on CWs. It is a result of the search of Web of Science database using keywords such as “constructed wetland”, “domestic water/wastewater treatment” and “water/wastewater reuse (in agriculture)”. The time period of publication was set from 2008 to 2019. According to the main objective of this review, 39 research publications which focus on domestic wastewater treatment, pollutants removal effect and reuse in agriculture were selected to be discussed. On the other hand, papers which did not report pollutant concentrations or removal rates were excluded from the selection.

The 39 articles analyzed cover 19 countries. Regular domestic wastewater (a mixture of kitchen, shower, toilet etc.) treatment is the main topic of 77% of the articles considered, while greywater (domestic wastewater from non-toilet sources) and blackwater (domestic wastewater from toilets) amount to 18% and 5%, respectively. Hybrid CWs or CWs combined with other systems are 64% of the total, while single-stage CWs are 36%. They were mostly of pilot-scale (51%), but full-scale (31%) and lab-scale studies (18%) were also well represented (Fig. 4.1a). Regarding experimental duration, 49% of the research studies lasted for more than one year, 33% were between six months and one year long, 10% were shorter than six months, while the remaining 8% did not provide a specific time (Fig. 4.1b). Plant species applied or tested in these studies were either a mixture of various species or different species planted separately.

In Table 4.1 are listed the 39 selected experimental case studies. Each case study was given a number from 1 to 39, as reported in Table 4.1 (system No.), and that number was later used to refer to a particular case study in the following sections of the paper. The efficacy of these various wastewater treatment systems was analyzed and conformity of their effluents to reuse limits evaluated. This research did not introduce studies on single free surface flow CWs, since not many cases suitable for this review were reported in the time frame considered. However, free surface flow CW were considered if combined with other technologies in a hybrid system. It was also noted that some studies in hybrid system section (Table 4.1) tested more than one system, either with similar (No. 16, 17, 18, 20, 35 and 37) or different configurations (No. 27a, 27b, 36a and 36b).

In the case when the paper did not provide influent, effluent concentration or removal efficiency, the following equation was used to calculate the missing variable:

$$\frac{C_i - C_e}{C_i} \times 100 = E \quad (1)$$

Where  $C_i$  = influent concentration of a pollutant ( $\text{mg L}^{-1}$ ),  $C_e$  = effluent concentration of a pollutant ( $\text{mg L}^{-1}$ ), and  $E$  = removal efficiency of a pollutant in a system (%).

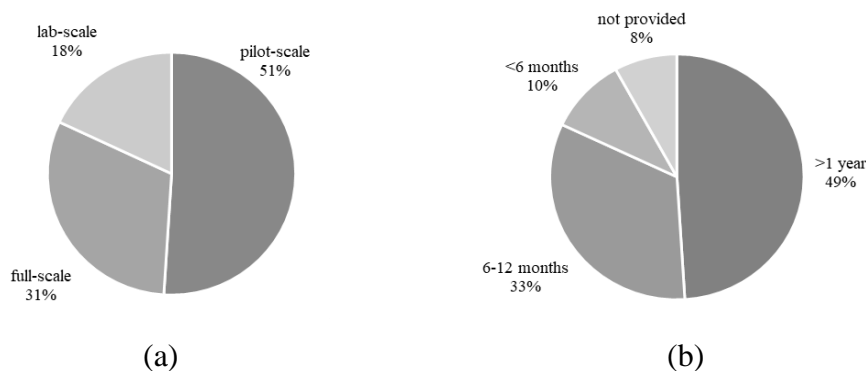


Fig. 4.1 - Distribution of case studies based on (a) constructed wetland scales and (b) experimental duration.

Table 4.1 - 39 Selected case studies of wastewater treatment systems recorded in the literature from 2008 to 2019.

CW types	System No.	Experimental scale	Country	Influent	Experimental period	Vegetation	Reference
Single-stage CW							
HSSF	1	Pilot-scale	Italy	RDW	>1 year	<i>Cyperus alternifolius</i> L. (unit 1), <i>Typha latifolia</i> L. (unit 2), unplanted (unit 3)	Tuttolomondo et al. (2016)
	2	Pilot-scale	Italy	RDW	6-12 months	<i>Vetiveria zizanioides</i> , <i>Miscanthus x giganteus</i> , <i>Arundo donax</i> , <i>Phragmites australis</i>	Toscano et al. (2015)
	3	Pilot-scale	Spain	RDW	>1 year	<i>Phragmites australis</i>	Morat ó et al. (2014)
	4	Pilot-scale	Turkey	RDW	>1 year	<i>Cyperus</i>	Ayaz (2008)
	5	Full-scale	Poland	RDW	>1 year	<i>Salix viminalis</i> L.	Józwiakowski et al. (2018)
	6	Pilot-scale	Spain	RDW	6-12 months	<i>Phragmites australis</i>	Andreo-Mart ínez et al. (2017)
VSSF	7	Pilot-scale	Italy	RDW	>1 year	<i>Typha latifolia</i> , <i>Phragmites australis</i>	Morari and Giardini (2009)
	8	Pilot-scale	Egypt	RDW	>1 year	<i>Canna</i> , <i>Phragmites australis</i> and <i>Cyperus papyrus</i>	Abou-Elela and Hellal (2012)
	9	Pilot-scale	Greece	RDW	6-12 months	<i>Atriplex halimus</i> , <i>Juncus acutus</i> and <i>Sarcocornia perennis</i> , <i>Phragmites australis</i>	Fountoulakis et al. (2017)
	10	Lab-scale	UK	RDW	>1 year	<i>Phragmites australis</i>	Almuktar et al. (2017)
Improved system GROW	11	Pilot-scale	India	GW	>1 year	8 varieties of Indian native plant species	Ramprasad et al. (2017)
Improved system RVFCW	12	Lab-scale	Israel	GW	>1 year	<i>Juncus alpigenus</i> and <i>Cyperus haspen</i>	Sklarz et al. (2009)
	13	Lab-scale	Israel	GW	<6 months	<i>Hydrocotyle leucocephala</i> and <i>Cyperus papyrus</i>	Travis et al. (2010)
Improved system upflow subsurface CW using green sorption media	14	Pilot-scale	USA	GW	<6 months	<i>Juncus effuses</i> (cell 1), <i>Panicum hemitomon</i> (cell 2), <i>Zizaniopsis miliacea</i> (cell 3)	Xuan et al. (2009)
Hybrid systems							



CW types	System No.	Experimental scale	Country	Influent	Experimental period	Vegetation	Reference
VSSF-HSSF	15	Full-scale	South Korea	RDW	>1 year	<i>Phragmites australis</i> and <i>Phragmites japonica</i> (VF), <i>Miscanthus sacchariflorus</i> , <i>Carex dispalata</i> , <i>Juncus effuses</i> , and <i>Iris pseudacorus</i> (HF)	Kim et al. (2016)
Two VSSF-HSSFs in parallel <sup>a</sup>	16	Pilot-scale	Spain	RDW	6-12 months	<i>Phragmites australis</i> and <i>Scirpus</i> sp.	Herrera-Melián et al. (2010)
Four separate HSSF-VSSF <sup>b</sup>	17	Pilot-scale	Iran	RDW	6-12 months	<i>Phragmites australis</i> , <i>Typha latifolia</i> , <i>Arundo donax</i> , unplanted	Haghshenas-Adarmanabadi et al. (2016)
HSSF-HSSFs, VSSF-VSSFs, VSSF-HSSF <sup>c</sup>	18	Lab-scale	Colombia	RDW	Not provided	<i>Papyrus</i>	García et al. (2013)
FWS-SSF	19	Pilot-scale	USA	GW	6-12 months	<i>Typha latifolia</i> (FWS), <i>Scirpus acutus</i> (SSF)	Jokerst et al. (2011)
HSSF-VSSF <sup>d</sup>	20	Lab-scale	Spain	RDW	>1 year	Common reed and <i>Papyrus</i> (HSSF), not provided (VSSF)	Herrera-Melián et al. (2018)
IVFCW-HSSF	21	Pilot-scale	China	RDW	6-12 months	<i>Canna indica</i> L. (down-flow VSSF), <i>Juncus effusus</i> L. (up-flow VSSF), <i>Scirpus validus</i> Vahl (HSSF)	He et al. (2018)
Saturated VSSF-free-drain VSSF-HSSF	22	Pilot-scale	Czech Republic	RDW	>1 year	<i>Phragmites australis</i> (saturated VSSF), <i>Phragmites australis</i> (free-drain VSSF), <i>Phalaris arundinacea</i> (HSSF)	Vymazal and Kröpfelová (2015)
VSSF-HSSF-FWS	23	Full-scale	Spain	RDW	Not provided	<i>Phragmites australis</i> (VSSF), <i>Phragmites australis</i> (HSSF), <i>Typha</i> spp., <i>Scirpus</i> spp., <i>Iris pseudacorus</i> , <i>Carex flacca</i> , <i>Cyperus rotundus</i> and <i>Juncus</i> spp. (FWS)	Ávila et al. (2015)

CW types	System No.	Experimental scale	Country	Influent	Experimental period	Vegetation	Reference
Settling cum equalization tank-UFDF sand filter-HSSF-charcoal filter-water hyacinth system	24	Full-scale	India	GW	6-12 months	<i>Canna indica</i>	Patil and Munavalli (2016)
BCO pretreatment-greenhouse-structured HSSF	25	Full-scale	China	RDW	>1 year	ornamental plants including <i>Hemerocallis lilioasphodelus</i> L., <i>Iris tectorum</i> , <i>Oxalis violacea</i> , <i>Sedum erythrostictum</i> Mig and <i>Hosta ensata</i>	Gao and Hu (2012)
Ice-block unit <sup>o</sup> -VSSF	26	Pilot-scale	Mongolia	GW	6-12 months	not provided	Uddin et al. (2016)
HSSF-lagooning	27a	Full-scale	Italy	RDW	6-12 months	<i>Phragmites australis</i> , <i>Typha latifolia</i>	Russo et al. (2019a)
HSSF-UV treatment	27b	Full-scale	Italy	RDW	6-12 months	<i>Phragmites australis</i> , <i>Typha latifolia</i>	Russo et al. (2019a)
RVFCW-UV disinfection	28	Lab-scale	Israel	RDW	>1 year	<i>Cyperus haspen</i> , <i>Juncus alpigenus</i> and <i>Hydrocotyle vulgaris</i> L.	Sklarz et al. (2013)
SSF-UV/TiO <sub>2</sub> /O <sub>3</sub>	29	Pilot-scale	Brazil	BW	>1 year	<i>Hymenachne grumosa</i>	Horn et al. (2014)
Anaerobic pretreatment-HSSF-VSSF	30	Pilot-scale	Turkey	RDW	<6 months	<i>Phragmites australis</i>	Ayaz et al. (2015)
HUSB reactor-VSSF-HSSF-FWS	31	Lab-scale	Spain	RDW	>1 year	<i>Phragmites australis</i>	Ávila et al. (2016)
OP-FWS-Cascade-FWS-SSF	32	Full-scale	China Taiwan	RDW	6-12 months	<i>Typha latifolia</i> and <i>Phragmites australis</i> (FWS), <i>Phragmites australis</i> (SSF)	Yeh et al. (2010)
FP-FWS-SSF	33	Full-scale	Spain	RDW	6-12 months	<i>Typha latifolia</i> (SF), <i>Salix atrocinerea</i> (SSF)	Reinoso et al. (2008)
Sedimentation tank-HSSF-VSSF	34	Pilot-scale	Egypt	BW	Not provided	<i>Phragmites</i>	Abdel-Shafy et al. (2017)

CW types	System No.	Experimental scale	Country	Influent	Experimental period	Vegetation	Reference
Combinations of HSSF, VSSF or stabilization pond <sup>f</sup>	35	Pilot-scale	Mexico	RDW	>1 year	<i>Zantedeschia aethiopica</i> , after 8 months (replaced with) <i>Canna indica</i> (HSSF), <i>Strelitzia reginae</i> (VSSF)	Zurita and Carreón-Álvarez (2015)
HSSF-biological pond-storage reservoir-sand and disk filters	36a	Full-scale	Italy	RDW	<6 months	<i>Phragmites australis</i>	Licciardello et al. (2018)
HSSF-sand and disk filters-UV treatment	36b	Full-scale	Italy	RDW	<6 months	<i>Phragmites australis</i>	Licciardello et al. (2018)
ABR-VSSF/HSSF-FWS <sup>g</sup>	37	Full-scale	Pakistan	RDW	6-12 months	<i>Typha latifolia</i> , <i>Phragmites australis</i> and vetiver grass (VSSF, HSSF), <i>Pistia stratiotes</i> (FWS)	Ali et al. (2018)
Two settling tanks in series-VSSF-a zeolite tank	38	Full-scale	Greece	RDW	>1 year	Unplanted (cell 1), <i>Phragmites australis</i> (cell 2)	Gikas and Tsihrintzis (2012)
A septic tank-an Imhoff tank-two parallel VSSFs-HSSF	39	Full-scale	Spain	RDW	>1 year	<i>Typha latifolia</i> (HSSF)	Vera et al. (2013)

HSSF = horizontal subsurface flow constructed wetland, VSSF = vertical subsurface flow constructed wetland, RDW = regular domestic wastewater, GW = greywater, BW = blackwater, GROW = green roof-top water recycling system, RVFCW = recirculating vertical flow constructed wetland, FWS = free water surface constructed wetland, SSF = subsurface flow constructed wetland, IVFCW = integrated vertical flow constructed wetland, UFDF = up-flow down-flow filter, BCO = bio-contact oxidation, UV = ultraviolet, HUSB = hydrolytic up-flow sludge blanket, OP = oxidation pond, FP = facultative pond, ABR = anaerobic baffled reactor

<sup>a</sup> System 1: VSSF-HSSF both with lapilli, system 2: VSSF-HSSF both with gravel

<sup>b</sup> System 1: *Phragmites* HSSF-*Phragmites* VSSF, system 2: *Typha* HSSF-*Typha* VSSF, system 3: *Arundo* HSSF-*Arundo* VSSF, system 4: unplanted HSSF-unplanted VSSF

<sup>c</sup> System 1: HSSF-HSSF planted, system 2: HSSF-HSSF unplanted, system 3: VSSF-VSSF planted, system 4: VSSF-VSSF unplanted, system 5: VSSF-HSSF planted, system 6: VSSF-HSSF unplanted

<sup>d</sup> System 1: Mulch-based HSSF-gravel-based VSSFs, system 2: Mulch-based HSSF-mulch-based VSSFs

<sup>e</sup> The system functioned as the storage pond of frozen wastewater in winter and transferred to septic tanks for treating melted wastewater in summer

<sup>f</sup> System 1: HSSF-stabilization pond, system 2: HSSF-VSSF, system 3: VSSF-HSSF

<sup>g</sup> System 1: ABR-Saturated VSSF-FWS, system 2: ABR-HSSF-FWS

## 4.3 Single-stage CW

On the basis of wetland flow, CWs are classified into free water surface (FWS) and subsurface flow (SSF). SSF CWs are the most widely used systems (Fonder and Headley, 2013) and can be subdivided into two specific types, horizontal (HSSF) and vertical (VSSF) one. Generally, SSF systems show a better performance than FWS ones, especially when hydraulic loading rate (HLR) is high (Liu et al., 2009). Certain modifications of the original CW types yielded also some novel technologies such as a green roof-top water recycling system (GROW) CW (Avery et al., 2007) and a recirculating vertical flow constructed wetland (RVFCW) (Gross et al., 2007). Improved systems are thought to be an option to save land resource (Ramprasad et al., 2017), optimize organic matter and biogenic compounds removal (Sklarz et al., 2009; Xuan et al., 2009), decrease the likelihood of human contact with wastewater (Sklarz et al., 2009) and environmental risks of its reuse (Travis et al., 2010), meanwhile achieving a relatively good overall treatment efficiency (Ramprasad et al., 2017; Sklarz et al., 2009; Travis et al., 2010). Furthermore, CWs can be filled with different substrates and vegetated by aquatic plants (Chen, 2011; Herrera-Melián et al., 2010; Toscano et al., 2015), which were suggested to be able to improve pollutants removal (Arden and Ma, 2018; Liu et al., 2009).

### 4.3.1. Horizontal subsurface flow CW (HSSF CW)

Until now, many studies on HSSF CWs have been implemented and reported. More HSSF systems were operated in Europe and United states than VSSF CWs (Nivala et al., 2019), and showed reliable capacity for total suspended solids (TSS) and biochemical oxygen demand (BOD) removal (Lavrnić et al., 2017). However, it is worth noting that these systems may not ensure a stable and good removal efficiency of phosphorus, nitrogen and organics (Andreo-Martínez et al., 2017), as a result of a lack of metal ions (e.g. Ca, Mg, Fe and Al) in conventional substrates (Vohla et al., 2011) and a lack of dissolved oxygen (DO) in water (Vymazal, 2007).

In the west of Sicily, Italy, Tuttolomondo et al. (2016) carried out a two-year experiment on a pilot-scale HSSF CW system (containing three independent units) where one unit was planted with *Cyperus alternifolius*, one with *Typha latifolia* and the third was unplanted. The system was filled with silica quartz river gravel (a particle size of 20 to 30 mm) and operated at a HLR of 12 cm d<sup>-1</sup>. The mean removal rates based on concentrations of parameters chemical oxygen demand (COD) and BOD<sub>5</sub> are shown in Table 4.2 (Table 4.2, No. 1). Evapotranspiration (ET), as a primary factor of the system water balance, had an influence on available treated water volume. The findings

showed that the observed removal efficiency of BOD<sub>5</sub> and COD was negatively correlated with ET. Therefore, ET should be taken into consideration especially for arid areas when wastewater reuse in irrigation is the objective. Similarly, in Sicily, Toscano et al. (2015) tested pilot HSSF CWs filled with volcanic gravel to a depth of 0.6 m for tertiary treatment of domestic wastewater from March to November 2012. The system contained two lines, each of which consisted of five parallel HSSF beds. Four were planted with macrophytes (*Vetiveria zizanoides*, *Miscanthus x giganteus*, *Arundo donax*, *Phragmites australis*, respectively) and the fifth one was unplanted. The authors indicated that the vegetated CWs were more effective in contaminants removal. The average removal efficiencies based on concentrations for planted CWs were 92.8% for TSS, 68.1% for COD and 61.3% for total nitrogen (TN), and they were higher than the ones of unplanted systems that were 89.4%, 55.4% and 43.1%, respectively. The best performance of pollutants removal was attained by the system planted with *Phragmites australis*, with efficiencies of 99.9% for *Escherichia coli* (*E. coli*), 88% for TSS, 63% for COD and 61% for TN (Table 4.2, No. 2), thus regarded as the most suitable plant species for wastewater treatment in this case. Similarly to the previous study, it was reported that vegetation has affected water balance of systems leading to different ET values measured in planted and unplanted wetlands.

In Spain, Morató et al. (2014) evaluated the effect of design factors (water depth and gravel granulometry) on treatment efficiency of HSSF systems. As can be observed in Table 4.2, the mean removal efficiencies of COD and BOD<sub>5</sub> were 63.8% and 65%, respectively (Table 4.2, No. 3). It was found that the system with the water depth of 0.27 m and the size of granular medium of 3.5 mm, was more effective for microbial removal in comparison with other systems (0.5 m water depth, 10 mm size of granular medium). The removal effectiveness in the system with a fine medium may be explained by a larger proportion of water volume contacting with root systems of the vegetation, beneficial to microbial reduction. Microbial removal was primarily attributed to mechanisms of filtration and sedimentation occurring near the inlet of HSSF CWs. Moreover, seasonal variations affected removal of some bacterial groups - a higher removal of *E. coli*, total coliforms (TC) and *Clostridium* spores was achieved in summer, while for heterotrophic plate count it was during winter. Ayaz (2008) also demonstrated the effect of seasonal changes on removal efficiency, since removals of BOD<sub>5</sub> and COD were greater during summer. Furthermore, the author indicated that HSSF CWs were more effective for SS (80%), BOD<sub>5</sub> (65%) and COD (50%) removal in comparison with VSSF and FWS CWs. The only exception was total organic carbon (TOC), which obtained low removal efficiency (sometimes even negative one) in all wetlands due to the additional generation of organic carbon by planted vegetation. The mean removal efficiency of fecal

coliforms (FC) and TC both amounted to more than 94% (Table 4.2, No. 4).

Jozwiakowski et al. (2018) carried out a 14-year investigation on a HSSF wetland in Poland operated under a HLR of 0.6 cm d<sup>-1</sup>. The 1.2 m deep system was filled with sand and *Salix viminalis* L. was planted in the humus layer distributed over the sand layer. It was observed that for TN and total phosphorus (TP) it was not possible to achieve a continuous and satisfactory removal in the long-term (Table 4.2, No. 5). It could be explained by the fact that HSSF system did not provide sufficient conditions for nitrification, which in turn negatively affected TN removal. Regarding TP, the sorption capacity of substrate declined over time, causing a lower abatement efficiency of TP during later period of the experiment.

#### 4.3.2. Vertical subsurface flow CW (VSSF CW)

VSSF CWs differ from horizontal ones mainly by flow direction. They have a greater oxygen transfer rate that is beneficial for nitrification and organic matter removal (Sklarz et al., 2009), and that is leading to a lower surface area required in comparison to HSSF CWs (Herrera-Melián et al., 2018; Lavrnić et al., 2017). Several research studies have recently focused on them.

In Italy, two type of pilot-scale VSSF CWs planted with *Typha latifolia* and *Phragmites australis*, respectively, were evaluated for two years by Morari and Giardini (2009). The systems were both filled with gravel (30-50 mm, 4-8 mm and 8-12 mm size of granular medium) and topped with sand (effective size of 0.16 mm). The mean removal efficiency of parameters tested in two years is shown in Table 4.2 (No.7). It was also found that the systems performed much better in the second experimental year, especially regarding COD (>93%), BOD (>92%), N (>90%) and K (>86%) removal. The authors attributed such a result to macrophytes that were completely established by the second year. The vegetation showed a positive effect during a treatment process by uptake of nutrients, providing a habitat for microbial populations, etc. However, the treatment efficiency did not differ for tested macrophyte species.

In Egypt, Abou-Elela and Hellal (2012) conducted a pilot-scale VSSF CW experiment for two years. There were three types of macrophytes (*Canna*, *Phragmites australis* and *Cyprus papyrus*) in different sections of this wetland unit. The top 60 cm of the bed were filled with 10 mm gravel and the bottom 25 cm with 20 mm gravel. The average removal rates of TSS, BOD and COD were 92%, 90%, 88% (Table 4.2, No. 8), respectively. However, different mean concentrations of pollutants accumulated in roots of *Canna*, *Phragmites australis* and *Cyprus papyrus* also proved different vegetation species could influence the removal rates of some contaminant. It was found that *Cyprus papyrus* increased removal of heavy metals, TN and TP, while *Canna* was better for pathogen reduction.

Fountoulakis et al. (2017) reported on the use of a VSSF system planted with halophytes, namely *Atriplex halimus*, *Juncus acutus* and *Sarcocornia perennis*, for treatment of primary treated domestic wastewater in Heraklion, Greece, and compared it with another VSSF CW planted with *Phragmites australis*. Both beds were filled with a 15 cm depth drainage layer of 20-40 mm gravel, a 10 cm depth transition layer with 8-20 mm gravel and a 55 cm depth main layer of 1-3 mm coarse sand, while HLR was 95 mm d<sup>-1</sup>. The mean removal efficiency of all systems were 78.5% for COD, 26.5% for TN and 30% for TP (Table 4.2, No. 9). The authors indicated that *Atriplex halimus* was better for salt accumulation (especially Na ones), biomass production and pathogen removal. However, there was no significant difference on the removal efficiency of phosphorus and organic matter among CWs planted with halophytes and common reed, except for slightly lower TN removal rate achieved by the system planted with halophytes.

In the UK, Almuktar et al. (2017) adopted a completely randomized design when testing ten different VSSF CWs located at an aerated greenhouse. They were filled with pea gravel (a depth of 60 cm) differing in four parameters (aggregate diameter, loading rate, contact time, resting time). They were operated for more than 4 years (June 2011 to September 2015). The findings indicated that effluent concentrations of TP ( $4.2 \pm 0.48 \text{ mg L}^{-1}$ ), NH<sub>3</sub>-N ( $4.2 \pm 2.64 \text{ mg L}^{-1}$ ), potassium ( $7.0 \pm 3.03 \text{ mg L}^{-1}$ ) and TC ( $69647 \pm 64852.6 \text{ cfu } 100 \text{ mL}^{-1}$ ) were significantly higher than the irrigation limits, despite a good treatment efficiency for other pollutants. However, nutrients from the effluents were recycled for irrigation and lead to greater chilies weights and dimensions, consequently more marketable profits. Furthermore, the best quality fruits came from the chilies irrigated by the systems of small aggregate diameters, long resting and contact time, under high inflow loading rates.

#### 4.3.3. Improved single-stage constructed wetlands

In recent years, there were studies focusing on novel constructed wetlands for wastewater treatment. After some improvements (e.g. location of wetlands, recirculation and aeration), these systems could achieve a greater treatment capacity for some contaminants (e.g. TN, TP) than common single-stage CWs (Mart ínez et al., 2018; Xuan et al., 2009), while also reducing surface area needed in comparison with hybrid CWs (Ramprasad et al., 2017).

For instance, based on HSSF CWs, GROW was first established in the UK. The system was made up of interconnected weirs and troughs functioning as the beds for wastewater treatment. Besides, it was placed on the roof in order to save ground space. Thus it was proposed as a viable alternative to treat greywater, addressing the limitation of land resource (Ramprasad et al., 2017). Similarly, a research in the Netherlands proposed the design of a shallow constructed wetroof (Zapater-Pereyra



et al., 2016). It was also reported that TN (>87%), TP (>86%), TSS (>80%), COD (>79%) and BOD<sub>5</sub> (>95%) were reduced effectively in the tested system at a low organic loading rate, predominantly due to the treatment of roots, organic soil and sand in the system. In India tropical conditions, from November 2013 to April 2015 Ramprasad et al. (2017) monitored a pilot-scale GROW system planted with eight kinds of local common plants and filled with a 15 cm depth layer of mixed substrates, containing sand, brick bats and gravel (1:1:1). The results showed that overall removal rate of all tested parameters was very high, in particular COD (92.5%), BOD (90.8%), TSS (91.6%), TN (91.7%), TP (87.9%) and FC (91.4%) (Table 4.2, No. 11). The author pointed out that the high removal of solids was attributed to the baffled CW configuration, which increased the flow path and improved filtration process. It was also concluded that 3.125 cm day<sup>-1</sup> was the most suitable HLR for organics removal (BOD and COD) in comparison with other tested HLRs. A higher HLR than 3.125 cm day<sup>-1</sup> caused shorter hydraulic retention time (HRT), consequently less removal. Moreover, the system was affected by the factor of seasonal changes, attaining the highest treatment efficiencies of pollutants (i.e. organics, nutrients and FC) in summer, which was consistent with Ayaz (2008).

In Israel, the research related to RVFCW reported that the recirculation was beneficial to organics abatement (Sklarz et al., 2009). In this study, Sklarz et al. (2009) tested two RVFCW systems with or without soil-plant component (a layer of peat vegetated with *Juncus alpigenus* and *Cyperus haspan* for a depth of 8 cm) and revealed that even in the recirculating system without soil-plant component, organics from wastewater can be removed with a high efficiency, namely 95% for BOD<sub>5</sub> and 84% COD on average, besides a 90% TSS removal (Table 4.2, No. 12). Similarly, in another 40-day study (Travis et al., 2010) on a lab-scale RVFCW planted with *Hydrocotyle leucocephala* and *Cyperus papyrus* and fed with domestic greywater, it was observed that TSS decreased by around 95% and BOD<sub>5</sub> by about 99% (Table 4.2, No. 13).

In addition, artificial aeration and innovative media were introduced into some CW systems to address a lack of oxygen and the unfavorable adsorption rates of conventional media. Andreo-Martínez et al. (2017) improved a HSSF wetland in Spain by filling it with blast furnace slags (BFS) and feeding it with artificially aerated municipal sewage. Those changes produced higher quality effluents. The data demonstrated that the application of BFS and aeration optimized TP removal. Moreover, the average removal efficiency of turbidity ( $99.5 \pm 0.3\%$ ), TSS ( $97.5 \pm 1.3\%$ ) and TN ( $91.5 \pm 5.3\%$ ) had also been improved due to the aeration (Table 4.2, No. 6). Sklarz et al. (2009) also pointed out the favorable effect of passive aeration on organics removal in their research. Additionally, Xuan et al. (2009) introduced green sorption media (recycled and natural

materials) into upflow subsurface CWs and tested them for three months. It was found that TP and TN were effectively reduced by 94.9% and 75.4% (Table 4.2, No. 14) in the CWs and reduced by 95.8% and 81.3% in the integrated system with the combination of a septic tank and CWs, respectively. It was verified the tested CWs had a good capacity for TN and TP abatement, especially for TP, which was hardly affected by the septic tank.

Table 4.2 shows the mean removal efficiency based on concentrations of investigated systems coming from previously discussed 14 single-stage CW studies. Actually, the removal efficiency of pollutants can be assessed by two methods - using concentrations or mass loads for calculation. Values attained from the two methods were similar when ET was low, while obviously different under very high ET in summer, as Tuttolomondo et al. (2016) stated in their study. High ET losses caused unsatisfactory residual concentration of pollutants, as they concentrated the elements in effluents, offsetting the effect of treatment (Morari and Giardini, 2009). Therefore, removal efficiency based on mass loads is considered to be more accurate (Tuttolomondo et al., 2016). However, most researchers provide removals based on concentrations in their studies due to easier measurement of related parameters.

Among all the systems considered (Table 4.2), improved systems GROW and RVFCW displayed superior overall removal efficiency, beyond 90% for the most of contaminants measured (Table 4.2, Nos. 11-13). According to Table 4.2, the majority of VSSF and HSSF CWs reached a good effectiveness of removing solids. Generally, VSSF systems (Table 4.2, Nos. 7-10) showed better effect on organic matter removal in comparison with HSSF ones (Table 4.2, Nos. 1-5). One exception was the improved HSSF system (Table 4.2, No. 6), that showed a greater capacity of nutrients removal comparing with other HSSF and VSSF CWs. Regarding bacterial indicators, the HSSF systems No. 2, 4 and the VSSF system No. 8 (Table 4.2) removed them most effectively. Their abatement higher than 94% could be related to different factors: i) longer HRT (Ayaz, 2008), ii) high temperature and oxygen concentration in the VSSF CW, providing aerobic environment unfavorable for coliforms survival (Abou-Elela and Hellal, 2012). It was revealed that organics and solids removal were also high in the system No. 8 (Table 4.2). Settleable organics could be eliminated through filtration and deposition, while the high removal of organic compounds can be explained by the fact that this system provided both oxygen and a more favorable habitat for microorganisms due to the presence of various plants. Moreover, the diversity of roots increased HRT, beneficial for pollutants removal (Abou-Elela and Hellal, 2012).

The role of plants was reported in several studies. Plant types (i.e. macrophytes and halophytes) affected pollutant removal rates (Fountoulakis et al., 2017). Some research stated the positive effect

of plants (Morari and Giardini, 2009; Toscano et al., 2015). In contrast, plants can also result in the pollutant concentrations increase by the generation of organic carbon (Ayaz, 2008) and the higher ET values (Toscano et al., 2015). Furthermore, Morari and Giardini (2009) highlighted the macrophytes did not show their full capacity by the time they were mature.

Seasonal variations affect the removal capacity of systems. It is found that higher removal efficiencies of organics, microbes (e.g. *E. coli*, FC and TC) and nutrients can be achieved in summer (Ayaz, 2008; Morató et al., 2014; Ramprasad et al., 2017).

Interestingly, the water depth also plays a role in removal performance. Shallow CWs (e.g. GROW, constructed wetroof), providing high aerobic conditions for pollutants and a larger fraction of water volume in contact with plants roots, were favorable for enhancing removal efficiencies in comparison with conventional CWs (Morató et al., 2014; Zapater-Pereyra et al., 2016).

The continuous recirculation can also improve organics abatement (Sklarz et al., 2009), while the aeration can help reducing the values of nutrients, turbidity, TSS and organics (Andreo-Martínez et al., 2017; Sklarz et al., 2009). The system filled with a smaller size medium performed better in microbial removals than the one filled with a bigger size medium (Morató et al., 2014).

Table 4.2 - The mean removal efficiency based on concentrations of main pollutants in single-stage systems analyzed.

System No.	HLR (cm d <sup>-1</sup> )	Removal efficiency (%)							
		Organic matter		Nutrients		Solids	Bacterial indicators		
		COD	BOD <sub>5</sub>	TN	TP	TSS	<i>E. coli</i>	FC	TC
1	12	60.5	53.8	-	-	-	-	-	-
2	36	63	-	61	-	88	99.9	-	-
3	3.6	63.8	65	-	-	-	-	-	-
4	-	50	65	-	-	-	-	>94	>94
5	0.6	-	-	51.3	72.7	-	-	-	-
6	2.62	92.7	97.8	91.5	96.9	97.5	-	-	-
7	1.8-4	79	76.5	-	62.8	59.3	-	-	-
8	-	88	90	-	-	92	94-99.9	94-99.9	94-99.9
9	9.5	78.5	-	26.5	30	-	-	-	-
10 <sup>a</sup>	-	-	-	-	-	-	-	-	-
11	1.94-3.75	92.5	90.8	91.7	87.9	91.6	-	91.4	-
12	-	84	95	-	-	90	-	-	-
13	-	-	99	-	-	95	-	-	-
14	-	-	-	75.4	94.9	-	-	-	-

<sup>a</sup> The research did not provide either removal efficiencies of main pollutants or influent concentrations, so the efficiency values could not be calculate.

## 4.4. Hybrid systems

In the expectation of strengthening the treatment capacity of pollutants and attaining effluents of higher quality, some researchers combined different CW types or CWs with other technologies into hybrid systems, making use of their respective advantages (Ávila et al., 2015; Haghshenas-Adarmanabadi et al., 2016; Ramprasad and Philip, 2018; Zurita and White, 2014).

### 4.4.1. Hybrid constructed wetlands

The most widely used hybrid CW system is the combination of two subsurface flow CWs - horizontal and vertical one (Ramprasad et al., 2017). For example, in South Korea, Kim et al. (2016) carried out a full-scale experiment on VSSF-HSSF CWs from 2002 to 2013. The VSSF system was planted with *Phragmites australis* and *Phragmites japonica*, while the HSSF CW was planted with *Miscanthus sacchariflorus*, *Carex dispalata*, *Juncus effuses* and *Iris pseudacorus*. Both of them were filled with coarse sand. The study revealed that the hybrid system had a stable TN removal in the whole 12-year operation period, with the average removal rate of 71.8% (Table 4.3, No. 15). TN removal efficiency was associated with the factors like season (greater removal rates in summer), operating stage (greater removal rates during the middle stage of operation, between 2006 and 2009) and nitrogen load. The highest removal rate simulated was at the inflow nitrogen load of under  $2.8 \text{ g m}^{-2} \text{ day}^{-1}$  in summer. Besides, the VSSF had a better removal efficiency of TN than the HSSF. It can be explained by the fact that HSSF cannot satisfy the needs of denitrification/nitrification for a complete anaerobic/anoxic condition, while VSSF can provide a aerobic condition that favors nitrification process.

Herrera-Melián et al. (2010) tested two pilot VSSF-HSSF systems filled with different substrates (gravel and lapilli) for eight months in Spain. Both systems were effective in municipal wastewater treatment, achieving the removal efficiency of more than 86%, 78%, 84%, 95%, 96% and 98.7% for BOD<sub>5</sub>, COD, NH<sub>4</sub><sup>+</sup>-N, SS, turbidity and fecal indicators, respectively. The best COD and FC removal was attained by gravel-based system and lapilli-based system, respectively, at a high HLR of  $7.9 \text{ cm d}^{-1}$ , while the best BOD<sub>5</sub> removal was from lapilli-based system, at low HLRs in the range of  $3.7\text{-}4.1 \text{ cm d}^{-1}$ . Pollutants (COD, FC and NH<sub>4</sub><sup>+</sup>-N) removal efficiencies were not significantly different between two hybrids with gravel and lapilli when HLRs varied. However, for BOD<sub>5</sub>, the lapilli-based system achieved significantly higher removal than gravel-based one. This better performance on BOD<sub>5</sub> removal could be attributed to a higher porosity and smaller diameter of lapilli particles applied, thus leading to greater removal during the treatment processes (e.g.

filtration, sedimentation and BOD<sub>5</sub> degradation). Hence, the marginally greater system is lapilli-based hybrid CWs at high HLR, with the removal of 82% for COD, 89 % for BOD<sub>5</sub> and 99.9% for FC (Table 4.3, No. 16).

In Iran, Haghshenas-Adarmanabadi et al. (2016) evaluated four pilot HSSF-VSSF CWs from September 2013 to August 2014, three of them planted with different vegetation types and the last one unplanted. The average HLR was 5.3 cm d<sup>-1</sup>. It was found that the hybrid systems can abate the main contaminants with high removal rates. The best removal efficiencies belonged to *Phragmites* hybrid CW, 84% for BOD<sub>5</sub>, 79% for COD, 78% for TSS, 99% for FC, 43-97% for TP. (Table 4.3, No. 17). However, no significant difference was found among various hybrid systems for those pollutants removal, except for TP removal. Planted CWs performed significantly better than unplanted CW for TP reduction. It can be attributed to plants uptake and sequestration in microbial biomass. Furthermore, adding VSSF CWs after the HSSF CWs was highly effective for optimizing main pollutants removal except for nitrates, which can be produced by ammonium nitrification in the VSSF stages.

In Colombia, Garc á et al. (2013) reported the high performance of a series of two-stage CWs in pathogen removal. These systems differed by feeding modes, order or combination of CWs (i.e. VSSF, HSSF) and vegetation conditions. The detailed description can be found in Table 4.1 (No. 18). It was shown that the type of planted VSSF-HSSF combination can reduce 99.984% *E. coli*, 99.987% TC and 90.741% Helminth eggs. The system had the best nitrogen removal (>90%) of all, also providing high removals of organics and solids (>90% for both BOD<sub>5</sub> and COD, >85% for TSS), regarded as the system with the best overall performance (Table 4.3, No. 18).

In Spain, Herrera-Meli án et al. (2018) tested two hybrid systems, i) mulch-based HSSF followed by gravel-based VSSFs, ii) mulch-based HSSF followed by mulch-based VSSFs, under different feeding modes (continuous and intermittent). The highest removals of various pollutants (82% for COD, 97% for BOD<sub>5</sub>, 99% for TSS, 99.8% for FC) were provided by the second hybrid system, the combination of HSSF and mulch-based VSSFs, under the continuous feeding mode (Table 4.3, No. 20). The authors argued that the intermittent feeding mode could have caused a shorter HRT compared to the continuous one, thus the lower removal efficiencies were attained. Furthermore, the greater performance of VSSFs with mulch than the ones with gravel can be attributed to its characteristics (e.g. compressibility and small particle size). A small particle size provides several benefits, longer HRT, greater water distribution on the reactor surface and retention of pollutants (e.g. TSS, turbidity).

In USA, Jokerst et al. (2011) operated a pilot-scale FWS-SSF hybrid CW in a semi-arid, temperate

climate for one year. The FWS and SSF beds were planted with *Typha latifolia* and *Scirpus acutus*, respectively. The FWS CW was filled with amended soil up to a depth of 0.9 m, a mixture of 50% sandy-loam soil and 50% sphagnum peat, while the SSF CW was filled with about 15 mm-diameter granite stone. The author reported the seasonal performance of the system. It was shown that during non-winter periods the removal efficiency based on mass loads of contaminants BOD<sub>5</sub>, TN and TP was as high as 92%, 85% and 78% on average, respectively. However, the treatment effect seemed to decrease in winter season. The mean yearly removal efficiency based on concentrations is given in Table 4.3 (Table 4.3, No. 19). In addition, TSS removal was probably negatively affected by a considerable number of growing algae during warm months of spring in the FWS bed.

In recent years, multistage hybrid CWs have been applied. In China, He et al. (2018) tested for nine months a system consisting of a down-flow VSSF CW, an up-flow VSSF CW and a HSSF CW. The highest intensity of nitrification and denitrification of the media was in the down-flow VSSF bed and the HSSF bed, respectively, which may be associated with the abundance of nitrifying and denitrifying bacteria in these wetlands. During the experimental period the removal rates of COD, TP, TN and NH<sub>4</sub><sup>+</sup>-N reached 59%, 82.8%, 57.7% and 79.2% on average, respectively (Table 4.3, No. 21). COD removal was negatively affected by the application of media with relatively big diameters. Since the media could not have provided sufficient surface area for biofilm growth, microbial activities were limited and unsatisfactory COD removal efficiency was obtained. Despite this, the media decreased the possibility of clogging in the system.

In Czech Republic, a three-stage hybrid CW comprised of a saturated VSSF CW, a free-drain VSSF CW and a HSSF CW, was investigated by Vymazal and Kröpfelová (2015) for nineteen months. They reported good efficiencies of the hybrid system - 92.5% for BOD<sub>5</sub>, 96% for TSS, 88.8% for NH<sub>4</sub>-N, 83.8% for COD and 79.9% for TN (Table 4.3, No. 22). A similar removal robustness of a lab-scale three-stage hybrid system (two VSSF CWs operating alternatively, a HSSF CW and a FWS CW in series) located in Spain has been reported by Ávila et al. (2013). The removal efficiencies of that system were 91% for BOD<sub>5</sub>, 97% for TSS, 94% for NH<sub>4</sub>-N, 78% for COD and 46% for TN. A full-scale research (Ávila et al., 2015) also demonstrated the superb overall pollutants abatement capacity of hybrid systems (Table 4.3, No. 23), even for some emerging pollutants (more than 80% removal, e.g. analgesic and anti-inflammatory drugs, personal care products). These high removals of different pollutants in hybrid system were aided by high temperatures and synergies and combination of removal mechanisms (e.g. nitrification, denitrification, biodegradation and sorption) under different physicochemical conditions of CW configurations (Ávila et al., 2015; Vymazal and Kröpfelová 2015).

#### 4.4.2. CWs combined with additional technologies

Some investigations have concentrated on the addition of different technologies to existing CWs, such as filtration, ultraviolet (UV) treatment, etc. These physical, chemical or biological technologies were applied to improve wastewater treatment, especially for some pollutants like turbidity or microbial indicators (Patil and Munavalli, 2016; Russo et al., 2019a; Toscano et al., 2013).

In Sakharale, India, a tropical zone, Patil and Munavalli (2016) reported a three-stage treatment system, containing preliminary treatment, HSSF CW treatment and a post treatment. Pretreatment was done by a settling cum equalization tank and an up-flow down-flow filter (UFDF), and could significantly decrease turbidity while also partially removing COD. The post treatment included a vertical flow charcoal filter and water hyacinth system, improving further the quality of final effluents to fulfill the outflow requirements mainly attributed to the role of adsorption and plant uptake. The overall removal rates of the system achieved were 70% for COD, 70% for total kjeldahl nitrogen and 85% for pathogen (Table 4.3, No. 24).

In Heilongjiang, China, Gao and Hu (2012) combined bio-contact oxidation (BCO) technology with a greenhouse-structured HSSF CW. The utilization of double solar panels in the greenhouse provided a stable temperature for treating wastewater, and improved effectively pollutants removal rates especially during winter season. The overall removal efficiency of the parameters evaluated in the combined system was 85.01% (COD), 70.98% (NH<sub>3</sub>-N), 36.48% (TP) (Table 4.3, No. 25). The BCO treatment was responsible for 74.6% and 85.4% of overall removal of COD and NH<sub>3</sub>-N, respectively while the wetland contributed to 59% of overall TP removal.

In Ulaanbaatar, Mongolia, an 'ice-block unit' consisting of a storage tank, septic tanks, a VSSF CW and a collecting tank showed a great potential for application in cold climates. It stored greywater throughout freezing period in an ice-block, melting and treating it during warm months. The removal rates of main pollutants (e.g. COD, NH<sub>4</sub><sup>+</sup>, TSS, *E. coli*) ranged from 87% to 100% (Uddin et al., 2016) (Table 4.3, No. 26).

In Italy, Russo et al. (2019a) tested for one year a full-scale HSSF CW combined with an UV unit to treat domestic wastewater. The findings indicated that the whole system eliminated thoroughly microbial indicators such as *E. coli*, somatic coliphages and *C. perfringens* spores (Table 4.3, No. 27b). Sklarz et al. (2013) proved the effectiveness of UV light disinfection treatment through agricultural reuse experiments, since there was no *E. coli* detected in the soil irrigated with the treated wastewater. Moreover, wastewater treated after the integrated operation of the sedimentation tank, RVFCW, the filter and the UV disinfection unit caused a large decrease on concentration of



BOD<sub>5</sub>, COD, TSS and *E. coli* in comparison with the raw wastewater (Table 4.3, No. 28). Horn et al. (2014) concluded that the combination of photocatalytic ozonation (UV/TiO<sub>2</sub>/O<sub>3</sub>) technologies and SSF CWs was capable of improving disinfection efficacy of the system and removed effectively microbial contaminants, reducing the microbial load under the detection limit. Besides, it eliminated 88.7% of BOD<sub>5</sub>, 62.1% of COD and 63.4% of TP (Table 4.3, No. 29). However, the ramp of UV/TiO<sub>2</sub>/O<sub>3</sub> reactor was observed to saturate after running for 4 h as a result of physisorption and chemisorption.

In Turkey, Ayaz et al. (2015) reported a three-stage hybrid pilot system built in a small community, consisting of anaerobic pretreatment, a HSSF CW and a VSSF CW. The pretreatment, an ABR and an upflow anaerobic sludge bed (UASB) reactor running in parallel, removed partially organic matter and SS. The combined system removed 90% of nitrogen and 95% of organic matter on average and the mean removal rates of pollutants BOD<sub>5</sub>, COD and TSS are shown in Table 4.3 (Table 4.3, No. 30). The authors stated that the combination of HSSF-VSSF CWs optimized the removal of organics and SS, aided denitrification process of HSSF, effectively reduced phosphorus and stimulated nitrification in the VSSF, and in general showed greater performance than a single CW. Another way to increase TN removal is the recirculation of effluents in the system, which can also contribute to organics reduction (Sklarz et al., 2009).

Likewise, the study provided by Ávila et al. (2016) proved that an experimental integrated system containing an anaerobic reactor was a good alternative for wastewater treatment in small communities, particularly in warm zones. The system comprised of two alternating VSSF CWs, a HSSF CW and a FWS CW operating in series, following an anaerobic reactor (a HUSB reactor). The results showed the system was able to effectively reduce BOD<sub>5</sub> (93% removal), TSS (96% removal), COD (82% removal) and NH<sub>4</sub>-N (75% removal), whereas it did not perform well in PO<sub>4</sub>-P (11%) and SO<sub>4</sub><sup>2-</sup> (10%) removal (Table 4.3, No. 31). Another application of a HUSB reactor reported in Spain showed that it did not perform as well as a conventional settler before HSSF CWs (Pedescoll et al., 2011).

Yeh et al. (2010) carried out an experiment on the system made up of an oxidation pond, two FWS CWs with a cascade between them and a SSF CW operating in series. The findings (Table 4.3, No. 32) showed the system removed 81% of BOD and 48% of COD on average, mainly owing to microbial degradation, and reduced 65% of TN, primarily attributed to nitrification and denitrification occurring in the treatment processes.

Reinoso et al. (2008) operated a facultative pond (FP) followed by a FWS and a SSF system for 10 months in Spain and the results revealed that FP was the most effective in bacterial removal (e.g.

TC, *E. coli*), while the SSF CW had the most robust capacity for removing protozoan pathogens and coliphages among the three technologies tested. It was observed that 99.33% of *E. coli* and 97.12% of TC were removed on average (Table 4.3, No. 33).

In Egypt, Abdel-Shafy et al. (2017) tested a pilot system including a sedimentation tank, a HSSF CW and a VSSF CW running in series for treating blackwater. The hybrid system was proven to be capable of treating a high hydraulic and organic load, with the removal efficiency of 98.5%, 98%, 97.4%, for COD, BOD and TSS, respectively (Table 4.3, No. 34). Furthermore, it was found that a high surface area and a low velocity of the integrated system (HSSF and VSSF CWs) principally contributed to the improvement of wastewater quality. However, a disadvantage of the system was exactly its high surface requirement.

In Greece, a three-stage hybrid system consisting of two serial settling tanks, a VSSF CW and a zeolite tank, was operated for 40 months. The overall removal efficiencies were satisfactory for BOD (95.8%), COD (94.9%), TSS (96%) and TC (99.97%) (Table 4.3, No. 38). It was observed that organics removal was mostly attributed to the septic tanks followed by the CW. Finally, the zeolite tank further enhanced the treatment performance, since relatively large pores of zeolite were favorable for the adsorption of organics. Besides, the superb removal of TC was achieved mainly by sedimentation and filtration in the septic tanks and the CW, respectively. It was also reported that plants played a significant role on removal organics and nutrients (except for TSS and TC). Moreover, their growth and movement may be beneficial for preventing clogging (Gikas and Tsihrintzis, 2012). Similarly, delaying or preventing system clogging can also be achieved by the application of media with bigger sizes (He et al., 2018), the setup of pretreatment (e.g. a HUSB reactor, a UFDF sand filter), adjustment of treatment operation and systems structure (e.g. intermittent discharge, upflow structure of CWs) (Pedescoll et al., 2011; Vera et al., 2013), as well as the application of earthworms or a low organic loading rate (Lavrnić et al., 2019; Zapater-Pereyra et al., 2016).

Vera et al. (2013) reported a system composed of a septic tank and an Imhoff tank in series as pretreatment, and two parallel VSSFs followed by a HSSF as secondary treatment. It was found that the overall removal rates were 98% for TSS, 93% for BOD<sub>5</sub>, 89% for COD, 61% for TN and 47% for TP (Table 4.3, No.39). Despite variability in removal efficiency among the stages affected by different factors (e.g. seasonal change and influent quality), overall performance was relatively stable during the experimental period of 2 years.

#### 4.4.3. Comparison of various hybrid systems

As reported in section 4.4.1, Haghshenas-Adarmanabadi et al. (2016) tested four pilot HSSF-VSSF

hybrid CWs for one year, three units planted with different emergent vegetation (*Phragmites australis*, *Typha latifolia* and *Arundo donax*) and one left unplanted, with a mean HLR of 5.3 cm d<sup>-1</sup>. The authors pointed out that the hybrid systems offered better conditions for reaching reuse standards than single CWs. Besides, there were no significant differences among four systems in removal capability of BOD<sub>5</sub>, COD, TSS and coliforms. However, with regards to nutrient removal, the planted systems (especially the unit planted with *Phragmites australis*) performed better in comparison with the unplanted one.

As reported in section 4.4.1, García et al. (2013) assessed the performance of six planted or unplanted two-stage hybrid systems consisting of HSSF or/and VSSF CWs. The results highlighted that the combination VSSF-HSSF was the most efficient for *E. coli* removal (4 log units), TC (3 log units) and nitrogen (>90%), whether planted or not. In addition, it statistically displayed that vegetation probably contributed a lot to the reduction of nutrients and *E. coli*.

In Mexico, Zurita and Carreón-Álvarez (2015) reported on the application of three integrated systems running for two years, namely HSSF+stabilization pond (SP), HSSF+VSSF and VSSF+HSSF systems. During the first year, it was observed the HSSF+VSSF and VSSF+HSSF system showed the best efficacy of TC removal (2.2 log units) and *E. coli* removal (3.8 log units), respectively. During the second year, both HSSF+VSSF and VSSF+HSSF, performed well in TC and *E. coli* removal that was in the range 2.34-2.44, 3.44-3.74 log units, significantly better than HSSF+SP system. As a result, HSSF+VSSF system was the most effective for *E. coli* (99.94%) and TC (99.5%) removal during the two years study (Table 4.3, No. 35).

Multistage hybrid systems were also utilized in some research. For instance, Licciardello et al. (2018) evaluated two systems with similar costs, whose primary difference was whether UV disinfection was utilized or not. One system was comprised of a HSSF CW, a biological pond, a storage reservoir followed by sand and disk filters, while the other one was made up of a HSSF CW, sand and disk filters and UV treatment, both of them operating in series. It was found that the system including UV treatment was more effective. The removal efficiencies achieved were 76.48%, 80.43% and 90.87% for COD, BOD<sub>5</sub> and TSS, respectively. Moreover, *E. coli* was reduced by 5.49 log units, mostly owing to the UV disinfection (Table 4.3, No. 36b).

According to Ali et al. (2018), the two full-scale systems tested in Pakistan, system I made up of an ABR, a saturated VSSF CW and a FWS CW and system II consisting of ABR, a HSSF CW and a FWS CW were both influenced by seasonal factors. They achieved greater removal efficiency in summer, similar with several findings concerning single or hybrid CWs (Ayaz, 2008; Kim et al., 2016; Ramprasad et al., 2017). Generally, the system I provided higher overall removals of COD

(73.6%), BOD<sub>5</sub> (76.2%) and NH<sub>4</sub>-N (52.8%). The exception was TSS removal (82%), that was slightly lower than in the system II (91%) (Table 4.3, No. 37). The differences of removal efficiency between the two systems were mainly caused by the use of HSSF or VSSF as the secondary treatment. It was also found that the first stage treatment (ABR) of both systems predominantly contributed to solids removal and organics degradation.

Table 4.3 shows the overall pollutants removal efficiency of hybrid systems from 25 studies previously discussed. These integrated systems showed nearly complete removal of bacterial indicators and quite a good effectiveness in TSS abatement (>85% for most systems), except for the system HSSF-lagooning (Table 4.3, No. 27a). In comparison with single-stage CWs (Table 4.2), they had a better overall performance in organics reduction. These superb removal rates can be explained by the combination of technologies in hybrid systems that contributed to a greater overall removal efficiency. However, regarding nutrients, the hybrid systems displayed a wide range of removal variation, from 29.5% (Table 4.3, No. 28) to 94% (Table 4.3, No. 23) for TN and from 7% (Table 4.3, No. 28) to 82.8% (Table 4.3, No. 21) for TP. Low TP removal observed in a few studies could be attributed to a limited sorption of some substrates applied (e.g. crushed rock), and to the fact that no additional measures for enhancing TP removal were used (Vymazal and Kröpfelová 2015). Interestingly, there is one system HSSF-lagooning (Table 4.3, No. 27a) that showed low removal efficiencies for the most pollutants, although a majority of hybrid systems performed well. It can be attributed to the fact that the algae growth and decomposition in the lagooning unit contributed to the increase of TSS, BOD<sub>5</sub> and COD concentrations. Besides, the lagooning unit was not able to further reduce nutrients (TN and TP), due to the anaerobic decomposition of algae.

The same as section 4.3, the research discussed in this section also pointed out the role of plants, substrates, seasonal variation in hybrid systems. The presence of plants can improve the removal efficiencies of organics and nutrients (García et al., 2013; Gikas and Tsihrintzis, 2012), while the reduction of TSS could be negatively affected by a large number of algae (Jokerst et al., 2011). Regarding *E. coli* removal, the influence of plants was not consistent. The role of plants is still an open question and while García et al. (2013) reported there was significant difference between planted and unplanted systems tested, Headley et al. (2013) stated the opposite results.

In terms of substrates, the application of lapilli (Herrera-Melián et al., 2010), mulch (Herrera-Melián et al., 2018) and zeolite (Gikas and Tsihrintzis, 2012), was regarded as favorable for pollutants removal, due to their characteristics of a high porosity, small media sizes and good compressibility. He et al. (2018) indicated that the media with bigger sizes cannot provide enough surface area for microbial activities, thus resulting in the lower COD removal.

In addition, the seasonal variation also had an impact on treatment performance of the hybrid systems. Jokerst et al. (2011) highlighted the better removal efficiencies of BOD<sub>5</sub>, TN and TP during the warmer part of the year. Particularly, the greatest performance can be generally achieved in summer (Ali et al., 2018; Kim et al., 2016).

Moreover, in comparison to single-stage systems, the use of UV treatment can enhance the disinfection efficacy of hybrid systems, as a result of excellent microbial removals.

Table 4.3 - The mean overall removal efficiency based on concentrations of main pollutants in hybrid systems analyzed.

System No.	HLR (cm d <sup>-1</sup> )	Removal efficiency (%)							
		Organic matter		Nutrients		Solids	Bacterial indicators		
		COD	BOD <sub>5</sub>	TN	TP	TSS	<i>E. coli</i>	FC	TC
15	4.5-22.7	-	-	71.8	-	-	-	-	-
16	3.7-7.9	82	89	-	-	-	-	99.9	-
17	5.3	79	84	-	43-97	78	-	99	-
18	10	>90	>90	>90	-	>85	99.984	-	99.987
19	1.3-3.4	-	80.5	74.8	66.3	-	-	-	-
20	-	82	97	-	-	99	-	99.8	-
21	15-24	59	-	57.7	82.8	-	-	-	-
22	3.8-61.1	83.8	92.5	79.9	30	96	-	-	-
23	4.4	89	99	94	47	98	99.999	-	-
24	1.5-9.3	70	-	-	-	-	-	-	-
25	-	85	-	-	36.5	-	-	-	-
26	-	98-100	-	-	-	97	98	-	-
27a	-	28.26	19.23	50.93	11.86	22	99.937	-	99.899
27b	-	-	-	-	-	-	>99.99	-	99.996
28	-	85.5	96.8	29.5	7	90	99.999	-	-
29	-	62.1	88.7	-	63.4	-	-	-	-
30	11.1-21.9	94.3	91.9	-	-	97	-	-	-
31	27	82	93	-	-	96	-	-	-

		COD	BOD <sub>5</sub>	TN	TP	TSS	<i>E. coli</i>	FC	TC
32	-	48	81	65	-	-	-	-	-
33	-	-	-	-	-	-	99.33	-	97.12
34	-	98.5	98	-	-	97.4	-	-	-
35	6.8-14.5	-	-	-	-	-	99.94	-	99.5
36a	-	69.02	68.01	-	-	90.15	99.99	-	-
36b	-	76.48	80.43	-	-	90.87	99.999	-	-
37	-	73.6	76.2	-	-	82	-	-	-
38	-	94.9	95.8	-	67.3	96	-	-	99.97
39	-	89	93	61	47	98	-	-	-

When the research tested more than one hybrid system (No. 16, 17, 18, 20, 35 and 37), the one with the best overall removal efficiencies was reported in this table.

The study (No. 26) provided only the maximum removal rates.

## 4.5. Wastewater reuse in agriculture

Wastewater reuse can not only alleviate water scarcity, but it can also relieve pressure on conventional wastewater treatment plants (Ghaitidak and Yadav, 2013). Treated wastewater offers a more sustainable and stable use in comparison with natural resources, especially when seasons and climate change are considered (Zhang and Shen, 2019). Moreover, treated wastewater is rich in inorganic elements and organic compounds that can increase crop yields, while at the same time reducing use of fertilizers (Castro et al., 2011). Similar findings were also made by Almukhtar et al. (2017) that recycled nutrients from wastewater in agriculture, resulted in greater chilies weights and dimensions, as discussed in section 4.3.2.

Nevertheless, wastewater could negatively impact irrigated soil and plants, likely to be a potential threat on the environment and human health. Travis et al. (2010) presented hydrophobicity development of soil as a result of the application of raw wastewater in irrigation. Moreover, it can cause heavy metals accumulation as reported by Gola et al. (2016) and shallow groundwater pollution (Zhang and Shen, 2019). However, the use of suitable wastewater treatment before irrigation can effectively prevent modification of soil properties and diminish environmental risks (Sklarz et al., 2013). Further information on the effect that wastewater reuse can have on soil and crops can be found in Al-Isawi et al. (2016); Almukhtar et al. (2017) and Sklarz et al. (2013).

#### 4.5.1. EU standards on wastewater reuse for agricultural purposes

Currently, more and more attention is given to the issues of wastewater treatment and reuse. Treated wastewater is reused for crop irrigation in many areas of the world (Licata et al., 2017). Specific standards about use of reclaimed water in agriculture already exist, including those in developed countries (e.g. United States, Italy, Spain), developing countries (e.g. Egypt, Pakistan, Turkey, Iran, Mexico, Thailand, Colombia) and even some international institutions such as the World Health Organization. The European Commission is currently in the process of adopting its own guidelines (European Commission, 2018), that are characterized by detailed classification limits depending on crop types and irrigation methods (Table 4.4).

Table 4.5 shows main pollutants concentration in effluents from 39 studies analyzed in this review. According to the future European criteria, parameters BOD<sub>5</sub>, TSS and *E. coli* are classified into different levels that correspond to different agricultural purposes. It can be seen that a majority of studies presented here achieved Class B, C and D (25 mg L<sup>-1</sup>) for BOD<sub>5</sub> concentration in effluents, suitable for crops irrigation (e.g. industrial, energy, and seeded crops, processed food crops and non-food crops) under any method (Table 4.4). Besides, the better BOD<sub>5</sub> removals were exhibited by the improved single-stage CWs - GROW and RVFCW (Table 4.5, No. 11, 13) and several multistage hybrid systems (Table 4.5, No. 22, 23, 28, 32, 36b), consequently meeting the strictest standard of irrigation reuse (Class A). As expected, most

treatment systems showed a good performance on TSS removal leading to low effluent concentrations, except for two hybrid systems (Table 4.5, No. 27a, 37) exceeding the range of the guideline. The high TSS concentration in research No. 27a can be attributed to algae growth in the lagooning unit. The similar finding was also observed by Jokerst et al. (2011) as reported in section 4.4.1. Regarding *E. coli*, four studies (Table 4.5, No. 6, 27b, 33, 36b) meet Class A (10 cfu 100 mL<sup>-1</sup>) of the irrigation limits. Those results can be mainly explained by the positive effects of artificial aeration (Andreo-Martínez et al., 2017), UV treatment (Licciardello et al., 2018; Russo et al., 2019a) and the combination of different treatment systems (Reinoso et al., 2008). Moreover, it is found that not many studies can achieve excellent *E. coli* removal without the application of disinfection measures or other chemical agents (Andreo-Martínez et al., 2017). Thus, several researchers indicated that, in order to improve effluents quality and to reach the criteria recommended for agricultural reuse, at least two stages of wastewater treatment in hybrid systems are necessary for pathogen removals (Toscano et al., 2015; Zurita and White, 2014). In terms of COD and TN, effluent concentrations among various systems vary considerably (Table 4.5). As concluded in section 4.3.3, artificial aeration and recirculation treatment can improve the removal of

nutrients and organics, respectively. Furthermore, as reported in section 4.4.3, plants can also be beneficial for both pollutants removal.

#### 4.5.2. Irrigation applications of treated wastewater

Among the 39 experimental research selected, there are several papers focusing on reclaimed wastewater reuse in irrigation, besides treatment methodologies. Some researchers (Almuktar et al., 2017; Almuktar and Scholz, 2015) have carried out lab-scale experiments on an overall process of wastewater treatment and reuse in irrigation for a few years. The findings indicated that chilies were able to grow successfully if irrigated with effluents from a VSSF CW, that for the majority of parameters complied with the irrigation criteria. Sklarz et al. (2013) explored the influence on soil of using treated wastewater from RVFCW for irrigation, and concluded that after 3 years physical and chemical properties of soil were similar to a soil undergoing usual agricultural treatment (irrigated with fresh water and enriched with fertilizers). Travis et al. (2010) also stated that the RVFCW effluents did not have any obvious adverse impact on plants growth and soil, thus it could be considered as an effective irrigation source.

Table 4.4 - The European guidelines on pollutants threshold values of reclaimed water for agricultural irrigation (European Commission, 2018).

Pollutants	Reclaimed water quality class			
	Class A	Class B	Class C	Class D
<i>E. coli</i> (cfu 100 mL <sup>-1</sup> )	10	100	1000	10000
BOD <sub>5</sub> (mg L <sup>-1</sup> )	10	25	25	25
TSS (mg L <sup>-1</sup> )	10	35	35	35

**Class A:** All food crops, including root crops consumed raw and food crops where the edible portion is in direct contact with reclaimed water. Irrigation method: All irrigation methods allowed.

**Class B:** Food crops consumed raw where the edible portion is produced above ground and is not in direct contact with reclaimed water, processed food crops, non-food crops including crops to feed milk- or meat-producing animals. Irrigation method: All irrigation methods allowed.

**Class C:** Crop category applicable is the same as Class B. Irrigation method: Drip irrigation only.

**Class D:** Industrial, energy, and seeded crops. Irrigation method: All irrigation methods allowed.



Table 4.5 - The mean effluent concentration of tested main pollutants of analyzed systems and the class level for agricultural purpose of treated wastewater according to parameters BOD<sub>5</sub>, TSS and *E. coli* referring to the guidelines of European Commission (2018).

System No.	Parameters required by European Commission (2018)						Other parameters		
Single-stage CW	<i>E. coli</i> (cfu 100 mL <sup>-1</sup> unless stated otherwise)	Class	BOD <sub>5</sub> (mg L <sup>-1</sup> )	Class	TSS (mg L <sup>-1</sup> )	Class	COD (mg L <sup>-1</sup> )	TN (mg L <sup>-1</sup> )	TP (mg L <sup>-1</sup> )
1	-	-	12.2	B, C, D	-	-	21.1	-	-
2	-	-	-	-	7.4	A	28.6	10.8	-
3	-	-	49	None	-	-	61.5	-	-
4	-	-	3.9	A	-	-	16.5	-	-
5	-	-	21.7	B, C, D	29.7	B, C, D	57.8	32.9	6.1
6	~0	A	16.5	B, C, D	<20	B, C, D	100.3	16.1	1
7	-	-	-	-	-	-	-	-	-
8	1.1×10 <sup>3</sup> MPN 100 mL <sup>-1</sup>	-	13.2	B, C, D	8.5	A	30.6	-	0.4-2
9	2.3 log MPN 100 mL <sup>-1</sup>	-	-	-	-	-	48	62	7
10	-	-	19.1	B, C, D	7.1	A	51	-	-
11	-	-	<10	A	-	-	<20	-	0.8-1.4
12	-	-	-	-	-	-	-	-	-
13	-	-	1.2	A	8.5	A	38	0.5	7.9
14	-	-	-	-	-	-	-	11.2	0.35

Hybrid system	<i>E. coli</i> (cfu 100 mL <sup>-1</sup> unless stated otherwise)	Class	BOD <sub>5</sub> (mg L <sup>-1</sup> )	Class	TSS (mg L <sup>-1</sup> )	Class	COD (mg L <sup>-1</sup> )	TN (mg L <sup>-1</sup> )	TP (mg L <sup>-1</sup> )
15	-	-	-	-	-	-	-	10.8	-
16	-	-	16.2	B, C, D	-	-	79.46	-	-
17	-	-	14.4-96	B, C, D	21.34-127.6	B, C, D	42-336	-	-
18	1×10 <sup>3</sup>	C	<11.2	A, B	-	-	<33.9	-	-
19	-	-	16.8	B, C, D	8.2	A	-	3.4	1.4
20	-	-	17	B, C, D	-	-	99	-	-
21	-	-	-	-	-	-	62.3	8	0.1
22	-	-	7.7	A	2.6	A	39	6.5	2.8
23	<40	B	4	A	3	A	43	2.2	3.1
24	-	-	46	None	-	-	58	-	-
25	-	-	-	-	-	-	22.4	-	2.1
26	9.2×10 <sup>4</sup>	None	-	-	2.5-11.2	A, B, C, D	0-19.2	-	-
27a	1.8 log	B	21	B, C, D	39	None	33	10.6	5.2
27b	<1 log	A	-	-	-	-	-	-	-
28	-	-	5	A	8.9	A	25	25.3	6.5
29	-	-	25.3	None	-	-	100.8	-	3.1
30	-	-	11	B, C, D	5.6	A	28.4	26.2	-
31	-	-	21	B, C, D	8	A	73	-	-
32	-	-	3.2	A	-	-	8.8	4	-
33	3.23	A	-	-	-	-	-	-	-
34	-	-	18	B, C, D	7.6	A	18	-	8.9

Hybrid system	<i>E. coli</i> (cfu 100 mL <sup>-1</sup> unless stated otherwise)	Class	BOD <sub>5</sub> (mg L <sup>-1</sup> )	Class	TSS (mg L <sup>-1</sup> )	Class	COD (mg L <sup>-1</sup> )	TN (mg L <sup>-1</sup> )	TP (mg L <sup>-1</sup> )
35	1.11×10 <sup>3</sup> MPN 100 mL <sup>-1</sup>	-	-	-	4.1-4.6	A	35-56.9	58.6-111.6	5.5-12.2
36a	34	B	10.3	B, C, D	8.3	A	24.1	-	4.6
36b	1.6	A	6.3	A	7.7	A	18.3	-	4.8
37	-	-	30	None	84	None	47	-	-
38	-	-	20.2	B, C, D	14.9	B, C, D	48.9	-	2.9
39	-	-	48.8	None	8.46	A	138.3	45.2	8

In the studies (No. 16, 17, 18, 20, 35 and 37) when the research tested more than one hybrid system, the one with the least pollutants concentrations was reported in this table.

## 4.6. Conclusions

Constructed wetlands are recognized as an effective and inexpensive technology for wastewater treatment. This review analyzed recent experimental studies on single-stage and hybrid CWs, that tested different scales, operating times, influent strengths, plant species, etc. According to the 39 studies considered, it can be concluded that improved single-stage CWs mainly had a better performance on pollutants removal (i.e. solids, nutrients and organics) than conventional systems. The multiple-stage treatments (e.g. hybrid CWs) and in particular the application of additional technologies (e.g. UV treatment, anaerobic reactors) combined with CWs, were able to further increase and optimize overall removal effectiveness.

In addition, seasonal variation can affect pollutants removal and generally the highest removal efficiency was achieved in summer. Plants could be beneficial for removal of organics, nitrogen and phosphorus, especially after they are fully established. However, it was noted that they could also cause the negative effect on treatment performance - additional generation of organic carbon, higher ET values and increased TSS concentration due to algae growth. Regarding the substrate type, it could be concluded that the ones with a higher porosity, small media sizes and good compressibility were favorable for removal efficiencies. Also, ET, depending on plant species and climate, was shown to be able to offset the effect of wastewater treatment.

The potential of considered treatment systems for irrigation was different to a large extent. Effluent quality of systems analyzed varied in a wide range, and it could not always meet the standards for agricultural reuse imposed by the new European regulations. The improved single-stage CWs and multistage hybrid systems generally had more possibilities to produce effluents with lower BOD<sub>5</sub> concentration (class A) than single-stage CWs. Also, hybrid systems showed a better overall performance on TSS reduction. However, *E. coli* concentration could not always be reduced to a level needed for agricultural reuse without the application of specific disinfection measures. Therefore, additional technologies and treatment steps should be introduced before irrigation application in order to decrease the environmental and public health risks.

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# CHAPTER 5 - Optimization of constructed wetlands efficiency for agricultural reuse of treated wastewater

This chapter is based on a deliverable of the FIT4REUSE project:

Lavrnić, S., Mancuso, G., Nan, X., Toscano, A., Jaouani, A., Manai, I., Khadija, K., Panagou, I., Noutsopoulos, C., Malamis, S., and Mamais, D. (2020). Scientific and practical report on design, test and start-up of the NBS units. Deliverable D2.1 of the FIT4REUSE project funded under the European Union's Horizon 2020 research and innovation program GA No: 1823.

*Key words: Nature-based solutions; Constructed wetlands; Wastewater treatment*

## 5.1. Introduction

The findings presented in chapter 4 highlighted the potential of treated domestic wastewater to be reused in agriculture in order to mitigate negative effects of water scarcity and reduce the use of chemical fertilizers. In order to optimize constructed wetlands and increase their treatment efficiency so that their effluent can meet the EU or Italian reuse regulations, a pilot plant based on constructed wetland (CW) technology for domestic wastewater treatment was built within a wastewater treatment plant near the city of Bologna, in Italy. This facility and practical activities presented in this chapter are part of the FIT4REUSE project.

FIT4REUSE was financed by the PRIMA Foundation program, supported under Horizon 2020, the European Union's framework program for research and innovation. It addresses both direct and indirect use of non-conventional water resources providing guidelines and performing a holistic assessment of the use of non-conventional water resources to improve public and legal acceptance of treated wastewater. The main objective of FIT4REUSE is to provide safe, locally sustainable and accepted ways of water supply for the Mediterranean agricultural sector by exploiting non-conventional water resources.

To date, the work on design, construction and start-up of the CW units have already been completed. However, due to the impact of COVID-19 pandemic and delays caused by it, experimental activities and data analysis are still in progress.

## 5.2. Technical description

The pilot plant is located at the Granarolo dell'Emilia wastewater treatment plant (WWTP) near the city of Bologna (Italy). The WWTP treats wastewater from combined sewage system and it was designed for 9,500 PE and an average nominal daily flow rate of approximately  $2,300 \text{ m}^3 \text{ d}^{-1}$  (Fig. 5.1).

The WWTP consists of pre-treatment (coarse screening, fine screening, oil and sand removal), two parallel lines of biological treatment with activated sludge system (denitrification, oxidation/nitrification and final sedimentation), and disinfection processes. The WWTP is also equipped with a sludge line.



Fig. 5.1 - Granarolo dell'Emilia wastewater treatment plant.

The FIT4REUSE pilot plant is fed with the pre-treated wastewater of Granarolo dell'Emilia WWTP and consists of a storage tank, a sedimentation tank, 6 horizontal flow constructed wetlands (HFCWs) and 6 vertical flow constructed wetlands (VFCWs).

Sedimentation tank works as primary treatment in order to prevent clogging of the further section of the pilot plant. The number of CW units will allow testing of three different substrates and two different plant species, and choosing the ones most suitable for domestic wastewater treatment. Some specific data regarding both HFCW and VFCW are given in Table 5.1.

Table 5.1 - Specific information of the constructed wetland units.

	<b>HFCW</b>	<b>VFCW</b>
Shape	Rectangular	Circular
Length (m)	0.70	-
Width (m)	0.35	-
Radius (m)	-	0.20
Depth (m)	0.60	1.00
Substrate depth (m)	0.50	0.90
Saturated	Yes	No
Minimal flow (L day <sup>-1</sup> )	12.5	12.5
Maximal flow (L day <sup>-1</sup> )	50.0	50.0

HFCW = horizontal flow constructed wetland, VFCW = vertical flow constructed wetland

### 5.3. Design criteria, assumptions and expected water quality in relation to EU and local water reuse regulation

Both HFCW and VFCW were designed to be inside a certain range of hydraulic loading rate, 0.05-0.20 m d<sup>-1</sup> and 0.10-0.40 m d<sup>-1</sup>, respectively. Moreover, flow rates for both systems are in the range 12.5-50.0 L d<sup>-1</sup>. Consequently, HFCW has hydraulic retention time (HRT) of 0.9-3.6 days. On the other hand, the HRT of VFCW cannot be precisely calculated since it is not a saturated system but rather depends on the type of substrate and its depth.

As previously said, the pilot plant is treating the Granarolo dell'Emilia WWTP influent with characteristics given in Table 5.2.

Table 5.2 - Characteristics of the Granarolo wastewater treatment plant influent in the period 2018-2020.

<b>Parameter</b>	<b>Average (mg L<sup>-1</sup>)</b>	<b>St. error (mg L<sup>-1</sup>)</b>	<b>Sample size</b>
Chemical oxygen demand	296.7	25.3	23
Biological oxygen demand	145.5	12.8	23
Total suspended solids	102.4	10.9	23
Total phosphorus	5.0	0.6	11
Ammonium-nitrogen	56.1	4.6	11

The objective of FIT4REUSE and this pilot system is to produce effluent that is suitable for reuse in agriculture. That area is currently regulated by the Italian ministerial decree 185/2003 (Legislative Decree, 2003), known as a very strict and has been actually recognized as one of the limiting factors for wastewater reuse in Italy.

In May 2020, the EU parliament adopted new Regulation 2020/741 on minimum requirements for water reuse for agricultural irrigation that will enter into force in 2023 (European Commission, 2020). The quality of water required for irrigational reuse depends on the type of crops (e.g. eaten raw or processed; edible part is in direct contact with reclaimed water or not) and method of irrigation, taking into account 4 classes (Table 5.3).

Table 5.3 - Italian and EU limits for wastewater reuse.

	Italy reuse regulation		EU regulation for water reuse			
	General	Irrigational	Class A	Class B	Class C	Class D
Chemical oxygen demand (mg L <sup>-1</sup> )	100	100	-	-	-	-
Biological oxygen demand (mg L <sup>-1</sup> )	20	20	10	25	25	25
Total suspended solids (mg L <sup>-1</sup> )	10	10	10	35	35	35
Total nitrogen (mg L <sup>-1</sup> )	15	35	-	-	-	-
Total phosphorus (mg L <sup>-1</sup> )	2	10	-	-	-	-
<i>E. coli</i> (CFU 100 mL <sup>-1</sup> )	50	100	10	100	1,000	10,000

## 5.4. Process design calculations

This section gives the main equations used for the design calculations and the obtained results.

The needed surface area of a CW can be estimated using equation 1:

$$A = \frac{Q}{HLR} \quad (1)$$

where,

A - surface area (m<sup>2</sup>)

Q - flow rate (m<sup>3</sup> d<sup>-1</sup>)

HLR - hydraulic loading rate (m d<sup>-1</sup>)

Other important data is the voids volume that estimates the volume of water that a CW can effectively contain.

$$V_v = HRT * Q \quad (2)$$

where,

V<sub>v</sub> - volume of voids (m<sup>3</sup>)

HRT - hydraulic retention time (d)

Once obtained voids volume, the total CW volume can be calculated taking into account porosity of the substrate.



$$V = \frac{V_v}{n} \quad (3)$$

where,

n - porosity of CW substrate (-)

V - total CW volume (m<sup>3</sup>)

Finally, the substrate depth can be estimated through equation 4.

$$h = \frac{V}{A} \quad (4)$$

h – depth of the CW substrate (m)

The final results are given in Table 5.4.

Table 5.4 - The main parameters calculated.

	Horizontal flow constructed wetland	Vertical flow constructed wetland
Surface area (m <sup>2</sup> )	0.25	0.13
Flow rate (m <sup>3</sup> d <sup>-1</sup> )	12.5-50.0	12.5-50.0
Hydraulic loading rate (m d <sup>-1</sup> )	0.05-0.20	0.10-0.40
Volume of voids (m <sup>3</sup> )	≈0.045	≈0.045
Hydraulic retention time (d)	0.9-3.6	-
Total CW volume (m <sup>3</sup> )	≈0.113	≈0.113
Depth of the CW substrate (m)	0.5	0.9

## 5.5. List of operation units

The experimental pilot plant was equipped with:

- N. 1 Electrical cabinet.
- N. 1 Lifting submersible pump (DG BluePRO 75/2/G40V A1BM(T) – 60 L min<sup>-1</sup>) to collect wastewater from the influent storage unit of Granarolo dell'Emilia WWTP.
- N. 1 primary sedimentation tank of 1 m<sup>3</sup> volume for the influent.
- N. 1 Storage tank of 1 m<sup>3</sup> volume for the primary clarified effluent.
- N. 6 HFCW units (Length: 0.70 m, Width: 0.35 m, Depth: 0.60 m).
- N. 6 VFCW units (Diameter: 0,40 m, Depth: 1,0 m).

- N. 1 Steel support structure to accommodate CW units.
- N. 1 Main dosing pump (Etatron D.S. AD0084CA00100 – 84 L h<sup>-1</sup>) to feed HFCWs/VFCWs.
- N. 12 Electro-valves (Valbia s.r.l. - Ø1/4" G - VB015), one before each CWs unit.
- N. 2 Dosing recirculation pumps (Etatron D.S. AD0060CA00100 – 60 L h<sup>-1</sup>) for the combined CW units.
- N. 1 Air pump (Mapro International s.r.l. – Minicomp 3 – 10 mmH<sub>2</sub>O – 3.5 m<sup>3</sup> h<sup>-1</sup>, 2850 rpm) for the HFCW units.
- N.1 Air pump (Mapro International s.r.l. – Minicomp 3 – 10 mmH<sub>2</sub>O – 3.5 m<sup>3</sup> h<sup>-1</sup>, 2850 rpm) for the VFCW units.
- N.12 Collection tanks (Length: 0.30 m, Width: 0.30 m, Depth: 0.30 m)
- Piping system to convey wastewater to the CW units and from the collection tanks to the drainage.

## 5.6. Construction/installation and start-up

The designing and installation phases (Fig. 5.2) have been carried out by an external company under the supervision of the University of Bologna.



Fig. 5.2 - Installation of the pilot plant at the wastewater treatment plant of Granarolo (BO), Italy.

### 5.6.1. Construction and installation process

Fig. 5.3 and Fig. 5.4 show the scheme, and the main features and layers of horizontal and vertical flow units.

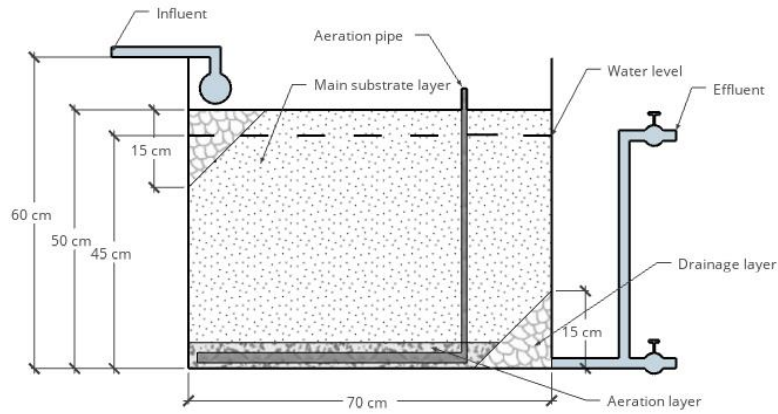


Fig. 5.3 - Scheme of the horizontal flow constructed wetland units.

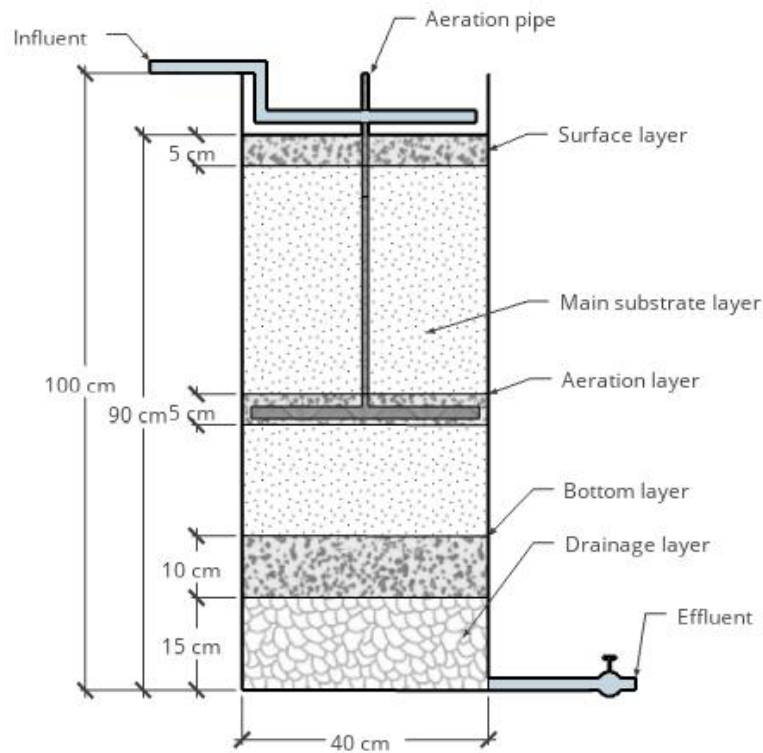


Fig. 5.4 - Scheme of the vertical flow constructed wetland units.

In HFCWs, drainage has been made through the application of a layer of gravel (10-20 mm) near the exit hole at the bottom (Fig. 5.5). Aeration pipes have been covered with a 5 cm-layer of gravel (4-6 mm) to facilitate the correct escape of air and to avoid the clogging of holes due to the possible infiltration of material from the main substrate above (Fig. 5.5).

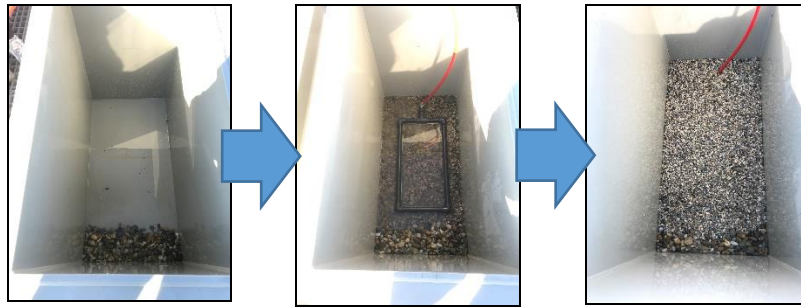


Fig. 5.5 - Drainage and aeration system in horizontal flow constructed wetlands.

In VFCWs, drainage has been made through the application of a 15 cm-layer of gravel (grain size of 10-20 mm) on the bottom (Fig. 5.6). Then, another 10 cm layer using gravel with a smaller particle size distribution (4-6 mm) has been put in the upper side to avoid the wash out of the finer material (Fig. 5.6).

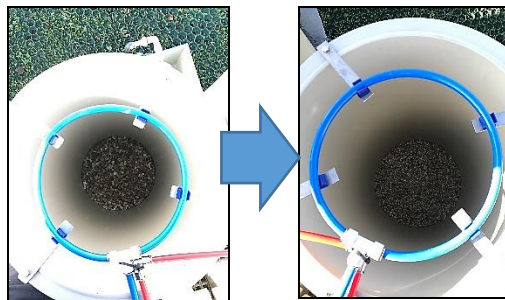


Fig. 5.6 - Drainage in vertical flow constructed wetlands.

In VFCWs, the aeration system was placed in the center of each vertical system, while in HFCWs it was on the bottom. Artificial aeration will be tested in order to improve the pollutant removal within the systems. The main substrates used in HFCW and VFCW and their characteristics are reported in Table 5.5.

Table 5.5 - List of main substrates to be used in horizontal and vertical flow constructed wetlands.

	<b>Substrate</b>	<b>Grain size (mm)</b>
HF1 – HF2	Gravel	4 - 6
HF3 – HF4	Pumice	3 - 6
HF5 – HF6	Agriperlite	2 - 6
VF1 – VF2	Sand	1 - 2
VF3 – VF4	Vermiculite	3
VF5 – VF6	Cork	2 - 3

As it can be observed from Table 5.5, tests with different main substrates will be done considering duplicates in both CW systems.

### 5.6.2. Start-up

A series of preliminary tests were carried out to verify the hydraulic seal of the system and the actual flow rate conveyed by the main pump to each CW unit (Fig. 5.7). At this stage, tap water from the WWTP of Granarolo was used. In addition, the correct operation of electro-valves, recirculation pumps and air pumps was checked (Fig. 5.8). It was also verified that the electrical cabinet allowed the correct implementation of the operating logics and data recording (flow rate administered to the system, electro-valves opening time, etc.) (Fig. 5.9).

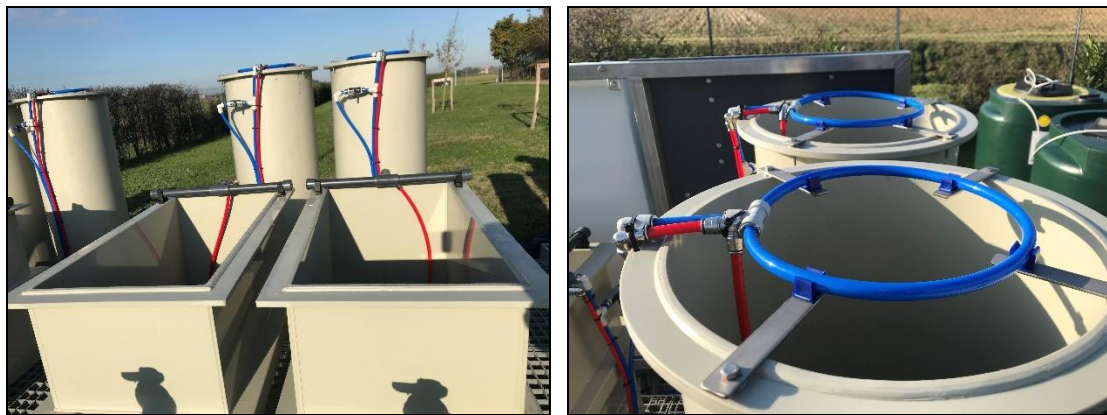


Fig. 5.7 - Measurement of the actual flow rate conveyed to each horizontal/vertical flow constructed wetland unit.



Fig. 5.8 - Check of the main hydraulic components.



Fig. 5.9 - Check of the electrical cabinet.

The acquisition system allows the download of data by means of a USB port (Cvs format). The recorded data can be then used for their subsequent elaboration.

The full installation and start-up phase were delayed due to the COVID-19 pandemics and subsequent lockdowns and they were completed only in late June 2021, when the pilot plant was considered fully operational (Fig. 5.10) and the monitoring period has started.



Fig. 5.10 - The constructed wetland pilot plant in operation (July 2021).

Based on the preliminary monitoring and data collected over the last few months, the concentration of each pollutant is reported in Table 5.6. The monitoring and data analysis are still ongoing.

Table 5.6 - The concentration of pollutants in horizontal and vertical flow constructed wetlands.

System	COD (mg L <sup>-1</sup> )	TN (mg L <sup>-1</sup> )	NH <sub>4</sub> -N (mg L <sup>-1</sup> )	NO <sub>3</sub> -N (mg L <sup>-1</sup> )	NO <sub>2</sub> -N (mg L <sup>-1</sup> )	TP (mg L <sup>-1</sup> )
Influent	367.5	66.9	-	0.4	0.0	8.7
V1	15.6	52.2	16	52.8	0.0	6.6
V2	13.0	50.9	18.4	52.1	0.0	18.9
V3	21.3	50.1	10.4	50.9	0.0	25.1
V4	14.9	48.2	16.1	48.6	0.0	14.8
V5	7.2	53.0	18.0	53.1	0.0	6.5
V6	26.3	50.4	10.4	52.7	0.0	8.1
H1	70.2	47.0	0.2	0.4	0.0	6.1
H2	104.5	53.0	0.3	0.4	0.0	6.6
H3	53.7	30.8	0.3	0.2	0.0	5.7
H4	64.3	47.1	1.8	0.4	0.0	5.9
H5	86.3	51.0	0.5	0.4	0.0	6.4
H6	41.9	30.9	0.5	0.2	0.0	5.6

COD = chemical oxygen demand, TN = total nitrogen, NH<sub>4</sub>-N = ammonium-nitrogen, NO<sub>3</sub>-N = nitrate-nitrogen, NO<sub>2</sub>-N = nitrite-nitrogen, TP = total phosphorus

## 5.7. Drawings

In this section technical drawings of the pilot system are reported. Specifically, Fig. 5.11 reports a schematic representation of the experimental setup, while Fig. 5.12 and Fig. 5.13 show the design details of HFCWs/VFCWs, collection tanks, and steel support structure, respectively. Fig. 5.14 provides the P&ID of the experimental setup.

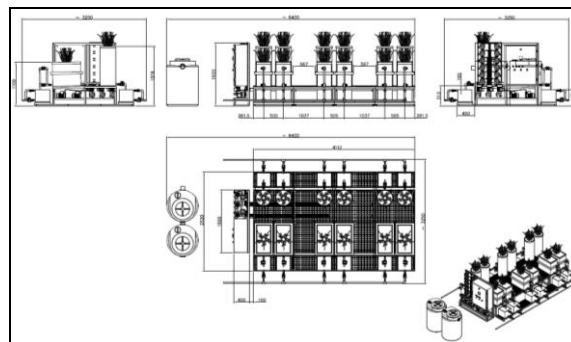


Fig. 5.11 - Design of the experimental setup.

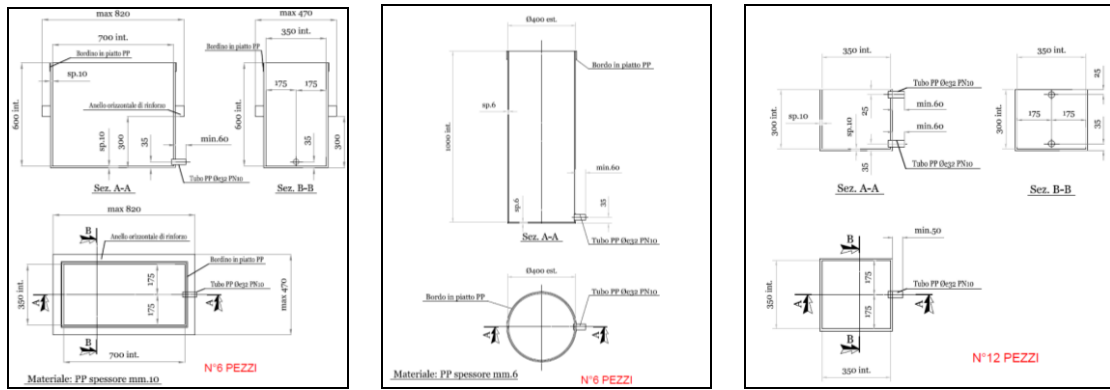


Fig. 5.12 - Design of the horizontal/vertical flow constructed wetland units and collection tanks.

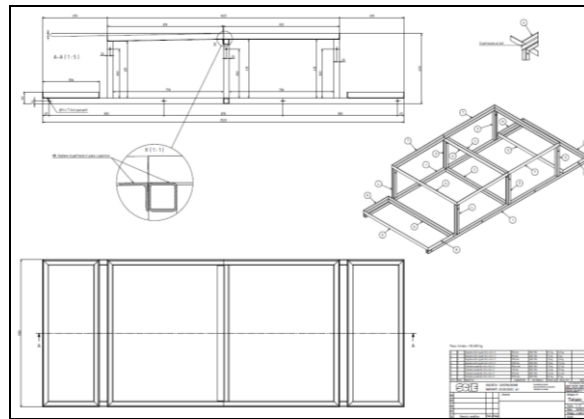


Fig. 5.13 - Design of the steel support structure.

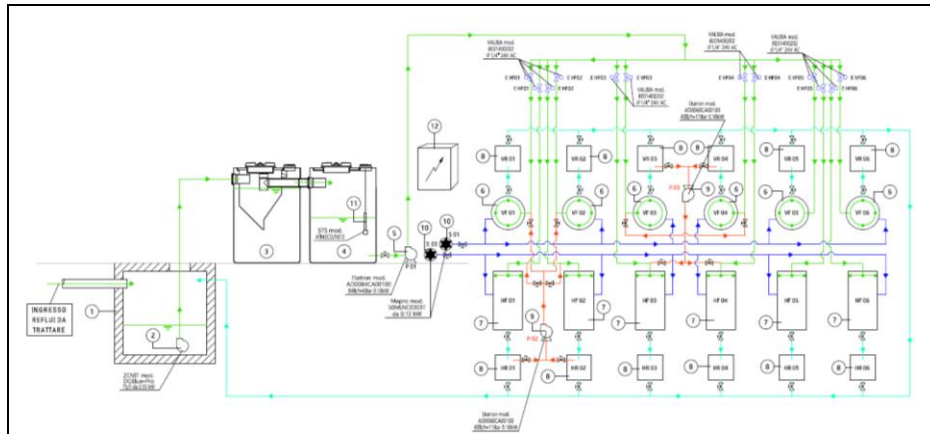


Fig. 5.14 - P&ID of the experimental setup.



## References

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# CHAPTER 6 - Conclusions



In this thesis, CWs as NBS for ADW and domestic wastewater treatment were studied and their potential was explored.

The effectiveness of CWs for the treatment of ADW has been demonstrated by both the experimental approach and the literature review. A case study on a full-scale FWS CW was carried out in Northern Italy. Although this CW has already served for two decades, it still has a good performance on the pollutant retention (e.g., TSS, TN,  $\text{NO}_3^-$ -N, TP). In particular, the monitoring data in 2018 and 2019 showed that the highest mass load retention of TSS, TN and  $\text{NO}_3^-$ -N can reach 82%, 78% and 78%, respectively. Therefore, this case study provides evidence that CWs as an ecological engineering solution can effectively treat ADW in the long term. On the other hand, the authors noticed high variability in inflow and outflow of the system, which depended on the rainfall regime and season. The water losses were mainly attributed to evapotranspiration and infiltration into the ground.

Based on the review of literature, it was found that the treatment efficacy of CWs can also be affected by design and operational conditions (i.e., location and size, hydraulic design and CW configuration, vegetation management). For example, the implementation of simple hydraulic structures can help CW systems to achieve better pollutant removal efficiencies. The vegetation management regimes (e.g., the time of vegetation establishment and harvest, the determination of vegetation species and density) are closely related to nutrient removal. In addition, it is recommended to control the length to width aspect ratio of CWs within a reasonable range (e.g., from 2 to 10), to ensure the effect of water treatment.

On the other hand, different types of CW systems for domestic wastewater treatment were reviewed. Generally, domestic wastewater quality can be effectively improved by the application of CWs or the combination of CWs and other technologies (e.g., ultraviolet treatment, anaerobic reactors). The pollutants that affect water quality mainly include organic matter, nutrients, solids and bacterial indicators. With respect to the performance of single-stage CWs, it was observed that vertical CWs had greater efficiency in organic matter removal than horizontal CWs, as the design of vertical systems provides higher oxygen concentration needed for aerobic degradation of organic matter. Nutrient removal rates of these systems varied widely, while solids removals were always high. As for microbiological indicators, the systems that showed better performance were those with longer HRT and elevated oxygen concentration.

In addition, Continuous recirculation can improve organics abatement, while the aeration can help to reduce the concentrations of nutrients, turbidity, TSS and organics. Seasonal variation was shown to affect pollutant removal (organics, microbes and nutrients) and the greatest efficiency was

usually achieved in summer. The presence of plants had contradictory effects. They might be beneficial for removal of organics, nitrogen and phosphorus, especially after they were fully established. However, it was noted that they could also have negative effects on treatment performance due to greater evapotranspiration values, increased concentration of TSS and organics and limited nutrients removal due to algae growth. Regarding the substrate type, it has been demonstrated that the ones with higher porosity, small media size and good compressibility were more likely to achieve high removal efficiency.

The quality of treated wastewater from various CW systems was quite different. However, effluents coming from hybrid systems and improved single-stage CWs had greater potential to meet agricultural reuse standards.

## 6.1. Limitations and further research

Experimental activities on ADW by CWs were finalized also taking into account the suggestions made by previous research studies (chapter 2 and 3). As for the topic of CWs for domestic wastewater treatment and consequent effluent reuse in agriculture, literature review presented in this thesis (chapter 4) provided useful suggestions for the experimental research and resulted in a specific experimental design (chapter 5) that was part of the FIT4REUSE project, but it could not be completed due to the COVID-19 pandemics. Therefore, it is still ongoing and will be continued in the future.

On the other hand, the influence of plant species and substrates on the removal efficiency of certain pollutants could be explored in the future work. In addition, more research on wastewater treatment based on hybrid and intensified CW systems is needed to clarify the advantages of different treatment technologies. However, the reasonable combination of these technologies could be expected to achieve better effluent quality and to meet agricultural reuse standards.

