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Efficiency of an Integrated Purification System for Pig Slurry Treatment under Mediterranean Climate

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Abstract: The intensification of pig production is considered a risky activity for the environment when the generated pig slurry is not adequately treated. In addition, intensive agriculture practices where pig slurry is applied to the cropland could cause nitrate leaching, salinization, and soil pollution (heavy metals and pathogens), thus the development of an eco-friendly system for pig slurry treatment is essential to avoid undesirable environmental impacts. The main objective of this study was to assess the efficiency of an integrated purification system (IPS) for treating pig slurry. The system included a pretreatment module (raw pig slurry tank, phase separator, aeration tank, and settlement tank), constructed wetlands (CWs) that included an unplanted cell and a planted cell, and a storage pond (SP). Pig slurry samples from the pretreatment modules, CWs, and SP were collected in triplicate and physic-chemical and microbial analyses were performed. Results showed that the pretreatment modules decreased the total suspended solids (TSS), total nitrogen (TN), and total phosphorus (TP) concentrations in the liquid fraction. Higher total nitrogen removal was reported in the planted cell, which decreased from 4.0 g L^{-1} to 1.9 g L^{-1} in the inflow and outflow, respectively. Total efficiencies over 85% were found in the planted and unplanted cells for TSS, Kjeldahl nitrogen (KN), chemical oxygen demand (COD), and TP. Microbial parameters were eliminated after the treatment in the planted cell. Therefore, the results indicated that filtration (pretreatment), purification (constructed wetland), and bioremediation (storage pond) constituted an appropriate IPS for treating pig slurry.

Keywords: horizontal subsurface constructed wetland; pig slurry purification; wastewater reuse; *Phragmites australis*; Suaeda vera; pollutant removal

1. Introduction

The intensification of pig production is considered a risky activity for the environment when the generated pig slurry is not adequately treated [1]. In addition, the intensive agricultural activities implicate the overuse of fertilizers (mineral and organic), which could cause nitrate leaching, salinization, and soil pollution (heavy metals and pathogens), thus the development of an eco-friendly system for pig slurry treatment is essential to avoid undesirable environmental impacts [2]. The main problems caused by pig slurry (PS) application on agricultural soils are related to eutrophication and acidification on water bodies, nitrate leaching and greenhouse gas emissions [3,4]. In addition, pig slurry is an important source of trace metals and pathogens. Trace metals, such as Cu, Zn, and Cd, are used worldwide in intensive animal breeding for preventing infectious diseases and as growth promoters [5], however, these heavy metals can be accumulated in the soil, uptake by plants and affect the health of animals and people through the food chain. Moreover, pig slurry is characterized by a high level of

bacterial population, including saprophytic microorganisms, pathogenic bacteria, viruses, and fungi, as well as eggs and oocysts of gastro-intestinal parasites, which can be accumulated in soils [6]. Also, PS is considered as one of the most agroindustrial wastewater pollutants, causing soil/water pollution, due to inappropriate treatment and management [7–9]. Nevertheless, PS includes a high amount of nutrients and water, which could be useful if it is correctly applied in agricultural soils, especially in arid and semiarid regions.

The European Economic Community (EEC) legislation regulates the nitrogen contribution to agricultural soils, establishing two main objectives: (a) reducing water pollution caused or induced by nitrates from agricultural sources, and (b) preventing further pollution [10]. This regulation encouraged countries to set up suitable agricultural practices and strategies focused on minimizing the environmental pollution, whereby it is therefore imperative to implement sustainable techniques of PS treatment and management to take advantage of it. Specifically, European legislation establishes several vulnerable zones [10], indicating that no more than 170 kg nitrogen ha^{-1} yr^{-1} can be applied to the croplands. Our study was located in the Murcia Region (SE Spain) concretely in a vulnerable zone, therefore, pig slurry management requires special attention before being applied to the land due to its high nitrogen concentration.

Spreading raw pig slurry on cropland is the traditional management of PS in agricultural areas, however, due to the above-mentioned environmental impacts, it is not the most appropriate way to manage it. Oppositely, treated pig slurry would allow the application of greater volumes on farmland [11], and it is a sustainable solution respecting the environment and legislation while offering agricultural benefits [12]. In semiarid areas such as the Mediterranean basin, the use of treated pig slurry is a good resource for the farmer because of the depletion and deterioration of the conventional water sources, the progressive salinization of aquifers and the lack of rain [13]. Therefore, the treated pig slurry is an additional non-conventional source of irrigation water and nutrients in arid and semiarid regions [7,13].

Among different wastewater treatment systems, conventional methods, where physicochemical technologies, biological reactors, activated sludge processes, or a combination of some of them are used, cannot be applied to farm level due to the high cost of investment and operation. However, new environmentally friendly and low-cost alternatives became a very attractive option for farmers, among them highlighting the constructed wetlands (CWs), which have been mainly used for urban wastewater treatment [14,15], and few studies have been described for animal wastewater treatment. Current studies have pointed out that several types of wastewater from different origins, such as agriculture, aquaculture, and industrial activities have been successfully treated with CWs [16–18]. Therefore, an innovative integrated system based on horizontal subsurface flow constructed wetland (HSFCW) could have the potential to remove pollutants from wastewaters at a low cost and at the same time respecting the environment [16,19].

This study differs from previous researches [20–22] because our study sought to develop a system that was as natural as possible without chemical treatments, and using two species of plants, combining phytoextractor and halophyte species. Therefore, the main objective of this research was to assess the efficiency of treatment of an integrated purification system, which included pretreatment module (raw pig slurry tank, phase separator, aeration tank, and settlement tank), constructed wetlands (unplanted and planted cells combining different species, *Phagmites australis* and *Suaeda vera*) and a storage pond, to treat pig slurry to be reused in agricultural applications.

2. Materials and Methods

2.1. Location of the Integrated Purification System

The integrated purification system was located in Lorca (Region of Murcia, Spain) at CIFEA (Integrated Center of Training and Agrarian Experiences) where a pilot plant was constructed (37°39′14.7″ N and 1°41′17.3″ E), under Mediterranean climate. During the experiment, the mean

annual temperature was 17.8 °C and the mean rainfall was 109.08 mm (SIAM, 2017). The pig slurry for the experiment came from a farm of 250 heads of Duroc-Jersey breed mother pigs with piglets, with an average weight of 300 kg.

2.2. Experimental Design and Sampling

The integrated purification system was composed of a pretreatment module (raw pig slurry tank, phase separator, aeration tank, and settlement tank), horizontal subsurface flow constructed wetland (HSFCW) and a storage pond (Figure 1). The PS from the pig farm was firstly pretreated in the physical pretreatment module; secondly, the PS was treated in the HSFCWs, and finally, it was collected in a storage pond.

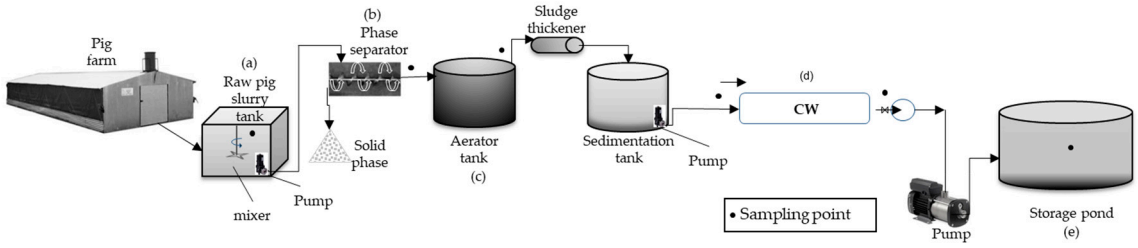

Figure 1. Scheme of the integrated purification system.

Therefore, the integrated purification system had six parts: (1) the raw pig slurry tank (RST), (2) phase separator (PHS), (3) aeration tank (AT), (4) settlement tank (ST), (5) constructed wetland (W1 = unplanted, W2 = planted) and (6) storage pond (SP). The pig slurry passed through each module until the purification process was accomplished.

2.3. Pretreatment Module

The pretreatment module included a subterranean tank (Figure 1a,) where the raw PS was collected from the pig farm and homogenized with a mechanical mixer (GTWS-44 type, Westfalia separator, Eisele, Germany) for 24 h intermittently, mixing for 1 h every 2 hours. The PS was pumped to a press screw separator (Figure 1b) with a capacity of 5 m^3 h^{-1}, then the liquid part was collected in the aerator tank (Figure 1c) of 10 m^3 staying in this tank for 48 h with an intermittent aeration process, which supplied air through a compressor of 30 m^3 h^{-1} (Josval, Herraiz, Spain). The retention time on the aerator tank was selected following the indication of other studies [7,21], which used 48 h of retention time in aeration tanks to reduce odors in similar climatic and management conditions than this study. In addition, Zhang and Zhu [23] indicated that short-term and low-intensity aeration is important to generate minimal odor during pig slurry treatment, also contributing to the decomposition of the total and volatile solids and BOD_5. Also, the alternation of aerated and low-oxygen conditions promotes nitrification and denitrification processes [24]. After aeration, the PS was passed through a sludge thickener to hold back the remaining solids and transfer the liquid part to a sedimentation tank of 10 m^3, where it stayed for 48 h. During this time, the coarse fraction of organic matter in the pig slurry was settled and the liquid fraction was pumped to the HSFCW.

2.4. Horizontal Subsurface Flow Constructed Wetland Characteristics

The horizontal subsurface flow constructed wetland (Figure 1d) consisted of two cells, each one with an area of 27 × 2.5 m and 1.2 m deep and a bottom slope of 1% (Figure 2). The porous medium used for filtration was 0.3 m of washed sand, 0.1 m of fine gravel, 0.5 m of coarse gravel and 0.30 m of fine gravel as shown in Figure 2 [7]. One cell was left unplanted as control assessing the effect of the filtration media and the other one was planted with a polyculture of *Phagmites asutralis* (*P. australis*) and *Suaeda vera* (*S. vera*) with a density of 10 plants m^{-2} and distributed as 50% of each species testing the filtration media plus plant effects. These species were chosen because *P. australis* is the most commonly

used macrophyte in wetland treatments [25] while *S. vera* is a halophyte that promotes the removal of soluble salt from PS. The HSFCWs worked in batch mode, that is, each cell works independently. The PS from the pretreatment module was pumped towards the inflow of the wetland. According to previous experiences [21,26], the selected hydraulic retention time (HRT) was seven days, during that period pollutants can be removed by microorganism activity, adsorption on the substrate and uptake by plants [27,28]. Finally, after seven days into the wetlands, the outflow was discharged to the storage pond (Figure 1e).

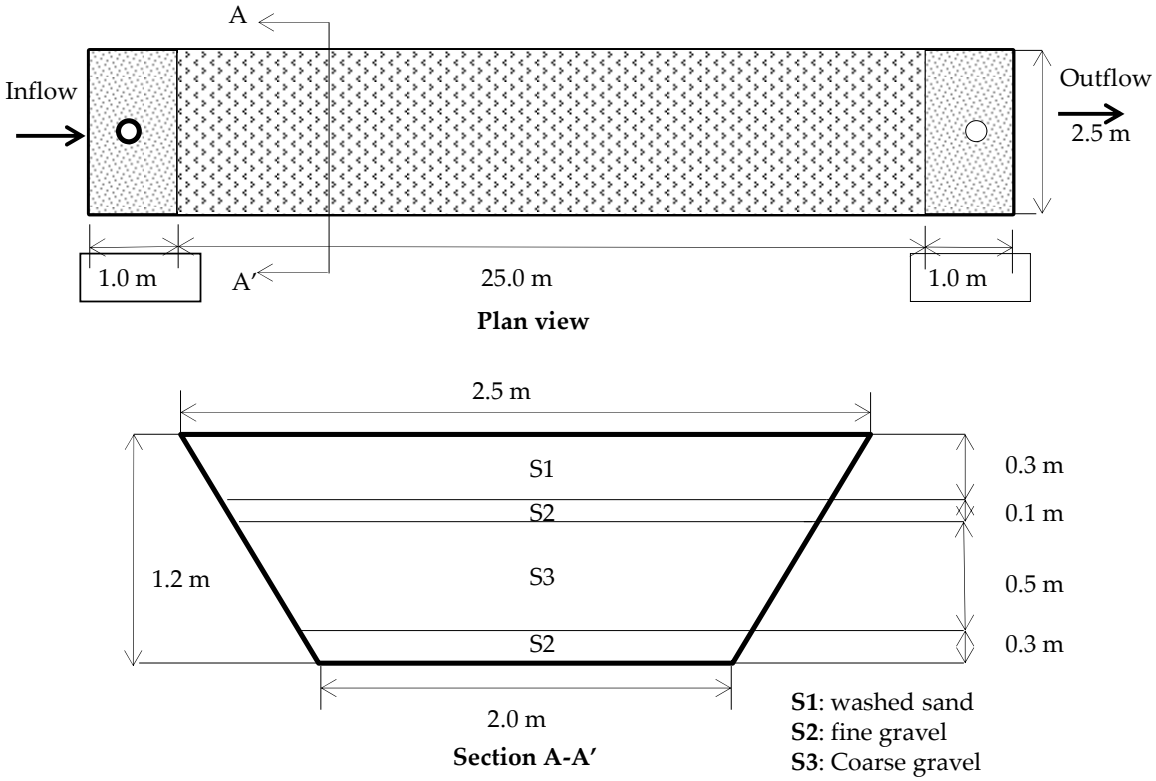

Figure 2. Horizontal subsurface constructed wetland (HSFCW) characteristics. Plan and section view. *S1* washed sand, *S2* fine gravel, and *S3* coarse gravel.

2.5. Storage Pond

The effluent from the HSFCW was pumped to a storage pond with a volume of 100 m^3 (Figure 1e). Sedimentation took place in the storage pond and algae species grew, adding a new biological process. The storage pond has been recommended by the World Health Organization because this biological process is effective in removing microbial such as nematode eggs and helminth eggs [29].

2.6. Sampling Design

All samples were taken in sterile containers of 250 mL at the eight sampling points indicated in Figure 1. Three cycles with an HRT = seven days per each cell (W1 and W2) were performed. At the beginning of the experiment enough volume of PS was stored (RST) to feed the wetlands in the three cycles, thus points a, b, and c were sampled one time for each cell (planted and unplanted). On the other hand, sampling points d and e were sampled in the three cycles for each cell (planted and unplanted). Finally, pig slurry from the storage pond (point f) was sampled at the end of the three cycles for each cell (planted and unplanted). This sampling design was followed taking into account the characteristics of pig slurry stored in RST did not change during the experimental time (three cycles of seven days), mainly because this tank was constructed underground, and the climatic condition did not change during that time. In addition, the conditions of management (retention times, aeration time,

etc.) on the pretreatment module (raw pig slurry tank, phase separator, aeration tank, and settlement tank) were strictly controlled and the effects on pig slurry characteristics were very homogenous.

2.7. Physical, Chemical, and Microbial Parameters and Analytical Methods

Temperature (T) and pH were determined by a portable probe (Hanna model HI 9025, Barcelona, Spain), the electrical conductivity (EC) was analyzed by a portable conductivity meter (Hanna model HI 9033, Barcelona, Spain), and all were measured *in situ*.

Kjeldahl nitrogen (KN) was determined by a modified Kjeldahl method [30], using 1 mL of pig slurry in the digestion. Ammonium nitrogen (NH_4^+-N) was determined by steam distillation and titration with HCl 0.1 N. Total nitrogen (TN) includes organic and inorganic nitrogen forms (Kjeldahl nitrogen plus nitrites and nitrates form). Total phosphorus (TP) was photometrically determined as molybdenum blue after acidic hydrolysis and oxidation at 120 °C (Macherey-Nagel GmbH & Co.KG. Nanocolor Test; ref. 985-055). Potassium (K^+) was determined using an atomic absorption spectrometer (Perkin Elmer AA-Analyst, 800).

Total suspended solids (TSS) were filtered through a weighed standard glass-fiber filter and the residue retained on the filter was dried to a constant weight at 105 °C (2440-D method, APHA-AWWAWEF 2012). Biochemical oxygen demand in five days (BOD_5) was determined by manometer OXITOP WTW equipment. Chemical oxygen demand (COD) was determined by photometric determination of chromium (III) concentration after 2 h of oxidation with potassium dichromate/sulphuric acid and silver sulphate at 148 °C (Macherey-Nagel GmbH & Co. KG. Nanocolor Test, Ref 985 028/29), (DIN 38 409-H41-1, DIN ISO 15 705-H45).

Anions (Cl^-, Br^-, SO_4^{2-}, F^-, NO_3^- and NO_2^-) were analyzed by high performance ion chromatography (IC) (Methrom, model 861), and cations (Na^+, Ca^{2+} and Mg^{2+}) were determined using atomic absorption spectrometer (Perkin Elmer AA-Analyst, 800).

Microorganisms were determined by preparing serial dilutions of samples with autoclaved peptone water following the normalized methods according to Standardized Work Procedures which uniform European Norm (EN), *Norme Française* (NF) and *Association Française de Normalisation* (AFNOR). Mesophilic aerobic (MA) bacteria count was determined by normalized method AF V 08-01, inoculating each sample in PCA (Plate Count Agar), incubated at 30 ± 1°C for 24–72 h. Total coliforms (TC) count was determined by normalized method NF V 08-050, inoculating each sample in VRBD (Violet Red Bile Dextrose) agar, incubated at 30 ± 1 °C for 24 ± 2 h. Fecal coliforms (FC) count was determined by normalized method NF V 08-054, inoculating each sample in VRBD (Violet Red Bile Dextrose) agar, incubated at 44 ± 1 °C for 24 ± 2 h. *Escherichia coli* colonies (*E. coli*) count was determined by normalized method NF V 08-053, inoculating each sample in a selective chromogenic medium incubated at 35 ± 1 °C for 24–48 h. Fecal streptococcus (FS) count was determined by normalized method NF EN ISO 7899-2, inoculating each sample in KAA (Kanamycin Aesculin Azide) agar incubated at 44 ± 1 °C for 24 ± 2 h. *Salmonella sp.* and *Shigella sp.* presence was determined by normalized method AF V 08-052, with an enrichment of two selective broths (Selenite Cystine and Rappaport Vassiliadis) incubated at 37 ± 1 °C and 42 ± 1 °C, respectively for 24 ± 2 h and transferring to a Petri plate with each sample inoculated in XLD (Xylose-Lysine-Desoxycholate) agar incubated at 37 ± 2°C for 24 ± 2 h. All samples were inoculated at the bottom of the Petri dish and aerobically incubated, except for *Salmonella sp.* and *Shigella sp.* which were inoculated on the surface.

2.8. Statistical Analysis

Statistical analyses were performed using SPSS (SPSS Version 23.0, Chigago, IL., USA). The Kolmogorov-Smirnov Test was used to study the normality of the data distribution and transformations into Ln were carried out to achieve normality. The homogeneity of the variance was confirmed by Bartlett's Test. One-way ANOVA was performed by post hoc Tukey's Test at $p < 0.05$ which were completed to identify significant differences through the comparisons of all possible pairs of means at the inflow and the outflow for both wetlands (W1 and W2).

3. Results

3.1. Physico-Chemical Parameters

3.1.1. Pretreatment

Tables 1 and 2 show the physic-chemical characteristics of pig slurry of each module for the unplanted cell (W1) and the planted cell (W2) respectively. Pretreatment modules of W1 and W2 operated separately, firstly the three cycles for W1 were performed and secondly the three cycles for W2. Significantly different ($p < 0.05$) of TSS concentrations were found in the raw pig slurry tank compared to the inflow of the wetlands, decreasing from 60.2 to 26.1 g L^{-1} in W1 and from 56.3 to 29.5 g L^{-1} in W2.

Non-significant differences among pretreatment modules were reported for nitrogen concentrations (NH_4^+, ON, NO_3^-, and NO_2^-) in W1. Contrarily, although the total nitrogen (TN) concentration did not differ significantly among the pretreatment modules in W2, a slight decrease of TN concentration when PS passed through the PHS in the planted constructed wetland (W2) was observed. In addition, the concentration of KN was significantly different ($p < 0.05$) between RST (5.0 g L^{-1}) and IW2 (4.0 g L^{-1}). Nitrates and nitrites had similar behavior with non-significant differences among the pretreatment modules for W2.

The maximum concentration of BOD_5 in pretreatment module RST of W1 was 4.4 g L^{-1} showing a significate decrease in IW1 (2.6 g L^{-1}). In contrast, an increase from 1.2 g L^{-1} to 3.3 g L^{-1} was observed in W2 between RST and IW2 respectively. For both cases, unplanted and planted cells, chemical oxygen demand (COD) did not vary statistically among pretreatment modules (raw pig slurry tank, phase separator, aeration tank, and settlement tank). COD concentrations of 37.0 and 26.6 g L^{-1} and of 35.3 and 28.7 g L^{-1} were reported in W1 and W2 for RST and IW, respectively.

Non-significant differences among pretreatment modules were reported for TP in W1, which mean values ranged between 1728 to 1093 mg L^{-1}. In contrast, the pretreatment exhibited a significant decrease ($p < 0.05$) of TP between RST and IW2 for W2, 1953 to 1398 mg L^{-1}, respectively.

Ca^{2+} and Mg^{2+} concentrations increased slightly when PS passed through the pretreatment module for W1, however Na^+ and K^+ were scarcely reduced, with mean concentrations of 421 to 347 mg L^{-1} for Na^+ and 1504 to 1316 mg L^{-1} for K^+. For W2 pretreatment (Table 2), Mg^{2+}, Na^+ and K^+ concentrations decreased when compared RST and IW2, except for Ca^{2+} which decreased after aeration tank from 112.0 to 107.0 mg L^{-1}.

3.1.2. Horizontal Subsurface Flow Constructed Wetland

The temperatures observed in W1 and W2 were relatively constant during the study (18 °C and 19 °C, respectively), with no significant differences ($p < 0.05$) between the inflow and the outflow of the wetlands. A slight decrease of EC from inflow to outflow for W1 and W2 was observed, with mean values ranged from 19.7 to 16.9 dS m^{-1} and from 22.5 to 20.4 dS m^{-1}, respectively.

Total suspended solids were significantly reduced ($p < 0.05$) after the wetlands, with mean concentrations of 26.1 g L^{-1} (inflow) and 2.1 g L^{-1} (outflow), 29.5 g L^{-1} (inflow) and 2.2 g L^{-1} (outflow), for W1 and W2, respectively.

The results showed higher mean concentrations of total nitrogen removal for W2 than those reported for W1, with mean concentrations of 3.5 to 3.1 g L^{-1} and 4.0 to 1.9 g L^{-1} from the inflow and outflow in W1 and W2, respectively (Tables 1 and 2).

Remarkable results were obtained for both, unplanted and planted cells, where BOD_5 and COD were significantly reduced (Tables 1 and 2), with mean concentrations of 2.6 to 1.0 g L^{-1} and 26.6 to 4.9 g L^{-1} for BOD_5 and COD removal, respectively in W1, while in W2, the mean concentrations ranged from 3.3 to 1.5 g L^{-1} and from 28.7 to 6.5 g L^{-1} for BOD_5 and COD, respectively.

The mean concentrations of total phosphorus significantly decreased ($p < 0.05$) from 1093 to 59.4 mg L^{-1} for W1, and from 1938 to 44.1 mg L^{-1} for W2.

Finally, concentrations of Cu, Fe, Mn, and Zn decreased at the outflow of W1 and W2, except for Cu in the planted cell (W2). Fe and Mn were statistically lower ($p < 0.05$) in the outflow for both W1 and W2, showing an important and positive efficiency, over 50% for Fe and over 80 % for Mn.

3.1.3. Storage Pond

Values of EC, Fe, and Mn significantly decreased ($p < 0.05$) after storage pond for W1, while for W2 decreased T, EC, NH_4^+, Cl^-, and SO_4^{2-}, oppositely SO_4^{2-} increased after the storage pond for W1 (Tables 1 and 2). However, it should be highlighted that although non-significant differences were observed, TSS, pH, TN, BOD_5, COD, TP, and Zn decreased at the outlet of the wetland and this behavior continued after the storage pond in both treatments, unplanted and planted cell.

3.2. Microbial Parameters

Remarkable percentages of reduction were identified for microbial parameters, with the highest efficiencies being for MA (22%), TC (100%) after W1 (Table 3), and FS (99%), *E. coli* (100%) after W2 (Table 4). In W2, FS, FC, and TC diminished by 100 %. *Salmonella sp.* and *Shigella sp* resulted negative for all modules. Microbial parameters such as FS, FC, and TC were eliminated in SP of W2 and *E. coli* in SP of W1 (Tables 1 and 2).

Table 1. Mean and standard deviation values, efficiencies of each module for unplanted cell (W1). Efficiency is referred to a previous module.

Parameters *	Module (n = 3)																						Efficiency (%)				
	RST				PHS				AT				IW1				OW1				SP				PHS	AT	IW1	OW	SP
T (°C)	**16.4**	**±**	**1.1**	**a**	**16.4**	**±**	**0.4**	**a**	**15.2**	**±**	**0.4**	**a**	**18.1**	**±**	**1.8**	**a**	**18.5**	**±**	**0.7**	**a**	**15.5**	**±**	**1.6**	a	0	8	−19	−2	16
TSS (g L^{-1})	60.2	±	0.8	c	38.9	±	0.6	bc	40.5	±	1.8	bc	26.1	±	26.2	ab	2.1	±	0.6	a	2.1	±	0.7	a	35	−4	36	92	4
pH	7.7	±	0.0	ab	7.8	±	0.0	ab	8.2	±	0.0	b	8.2	±	0.4	b	7.6	±	0.1	a	7.8	±	0.0	ab	−1	−6	0	7	−3
EC (dS m^{-1})	19.5	±	0.3	b	21.2	±	0.5	b	21.0	±	0.3	b	19.7	±	2.8	b	16.9	±	2.1	b	12.2	±	0.1	a	−9	1	6	14	28
TN (g L^{-1})	4.3	±	0.1	b	3.8	±	0.1	b	3.8	±	0.2	b	3.5	±	1.1	ab	3.1	±	0.9	ab	2.0	±	0.4	a	11	−1	10	10	37
KN (g L^{-1})	4.2	±	0.1	b	3.7	±	0.1	b	3.7	±	0.1	b	3.4	±	1.2	b	1.4	±	0.2	a	0.5	±	0.0	a	11	−1	10	58	64
NH_4^+ (g L^{-1})	2.8	±	0.2	b	2.6	±	0.1	b	2.5	±	0.1	b	2.3	±	0.6	b	1.1	±	0.2	a	0.4	±	0.0	a	6	4	10	50	63
ON (g L^{-1})	1.4	±	0.3	b	1.1	±	0.1	b	1.2	±	0.2	b	1.1	±	0.6	b	0.3	±	0.1	a	0.1	±	0.0	a	23	−12	9	73	66
BOD_5 (g L^{-1})	4.4	±	0.5	d	4.2	±	0.4	cd	4.2	±	0.5	d	2.6	±	1.2	bc	1.0	±	0.1	ab	0.6	±	0.2	a	5	−1	39	61	36
COD (g L^{-1})	37.0	±	2.0	b	32.3	±	3.2	b	34.7	±	2.3	b	26.6	±	19.7	ab	4.9	±	0.9	a	2.8	±	0.1	a	13	−7	23	82	42
TP (mg L^{-1})	1662	±	335	b	1608	±	60.3	b	1728	±	321	b	1093	±	1044	ab	59.4	±	19.0	a	21.8	±	1.9	a	3	−7	37	95	63
Cu (mg L^{-1})	2.5	±	0.5	b	1.3	±	0.1	a	1.3	±	0.2	a	1.5	±	0.8	ab	0.6	±	0.3	a	0.7	±	0.1	a	49	2	−19	60	−21
Fe (mg L^{-1})	0.7	±	0.4	a	13.6	±	2.0	cd	11.9	±	1.7	c	18.1	±	1.8	d	7.0	±	2.8	b	2.4	±	0.1	a	−1852	12	−52	61	66
Mn (mg L^{-1})	12.1	±	2.1	d	2.8	±	0.2	cd	2.6	±	0.5	c	2.2	±	0.4	c	0.3	±	0.2	b	0.1	±	0.0	a	77	7	14	85	67
Zn (mg L^{-1})	1.6	±	0.1	ab	6.1	±	0.8	ab	6.9	±	1.1	ab	8.2	±	5.0	b	3.0	±	2.0	ab	1.1	±	0.1	a	−275	−12	−19	64	62
Cl^- (mg L^{-1})	1000	±	68	b	1028	±	51.8	b	985	±	32.8	b	954	±	77.2	ab	903	±	91.7	ab	797	±	54.2	a	−3	4	3	5	12
Br^- (mg L^{-1})	12.1	±	0.2	ab	12.2	±	0.1	ab	13.3	±	0.9	ab	14.3	±	2.2	b	12.3	±	0.8	ab	11.9	±	0.2	a	−1	−9	−7	14	4
SO_4^{-2} (mg L^{-1})	88.9	±	9.3	a	157	±	29.3	a	74.5	±	18.2	a	152	±	159	a	196	±	181	a	607	±	20.6	b	−77	53	−104	−29	−209
NO_3^- (mg L^{-1})	35.1	±	1.2	a	37.1	±	4.2	a	34.9	±	1.0	a	36.5	±	1.8	a	36.1	±	5.8	a	28.4	±	1.1	a	−6	6	−5	1	21
NO_2^- (mg L^{-1})	65.7	±	17.6	a	86.2	±	9.2	a	49.7	±	9.9	a	78.7	±	64.7	a	1464	±	692	b	1200	±	30.3	b	−31	42	−58	−1761	18
F^- (mg L^{-1})	2.8	±	0.1	a	2.6	±	0.9	a	3.0	±	1.0	a	2.4	±	0.7	a	1.9	±	0.1	a	2.0	±	0.0	a	8	−18	19	21	−1
Ca^{+2} (mg L^{-1})	14.5	±	3.4	a	57.7	±	1.6	ab	54.4	±	8.4	ab	90.6	±	45.7	b	15.7	±	8.5	a	27.3	±	1.1	a	−297	6	−66	83	−74
Mg^{+2} (mg L^{-1})	122	±	25.6	ab	204	±	2.1	b	229	±	54.9	b	149	±	87.0	ab	43.2	±	20.0	a	119	±	37.1	ab	−67	−12	35	71	−176
Na^+ (mg L^{-1})	421	±	66.2	bc	262	±	64.1	ab	173	±	61.2	a	347	±	111	abc	455	±	46.2	c	430	±	17.8	bc	38	34	−100	−31	5
K^+ (mg L^{-1})	1504	±	143	b	1307	±	65.1	ab	1230	±	274	ab	1316	±	109	ab	1110	±	164	ab	936	±	9.0	a	13	6	−7	16	16

**T:* temperature, *TSS:* total suspended solids, *EC:* electrical conductivity, *TN:* total nitrogen, *KN:* Kjedahl nitrogen, *ON:* organic nitrogen, *BOD_5:* biochemical oxygen demand, *COD:* chemical oxygen demand, *TP:* Total phosphorous. *RST:* raw pig slurry tank, *PHS*: phase separator, *AT*: aeration tank, *IW1:* inflow unplanted wetland, *OW1:* outflow unplanted wetland and *SP:* storage pond.

Table 2. Mean and standard deviation values, efficiencies of each module for planted cell (W2). Efficiency is referred to a previous module.

Parameters *	Module (n = 3)																						Efficiency (%)				
	RST				PHS				AT				IW2				OW2				SP								
																							PHS	AT	IW2	OW2	SP
T (°C)	18.2	±	0.0	b	19.9	±	0.0	bc	20.3	±	0.6	c	19.3	±	0.1	bc	19.8	±	1.5	bc	16.0	±	0.4	a	−9	−2	5	−3	19
TSS (g L^{-1})	56.3	±	9.3	c	40.2	±	1.0	b	36.8	±	3.4	b	29.5	±	8.4	b	2.2	±	0.7	a	1.5	±	0.1	a	29	8	20	93	34
pH	7.8	±	0.1	ab	7.9	±	0.1	ab	8.3	±	0.0	b	8.1	±	0.3	b	7.6	±	0.2	a	8.0	±	0.0	ab	−1	−5	2	7	−5
EC (dS m^{-1})	22.2	±	2.0	bc	24.0	±	0.7	c	23.5	±	0.5	c	22.5	±	1.0	bc	20.4	±	0.9	b	14.4	±	0.2	a	−8	2	4	9	29
TN (g L^{-1})	5.1	±	0.3	b	4.5	±	0.2	b	4.3	±	0.4	b	4.0	±	0.5	b	1.9	±	0.4	a	1.7	±	1.1	a	11	3	7	52	14
KN (g L^{-1})	5.0	±	0.3	c	4.5	±	0.2	bc	4.3	±	0.4	bc	4.0	±	0.5	b	1.9	±	0.4	a	1.0	±	0.0	a	11	3	7	52	48
NH_4^+ (g L^{-1})	3.6	±	0.2	c	3.6	±	0.2	c	3.0	±	0.1	c	3.0	±	0.3	c	1.6	±	0.5	b	0.8	±	0.0	a	1	17	0	45	49
ON (g L^{-1})	1.4	±	0.1	b	1.1	±	0.1	b	1.3	±	0.3	b	1.0	±	0.3	b	0.3	±	0.1	a	0.2	±	0.0	a	22	−23	22	73	38
BOD_5 (g L^{-1})	1.2	±	0.3	ab	2.5	±	0.2	bc	3.0	±	0.5	c	3.3	±	1.0	c	1.5	±	0.4	ab	0.9	±	0.0	a	−103	−20	−10	54	41
COD (g L^{-1})	35.3	±	7.8	b	35.7	±	2.1	b	27.0	±	7.0	b	28.7	±	5.4	b	6.5	±	1.0	a	5.6	±	0.1	a	−1	24	−6	77	13
TP (mg L^{-1})	1953	±	292	d	1697	±	184	cd	1272	±	67.5	b	1398	±	43.3	b	44.1	±	11.1	a	31.4	±	0.5	a	13	25	−10	97	29
Cu (mg L^{-1})	1.5	±	0.4	a	2.3	±	0.4	a	2.7	±	0.2	a	1.6	±	0.8	a	2.3	±	0.9	a	1.8	±	0.0	a	−58	−16	41	−46	24
Fe (mg L^{-1})	13.9	±	2.4	b	20.9	±	3.1	cd	22.4	±	0.6	d	17.3	±	2.2	bc	8.5	±	0.2	a	8.7	±	0.4	a	−51	−7	23	51	−2
Mn (mg L^{-1})	2.3	±	0.5	bc	2.9	±	0.4	c	2.7	±	0.3	c	1.8	±	0.3	b	0.3	±	0.2	a	0.2	±	0.0	a	−29	7	35	82	27
Zn (mg L^{-1})	6.5	±	1.1	abc	10.3	±	1.5	cd	12.4	±	0.8	d	9.8	±	1.7	bcd	5.9	±	2.3	ab	5.4	±	0.2	a	−59	−21	21	40	8
Cl^- (mg L^{-1})	1359	±	33.2	a	1369	±	18.7	a	1425	±	31.2	ab	1277	±	44.1	a	1572	±	145	b	1365	±	39.4	a	−1	−4	10	−23	13
Br^- (mg L^{-1})	12.3	±	0.7	a	11.9	±	0.8	a	12.4	±	0.0	a	13.8	±	1.9	a	13.1	±	0.8	a	13.3	±	0.2	a	4	−5	−11	5	−1
SO_4^{-2} (mg L^{-1})	52.1	±	3.9	a	43.5	±	4.5	a	46.0	±	2.3	a	202	±	16.9	b	327	±	98.5	c	97.1	±	0.2	ab	16	−6	−339	−62	70
NO_3^- (mg L^{-1})	30.4	±	0.8	a	30.3	±	0.5	a	30.8	±	1.4	a	33.5	±	5.6	a	28.0	±	2.0	a	31.6	±	1.8	a	0	−2	−9	16	−13
NO_2^- (mg L^{-1})	4.5	±	0.4	a	4.7	±	0.5	a	4.8	±	0.5	a	4.5	±	0.4	a	4.2	±	0.5	a	4.1	±	0.2	a	−5	−3	7	7	2
F^- (mg L^{-1})	3.1	±	1.0	a	3.3	±	0.6	a	3.1	±	0.8	a	2.1	±	0.3	a	2.0	±	0.1	a	2.0	±	0.1	a	−5	6	32	6	−2
Ca^{+2} (mg L^{-1})	98.9	±	17.9	b	106	±	27.4	b	112	±	19.6	b	107	±	13.6	b	53	±	36.1	a	33.1	±	0.3	a	−7	−5	4	51	37
Mg^{+2} (mg L^{-1})	212	±	41.2	cd	269	±	16.0	d	192	±	0.2	cd	158	±	12.5	bc	82	±	50.3	ab	80.7	±	8.6	a	−27	29	18	48	2
Na^+ (mg L^{-1})	670	±	141	b	188	±	5.5	a	219	±	61.3	a	371	±	66.9	a	720	±	25.2	b	802	±	13.3	b	72	−16	−69	−94	−11
K^+ (mg L^{-1})	1907	±	234	bc	2202	±	63.2	c	2192	±	32.7	c	1855	±	105	b	738	±	115	a	827	±	8.9	a	−15	0	15	60	−12

* *T:* temperature, *TSS:* total suspended solids, *EC:* electrical conductivity, *TN:* total nitrogen, *KN:* Kjedahl nitrogen, *ON:* organic nitrogen, *BOD_5:* biochemical oxygen demand, *COD:* chemical oxygen demand, *TP:* Total phosphorous. *RST:* raw pig slurry tank, *PHS*: phase separator, *AT*: aeration tank, *IW1:* inflow unplanted wetland, *OW1:* outflow unplanted wetland and *SP:* storage pond.

Table 3. Microbiological parameters and efficiencies for each module on unplanted cell (W1). Efficiency is referred to a previous module and total efficiency was calculated SP with respect to RST.

Parameter *	Modules (n = 3)																Efficiency (%)				
	RST			PHS			AT			IW1		OW1			SP							
FS (UFC L^{-1})	**6.1×10^4**	**±**	**9.2×10^3**	**6.6×10^4**	**±**	**1.6×10^4**	**1.9×10^4**	**±**	**1.9×10^4**	**7.5×10^4**	**±**	8.4×10^4	5.1×10^3	±	4.2×10^3	7.7×10^3	±	8.5×10^3	−8	71	93	−50	87
MA (UFC L^{-1})	1.5×10^6	±	3.5×10^5	2.1×10^6	±	6.0×10^5	4.6×10^6	±	1.7×10^6	3.7×10^6	±	2.2×10^6	2.3×10^6	±	2.0×10^6	3.2×10^6	±	5.5×10^5	−40	−119	38	−42	−113
FC (UFC L^{-1})	1.7×10^5	±	4.6×10^4	2.0×10^5	±	0	5.7×10^4	±	1.6×10^4	1.4×10^4	±	1.2×10^4	5.5×10^3	±	1.1×10^4	2.7×10^4	±	7.6×10^3	−18	72	62	−391	84
TC (UFC L^{-1})	1.2×10^4	±	3.1×10^3	1.2×10^4	±	3.2×10^3	1.1×10^4	±	7.6×10^3	4.4×10^3	±	3.2×10^3	0	±	0	0	±	0	0	8	100	0	100
E. coli (UFC L^{-1})	8.5×10^4	±	7.6×10^3	1.3×10^5	±	1.5×10^4	1.2×10^5	±	1.5×10^4	1.2×10^4	±	1.5×10^4	1.7×10^3	±	4.2×10^2	0	±	0	−53	8	87	100	100

Table 4. Microbiological parameters and efficiencies for each module on planted cell (W2). Efficiency is referred to a previous module and total efficiency was calculated SP with respect to RST.

Parameter *	Modules (n = 3)																Efficiency (%)				
	RST			PHS			AT			IW2		OW2			SP							
FS (UFC L^{-1})	1.7×10^4	±	1.7×10^3	1.4×10^4	±	5.6×10^3	3.5×10^3	±	1.0×10^3	3.5×10^4	±	7.6×10^4	5.2×10^2	±	7.6×10^2	0	±	0	19	75	99	100	100
MA (UFC L^{-1})	1.1×10^6	±	3.9×10^5	1.6×10^6	±	2.9×10^5	8.9×10^5	±	5.2×10^5	1.0×10^6	±	6.6×10^5	8.1×10^5	±	4.3×10^5	1.6×10^6	±	7.4×10^5	−55	46	22	−97	−52
FC (UFC L^{-1})	1.0×10^3	±	1.5×10^3	2.0×10^3	±	4.0×10^2	0	±	0	4.7×10^2	±	1.2×10^2	9.0×10^2	±	9.3×10^2	0	±	0	−95	100	−93	100	100
TC (UFC L^{-1})	1.7×10^3	±	2.6×10^3	1.1×10^4	±	5.2×10^3	0	±	0	1.6×10^3	±	7.3×10^2	2.5×10^3	±	2.3×10^3	0	±	0	−557	100	−57	100	100
E. coli (UFC L^{-1})	0	±	0	0	±	0	0	±	0	2.9×10^2	±	1.1×10^2	0	±	0	0	±	0	0	0	100	0	0

4. Discussion

4.1. Physic–Chemical Parameters

4.1.1. Pretreatment

In order to guarantee the effectiveness of wetland, an adequate pretreatment of solid/liquid separation of pig slurry must be performed [31]. Results showed that TSS was reduced by 35% with the mechanical separation, previous studies have indicated that a mechanical pretreatment previous the use of HSFCWs avoid clogging of the wetlands and lengthens the life of the system [32]. Caballero-Lajarín et al. [7] only achieved 16% of TSS reduction, while Burton [33] pointed out that physical separation can remove up to 80%.

The EC is usually high due to high salt contents in raw slurry [34] likely caused by the animals' diet and their natural metabolism. The highest EC values were found in the PHS module, 21.2 and 24.0 dS m^{-1} for W1 and W2, respectively, with no significant differences in the modules of this pretreatment. Our results were in line with previous studies performed by Møller et al. [35] and Christensen et al. [3], who reported mean value around 21.0 dS m^{-1}. However, some studies have indicated that physical separation of the solid and liquid fraction from the raw pig slurry could slightly reduce the EC due to chemical precipitation of salts in the solid phase [13,36].

According to the results found by Møller et al. [35], a considerable amount of dry matter is retained in the solid fraction, therefore when the solid and liquid phases are separated by a screw press separation, the TN could be reduced in the liquid fraction, this study reached 6.6% of TN removal, while in our study 7%–10% were obtained. On the other hand, the mean temperature of the pig slurry was around 20 °C, maintaining NO_3^- and NO_2^- contents through the pretreatment, thereby this fact could indicate that the nitrification-denitrification process did not take place or maybe it was not completed, inhibiting NO_3^- and decreasing NO_2^- [37,38].

The decrease in the nitrogen concentration was around 10% in the pretreatment modules mainly because of solid/liquid separation. Highlight that the majority of KN in most of the livestock wastewater systems is in inorganic form, mostly as ammonium [7,26,39] and therefore could be adsorbed to the colloids of suspended organic matter. Bernal et al. [40] reported that losses of nitrogen ranged from 5% to 60% during storage or composting manure compared with the initial content, which was mainly caused by gaseous emissions of NH_3 [41]. Hjorth et al. [36] indicated that a screw press separator is able to retain up to about one quarter of N and P in the solid fraction of the pig slurry—likewise, Møller et al. [35] reported that screw press separation could increase, in the solid fraction, the concentration of dried matter, TN, and TP by up to five, three, and two times, respectively.

The separation carried out in this study by press screw separator of the solid to liquid fraction contributed to decrease organic matter content in PS, and therefore BOD_5 and COD were also decreased by 39% and 23%, respectively in the unplanted system (W1). However, higher percentages of reduction for COD (69.1%) were reported by Muñoz et al. [21], which depends on the physical and chemical manure composition as well as on manure age as indicated by Møller et al. [35].

A slight amount of TP was transferred from the liquid to the solid fraction during the screw press separation. Møller et al. [35] reported 2.13 g L^{-1} and 1.23 g L^{-1} in solid and liquid fraction respectively after separation with a screw press. Burton [33] pointed out that raising the pH allowed the chemical equilibrium to move towards the formation of phosphate (PO_4^{3-}), promoting subsequent precipitation as calcium phosphate. However, because the pH of our study was constant this process was incomplete, and the TP concentration did not decrease as much as expected.

During pretreatment the best results in this study were obtained after AT when W2 was performed, achieving percentages of removal of 4% and 18% for Ca^{2+} and Mg^{2+}, respectively. Na^+ and K^+ were reduced 34% and 6%, respectively after AT in W1, even though no significant differences were found. According to Burton [33], calcium and magnesium could be removed as an insoluble precipitate in the presence of some anions such as carbonate or phosphate. In addition, Burton [33] pointed out that the

physical removal of calcium, potassium, chlorine and all salts related is effective at a greater cost and using complex techniques such as membrane separation.

4.1.2. Horizontal Subsurface Flow Constructed Wetland

Temperatures of pig slurry reported during our study (from 18.1 °C to 19.8 °C) were adequate for nutrient removal inside the wetlands. Akratos and Tsihrintzis [42] indicated that removal efficiencies depend on temperature variations, where a temperature above 15 °C is adequate for organic matter, nitrogen and phosphorus removal.

The unplanted wetland achieved better results in salts removal than the planted one—this could be because of an evapotranspiration process, which is typical for the Mediterranean climate, triggering salt precipitation and accumulation [13]. Another factor that could influence the planted cell performance was the low biomass of the *S. vera* species, showing its inefficacy to uptake the salt concentration as expected.

The percentage of removal of over 90% of total suspended solids were in accordance with finding by Vymazal [19] treating sewage with constructed wetlands in an experience of 15 years of operation in the Czech Republic or similar to the results found by Caballero-Lajarín [7] purifying pig slurry in the Region of Murcia, Spain. Transport mechanisms create collisions between particles of suspended solids and binding mechanisms could retain TSS on the substrate [43]. In addition, the slope of the HSFCW promoted the sedimentation rate, as well as a filtration effect due to the narrow pores among particles.

Our results suggested that the optimal nitrogen removal could be due to the presence of the phytoextractors plants in W2 because they provide surfaces and oxygen for the growth of microorganisms in the rhizosphere, thus improving nitrification [44,45] and providing carbon from root exudates optimizing organic removal and the denitrification process [46–48]. Bachand and Horne [49] and Chung et al. [38] pointed out that denitrification is the major mechanism of total nitrogen removal in CWs. In addition, Huang et al. [18] verified that under mixed planting conditions, the lower relative abundance of anaerobes and the higher percentage of bacteria associated to the nitrogen metabolism could be the cause of nitrogen removal, therefore the combination of *S. vera* and *P. australis* in this study could be promoted nitrogen decrease in the treated pig slurry.

In agreement with our study, Tsalkatidou et al. [50] found higher percentages of reduction for BOD_5 in the effluent of CWs that were left unplanted, performing a combined stabilization pond-constructed wetland system in a pilot experiment carried out in Sindos near Thessalonica, Northern Greece. Polprasert and Kittipongvises [51] indicated that the main mechanism for BOD_5 and COD removal in constructed wetlands and storage ponds is due to a biodegradation reaction of bacteria, which explains the symbiotic reaction between plants and bacteria in CWs process and the symbiotic relationship between algae and bacteria in SP. A positive effect of plants on wastewater treatment was reported by Kaseva [45] assessing HF constructed wetlands in Tanzania—the COD concentration was reduced by around 33% in the unplanted cell and 56% and 60% in the planted ones. Likewise, better results were reported by Mbuligwe [52], where a planted cell with *Typha latifolia* and *Colocasia esculenta* was compared to an unplanted cell in a system treating anaerobically pretreated wastewater in Dar es Salaam, Tanzania. Therefore, the main mechanisms occurred in the planted cell relating to BOD_5 and COD reduction could be: sedimentation, filtration, hydrolysis, oxidation/reduction and bacterial metabolism (aerobic/anaerobic/anoxic) [51,53–55], however, to ensure which of them is more important, more in-depth studies should be carried out.

The percentages of phosphorus removal were greatly high with 95% and 97% for W1 and W2 respectively. Verma and Suthar [56] pointed out that the main parameters controlling the phosphorus decrease in CWs were redox potential, pH and temperature, while Vohla et al. [57] indicated that phosphorus is bound to gravel and sand substrates as a consequence of the precipitation reaction with calcium, aluminum, and iron in wetlands with a pH above 7. We considered that in our study the P reduction was mainly related to coprecipitation with Ca and Mg due to a limestone gravel bed. However, it has been reported other mechanisms of phosphorus removal in CWs:

adsorption, desorption, precipitation, dissolution, plant and microbial uptake, fragmentation, leaching, mineralization, sedimentation [56,58], therefore, other processes of P removal could be taking place in the CWs.

Regarding the removal of Fe, Mn, and Zn, the results from both cells (W1 and W2) were similar, with an important reduction in the effluent concentration of over 50%. The processes involved in metals removal included particulate settling, precipitation, adsorption (cation exchange) and plant uptake [53]. Higher removal rates in Zn were achieved by Gill et al. [54] and Terzakis et al. [55] for runoff wastewater treated with planted CWs in Ireland and Crete, respectively. In addition, Terzakis et al. [55] found a correlation between TSS and metals removal, explaining that the metals removed by wetlands were presumably due to those bound to solids which were either screened or settled in the systems, therefore we could infer in our study that this phenomenon could contribute to metals removal.

4.1.3. Storage Pond

The storage pond constitutes a supplementary module for wetland treatment which contributes to a better quality effluent. Based on our results (Tables 1 and 2) the planted and unplanted cells followed the same pattern, where a decrease in T, TSS, EC, TN, BOD_5, COD, TP and Mn, and an increase in pH was observed. Oppositely to our results, other studies [7,59] found that TSS tended to increase after the storage pond due to phytoplankton growth. The decrease produced in our study could be explained by the decrease in the temperature from 19.8 to 16.0 °C inhibiting microalgae development, however, this hypothesis must be tested. In this study, the percentage of TN removal in SP was slightly lower than that obtained by Caballero-Lajarín et al. [7], which was 60%.

As mentioned in the previous Section 4.1.2, BOD_5 removal depends on suspended bacteria growing in the pond water and biofilm bacteria developed on the pond side barrier. The growth of nitrifying bacteria (oxic conditions) and denitrifying bacteria (anoxic conditions) under suitable environmental conditions are the reactions in charge of nitrogen removal [51].

4.2. Microbial parameters

Regarding our microbiological results, they are comparable with those found by Massé et al. [60] who reported 99.7% of TC and *E. coli* removal when treating pig slurry with bioreactors. The elimination of pathogens could be favored by high pH when algal photosynthesis occurred, then grazing by zooplankton, sedimentation, and natural decay [51], in this study pH in the wetland ranged from 7.6 to 8.1. In addition, Ottová et al. [61] suggested that the potential for germ reduction in CWs is explained by complex mechanisms: physical (filtration, sedimentation, adsorption, and aggregation); biological (consumed by protozoa, lytic bacteria, bacteriophages, natural death); and chemical (oxidative damage, influence of toxins from other microorganisms and plants).

4.3. Treatment Efficiency

Purification efficiency varied from module to module in this study, but optimal efficiencies were achieved when pretreatment, HSFCW, and the storage pond worked as an integrated system. The press screw separator is a competent technique to separate the solid fraction (rich in dried matter and nutrients) and the liquid fraction (nutrients and scant dried matter) [62]; this technique proved beneficial in term of efficiency for TSS, TN, and Na for W1 and W2 and showed some success for removing TP, especially in W2.

There are different factors that contribute to CWs efficiency; including the design, type of substrate, plantation, and climate, among others. A diverse range of interactions involving the wastewater with substrate, microorganism, litter, plants, and atmosphere within the wetlands promote pollutant removal; accordingly, this dynamic has a significant influence on efficiency when the wastewater passes throughout the wetland. Nutrients removal depends on the season, consequently, vegetation uptake in temperate climates is maximum during spring, at moderate temperatures, but release via decomposition is maximum during fall, also at moderate temperatures [53]. This research was performed in spring,

therefore the temperature in the whole process ranged from 15 °C to 18 °C in W1 and 18 °C to 20 °C in W2 being adequate for an optimal plant development which improves the purification processes in the wetland.

Xu and Mills [63] reported average removal efficiency of a wetland for Cu and Zn, above 60%; these findings are in line with our results, over 60% for W1 (Table 1). Muñoz et al. [21] registered lower values when compared to the removal efficiencies of the outlet from wetlands planted with *P. australis* with a similar planting frame for EC, TSS, KN, NH_4^+, COD, TP, Cu and K^+.

In general, our integrated purification system seemed to achieve higher performance of purification, with the highest efficiency being observed for macronutrients (N and K) in the planted system (W2), while the similar pattern for TP (Figure 3) was found in both cells (W1 and W2).

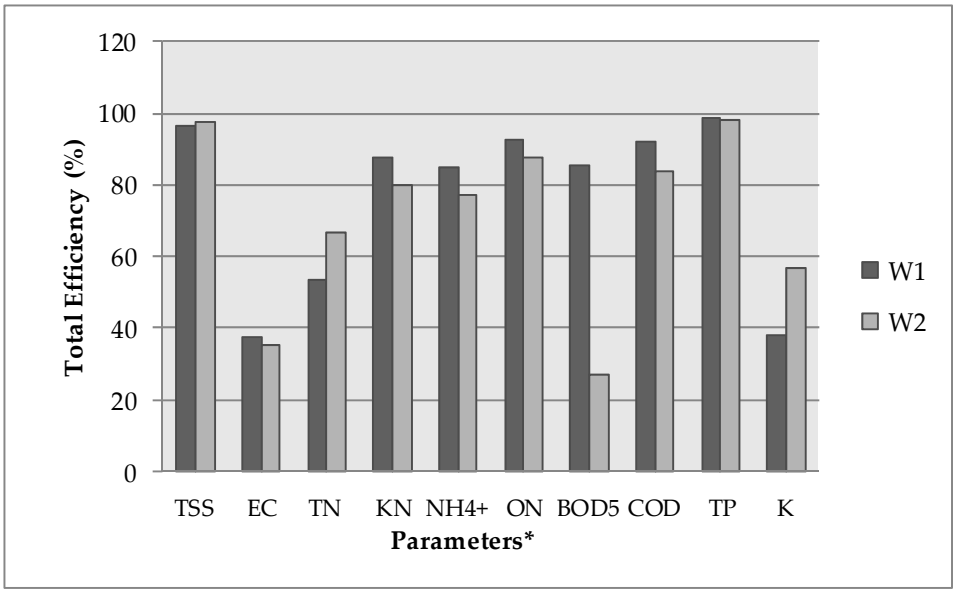

Figure 3. Total efficiency of the main parameters. **TSS* total suspended solids, *EC* electrical conductivity, BOD_5 biochemical oxygen demand *COD* chemical oxygen demand, *TN* total nitrogen, *KN* Kjeldahl nitrogen, *ON* organic nitrogen, *TP* Total phosphorous, *P* potassium.

5. Conclusions

Results showed that a high reduction of TSS was achieved during the pretreatment, around 20%–36%, in addition, after the wetlands the reduction was up to 93%. The mechanical separator removed part of the dried matter, decreasing TP and TN by around 13% and 11%, respectively. W1 showed better removal results than W2 in most of the analyzed parameters; however, W2 exhibited better results for macronutrients (N, P, and K), which are the most important for agricultural requirements, and therefore plants constituted an essential component in the HSFCWs when pig slurry is treated and played an important role in the purification process.

The majority of the microbial parameters were totally eliminated after the wetlands, especially in W2. The storage pond was shown to be quite effective in the biodegradation process due to microalgae activity complementing the wetland performance, thus optimizing the pig slurry quality.

Therefore, this study demonstrated that the combination of *P. australis* and *S. vera* improved the quality of the effluent, being a system that can be implemented as an economic alternative for farmers and whose purified pig slurry can be applied to croplands with a proper dose, which would mitigate the environmental risk of pollution.

However, this study suggests a need for further research on planted systems (preferably polyculture) to improve the removal of salt contents, including nitrates, or metals, which are the main potential pollutants that limit the use of treated pig slurry as a fertilizer.

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