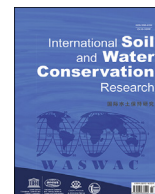




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Original Research Article

Performance of *Phragmites Australis* and *Cyperus Papyrus* in the treatment of municipal wastewater by vertical flow subsurface constructed wetlandsFernando García-Ávila ^{a,*}, Jhanina Patiño-Chávez ^a, Fanny Zhinín-Chimbo ^a, Silvana Donoso-Moscoso ^a, Lisveth Flores del Pino ^b, Alex Avilés-Añazco ^a^a Facultad de Ciencias Químicas, Universidad de Cuenca, Cuenca, 010107, Ecuador^b Facultad de Ciencias, Universidad Nacional Agraria La Molina, Lima, 12175, Peru

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ABSTRACT

The use of constructed wetlands to treat municipal wastewater reduces energy consumption and therefore economic costs, as well as reduces environmental pollution. The purpose of this study was to compare the purification capacity of domestic wastewater using two species of plants sown in subsurface constructed wetlands with vertical flow built on a small scale that received municipal wastewater with primary treatment. The species used were *Phragmites Australis* and *Cyperus Papyrus*. For this purpose, a constant flow of $0.6 \text{ m}^3 \text{ day}^{-1}$ was fed from the primary lagoon to each of the two wetlands built on a pilot scale with continuous flow. Each unit was filled with granite gravel in the lower part and with silicic sand in the upper part of different granulometry, the porosity of the medium was 0.34, with a retention time of 1.12 days and a hydraulic load rate of 0.2 m day^{-1} . To analyze the purification capacity of wastewater, physical, chemical and biological parameters were monitored during three months. Samples were taken at the entrance and exit in each experimental unit. The results obtained in the experimental tests for the two species of plants, indicated that the *Cyperus Papyrus* presented a greater capacity of pollutants removal as biochemical oxygen demand (80.69%), chemical oxygen demand (69.87%), ammoniacal nitrogen (69.69%), total phosphorus (50%), total coliforms (98.08%) and fecal coliforms (95.61%). In the case of *Phragmites Australis* retains more solids. The species with greater efficiency in the treatment of municipal wastewater for this study was *Cyperus Papyrus*.

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1. Introduction

Researches on the treatment of domestic wastewater through technologies that demand low costs, low energy and maintenance has been a priority in most countries around the world (Binder, Göttle, & Shuhuai, 2015; Laaffat, Aziz, Ouazzani, & Mandi, 2016). For the treatment of domestic wastewater, conventional treatments can be used in large populations and unconventional treatments in nuclei of small populations or in rural areas, where there is no sewerage service or buildings are widely dispersed (Tahir, Yasmin, & Hassan Khan, 2016; Zidan, El-Gamal, Rashed, & El-Hady Eid, 2015). Because it is a natural process, which has a low maintenance cost, and also avoids chemical treatment, constructed wetlands

(CW) have been increasingly recognized throughout the world in recent years (Mango, Makate, Tamene, Mponela, & Ndengu, 2017; Orimoloye, Kalumba, Mazinyo, & Nel, 2018; Talukdar & Pal, 2017; Vymazal, 2011). CW that try to mimic the layout and functions of natural wetlands have been widely used to improve water quality (Kyambadde, Gumaelius, & Dalhammar, 2004). CW have evolved over time adding the use of plants, assimilating them to natural wetlands, consists pollutants removal system through various natural processes, involving the operation of the support material or filter and the plant species. They are used throughout the world as a natural and economically favorable alternative to energy (Ansari & Golabi, 2018). CWs include biological, chemical and physical processes similar to the processes that occur in natural treatment wetlands (Stefanakis, Akrotos, & Tsihrintzis, 2014).

CWs can improve water quality by retaining or transforming constituents and have been used successfully to mitigate environmental pollution by eliminating a wide variety of wastewater

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pollutants such as organic compounds, suspended solids, pathogens, metals, colorants, pesticides and nutrients (Hansen, Kraus, Bachand, Horwath, & Bachand, 2018). Horizontal Subsurface Flow Constructed Wetlands (HSSFCW) have been used mainly to treat municipal wastewater (Zidan, El-Gamal, Rashed, & El-Hady Eid, 2015). CWs have been used and improved over time, using several types of plant species that adapt to the systems, flow types: horizontal, vertical or mixed, all carrying the same purpose that is the purification of wastewater (Sanmuga Priya & Senthamil Selvan, 2017; Stefanakis et al., 2014). Vegetation is an important factor affecting the effectiveness of Vertical Subsurface Flow Constructed Wetlands (VSSFCW) in the elimination of COD, BOD₅, TSS and NH₄ in all conditions tested (Abdelhakeem, Aboulroos, & Kamel, 2016; Coleman et al., 2001). Several species of plants sown in wetlands improve the elimination of COD throughout the year compared to the gravel only systems, especially at low temperatures (Abdelhakeem et al., 2016), works efficiently in tropical climates (Bohórquez, Paredes, & Arias, 2017).

It was found that the presence of vegetation causes small variations in the efficiency of elimination of chemical oxygen demand (BOD₅), suspended solids (SS), ammonia nitrogen (NH₃-N), total phosphorus (TP) and total nitrogen (TN) of livestock wastewater (Effendi, Widyatmoko, Utomo, & Pratiwi, 2018; Harmel et al., 2018). For all the above mentioned, the application of a VSSFCW was chosen on a pilot scale in the wastewater treatment plant (WWTP) “El Guabo” of the city of Santa Isabel, located in the province of Azuay-Ecuador. The choice of this type of subsurface flow wetland is due to the fact that they work with high rates of organic load, also during the operation of the system there were no presence of odors or pests. They have greater tolerance to cold, and possibly, a greater potential for assimilation per unit area than in surface flow systems (Pérez Villar, Dominguez, Hernandez, Sánchez, & Arteaga, 2012; Stefanakis et al., 2014). Common reed (*Phragmites Australis*) is the most widely used species in subsurface flow constructed wetlands (SSFCW) (Vymazal & Kropfelová, 2005; Vymazal & Kröpfelová, 2011), it has an appropriate efficacy in the elimination of microbiological parameters (Andreo-Martínez, García, Quesada, & Almela, 2017; Dadban Shahamat, Asgharnia, Kalankesh, & Hosanpour, 2018; Shahi et al., 2013).

The root structures of *Cyperus Papyrus* provided more microbial fixation sites, sufficient residence time of wastewater, entrapment and settlement of suspended particles, surface area for adsorption of contaminants, absorption, assimilation in plant tissues and oxygen for the oxidation of organic matter and inorganic in the rhizosphere. Which explains its high efficiency treatment, which is why it is a potential plant used to treat domestic wastewater (Kyambadde, Kansime, Gumaelius, & Dalhammar, 2004; Perbangkhem & Polprasert, 2010); *Cyperus Papyrus* has also been shown to reduce the content of phenols in wastewater (Vymazal, 2014). The hypothesis of this study, was that the wetland with *Phragmites Australis* would have greater removal of contaminants, compared to the *Cyperus Papyrus* wetland; since it has been reported a greater efficiency of *Phragmites Australis* (Zhang, Jinadasa, Gersberg, Liu, Jern, & Tan, 2014). The objective of this paper was to investigate the purification capacity of domestic wastewater using two species of vegetation (*Phragmites Australis* and *Cyperus Papyrus*) in pilot system of VSSFCW in the WWTP “El Guabo” and determine the improvement of water quality of download.

2. Materials and methods

2.1. Study site description

The experimental wetlands on the land next to the Wastewater

Treatment Plant “El Guabo” were implemented, located in the Santa Isabel city, Ecuador. The plant is located at the following geographic coordinates: Longitude 79.313732 °W and Latitude 3.298460 °S. This plant treats municipal wastewater through the lagooning system, which serves as the primary treatment. After the primary treatment, the water is conducted to the wetlands as a secondary treatment. Two experimental units were built, one for each species, *Phragmites Australis* and *Cyperus Papyrus*. Each wetland with its respective vegetation species operated independently. The flow was applied continuously in each wetland.

2.2. Design and constructed of vertical subsurface flow wetland

2.2.1. Sizing of the piloto wetland

The criteria for the SSFCW design were taken from Metcalf & Eddy (1991); Crites and Tchobanoglous (1998); Kadlec et al. (2000); García Serrano & Corzo Hernandez (2008); Abdelhakeem et al. (2016). To determine the size of the biological filtration system, the average temperature of the water (24.6 °C) was determined, thereby calculating the reaction rate constant, K_T (1.31 day⁻¹). Considering the initial concentration of BOD₅ (100 mg L⁻¹) entering the system, and the desired BOD₅ concentration in effluent (10 mg L⁻¹), the residence time of the water resulted in 3.25 days when the system operates intermittently and 1.12 days of retention when the system operates continuously.

The depth of the substrate, which can typically be 0.7 m. The deeper the substrate is, the greater the load that the system can process, but if the substrate is too deep, the bottom conditions become anaerobic and can result in reduced elimination of BOD₅. The effective porosity of the substrate (0.346 for sand and gravel) and the depth of the substrate (0.65 m taking as reference the greater root depth of the *Papyrus* plant) giving us as a result a final area of the 3 m² system necessary for the reduction of BOD₅. In Table 1, the design parameters of the pilot scale experimental units are presented.

The CWs operated with a HLR of 0.2 m d⁻¹ throughout the experiment. The granulometry of the material arranged in layers with their respective depths is shown in Table 2.

2.2.2. Construction of pilot-scale wetlands

For the construction of the wetlands, clearing work was initially carried out, that is, cleaning and extraction of the weeds from the site. Later the excavation was executed, in which the necessary area for the construction of the experimental units established in the design was delimited. In the surroundings, security channels were also built in order to prevent the entry of extra flows and materials carried during the rainy season. Then the wetlands were covered with high-density polyethylene (HDPE), to avoid drainage to the underground aquifer of 750 μm, which is 2.2 times cheaper than reinforced concrete (Stefanakis et al., 2014; Tsihrintzis, 2017). For the conduction of the treated water, PVC pipes of 50 mm diameter

Table 1
Data of the design parameters to implement the pilot-scale constructed wetlands.

Parameter	Value	Unit
Flow	0,6	m ³ d ⁻¹
Depth	0,65	m
Free edge	0,1	m
Area	3	m ⁻²
Long-wide relationship	3:1	
Long	3	m
Width	1	m
Time	1,12	día
Slope	1	%

Table 2
Depth of each layer of granulometric material.

Material	Effective size (mm)	Depth (cm)
Coarse sand	2	10
Fine sand	8	30
Fine gravel	16	7.5
Medium gravel	32	7.5
Coarse gravel	128	10

were used; the drainage system was also assembled with 50 mm PVC pipes and the necessary accessories such as elbows, tees and liquid solders; thus forming a circuit for water collection. Pipes were distributed throughout the base of the wetland, in the form of a fishbone (Stefanakis et al., 2014). In regard to aeration pipes, recommend installing 1 pipe for every 4 m^{-2} (Kadlec et al., 2000). At the bottom of the bed a drainage system for perforated pipes of \varnothing 50 mm and covered with coarse gravel was installed (Dąbrowski, Karolinczak, Gajewska, & Wojciechowska, 2017). A system of six vertical tubes connected to the drainage collector system ensured gravitational ventilation, with the objective of increasing the amount of oxygen to the wetland (Dąbrowski et al., 2017).

For the filling of the filtering medium, gravel and silicic sand available near the place of study were used. In the vegetation sowing, 12 plants of each species investigated for each wetland were used (Fig. 1). Both types of vegetation were planted with a density of 4 plants m^{-2} (Abou-Elela & Hellal, 2012; Brix & Arias, 2005; Ramprasada, Smithb, Memonc, & Philipa, 2017). Young plants developed of Phragmites Australis were extracted from the banks of the Minas River, close to the study site; while Young plants developed of Cyperus Papyrus were obtained from nurseries in the same zone, in such a way that a previous adaptation to the climate was not necessary.

Polyethylene hoses of 16 mm in diameter, were used for the continuous feeding of water to the wetlands. The same ones that were drilled in order to regulate the flow at each point of the wetland. The holes are located according to the surface area;

allowing to form a closed circuit and in turn distribute the liquid equally over the entire surface of the wetland.

2.3. Operation and monitoring of constructed wetlands

2.3.1. Operation of the VSSFCW

After building the VSSFCW and once planted Phragmites Australis and Cyperus Papyrus, the WCs were filled for a week. The flow of income to each wetland was taken from the lagoon exit of the primary treatment system. From the second week, it began with a continuous water supply and the flow was controlled through meters. In this growth stage of plants, periodic monitoring was carried out to verify the entry of water into the wetlands, observing the development of the plants and the purified effluent. Two months were expected in order to obtain a greater growth of the plants, after which, the monitoring stage of the influent and effluent of the wetland began for three months. After four months of planting, the plants reached a certain height. To avoid excessive accumulation of Cyperus Papyrus remains in the wetland, pruning was carried out. In the case of Phragmites Australis dried leaves were extracted.

The rainfall record in the study area was 561.4 mm of annual rainfall, distributed mostly in the months of January to May with 406.5 mm, which is equivalent to 72.4%. In this area there are two periods, one rainy from January to May and another dry from June to December. The experiment began in February and ended in July, that is, it was carried out during the rainy season.

2.3.2. Monitoring program

The monitoring allowed to evaluate the operation of the experimental units, identifying possible abnormalities, evidencing the fulfillment of the established objectives and checking if the physical, chemical and biological parameters in the effluent of the wetlands were within the maximum permissible limits established in the Ecuadorian environmental regulation. A period of two months was expected so that the plants could adapt to the new

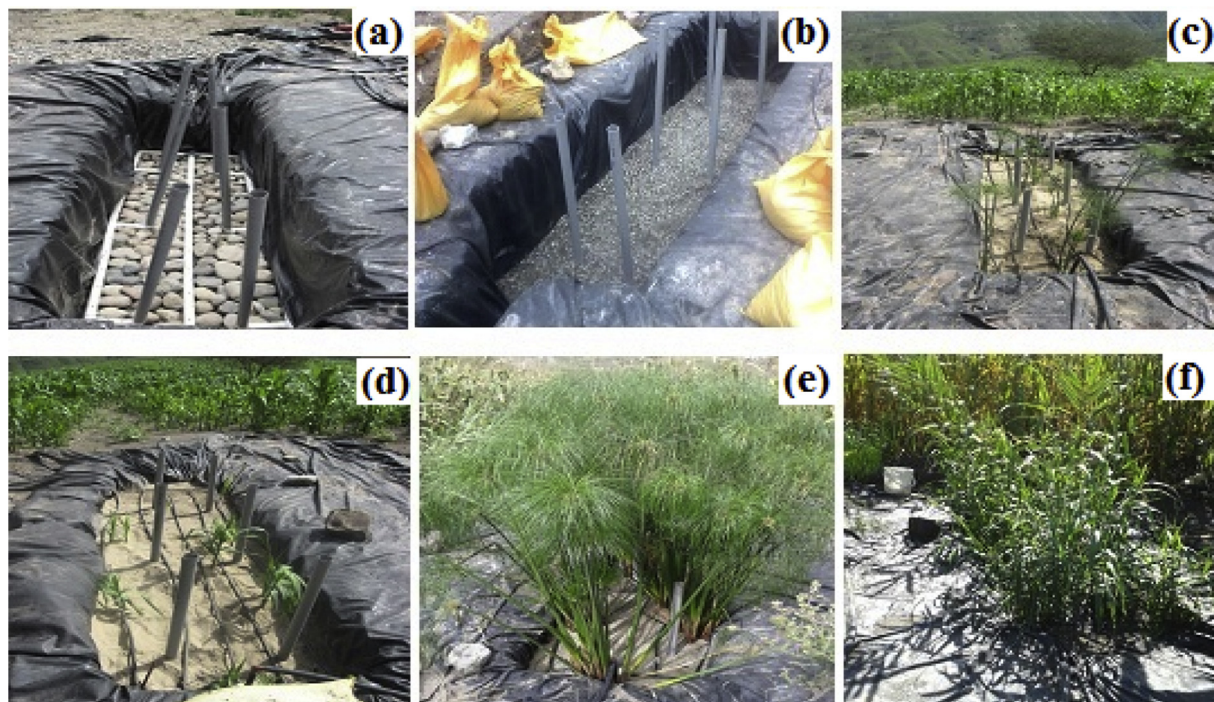


Fig. 1. Wetlands built to scale. (a) Coarse gravel placement, (b) Fine gravel placement, (c) Initial development of Cyperus Papyrus, (d) Initial development of Phragmites Australis, (e) Fourth month of Cyperus Papyrus development, (f) Fourth month of Phragmites Australis development.

environment of the CWs, until the plants reached a height of 0.5 m.

2.3.3. Location of sampling points and parameters analyzed

Three sampling points were identified: one point at the entrance of water to the wetlands (because the residual water that enters is the same for the two wetlands) and one point at the exit of each wetland (Fig. 2). The monitoring had a period of three months a fortnightly sampling was carried out. The following physical, chemical and microbiological parameters were analyzed: pH, Temperature, Total Alkalinity, Electric Conductivity, Total Suspended Solids, Total Dissolved Solids, Nitrates, Ammoniacal Nitrogen, Total Phosphorus, Biochemical Oxygen Demand, Chemical Oxygen Demand, Total Coliforms, Fecal Coliforms (*E. Coli*).

2.3.4. Data collection

The samples were kept at 4 °C and transported the same day for analysis to the Engineering Faculty Laboratory of the Cuenca University. Samples were collected in 2.5-L plastic bottles for physico-chemical parameters, and in 50-ml containers for microbiological parameters (García, Ramos, Pauta, & Quezada, 2018a; INEN 2176, 1998). Temperature, pH, and conductivity were measured in situ using a Hach Multiparameter model HQ40d. BOD₅ was quantified after five days of incubation at 20 °C with Oxytop head gas sensors

(WTW, Germany), COD was analyzed by the K₂Cr₂O₇ method according to APHA (1998). NH₄-N, NO₃-N and TP were analyzed with a GENESYS 10S UV–Vis Spectrophotometer following standard methods (APHA, 1998). Faecal coliforms (by membrane filter procedure), TSS and alkalinity as standard methods for water and wastewater examination (APHA, 1998) was analyzed.

2.4. Statistic analysis

Simple ANOVA was used for statistical analysis in order to determine significant statistical differences in the performance of wastewater treatment by wetlands between *Phragmites Australis* and *Cyperus Papyrus*. The variance analysis (ANOVA) was carried out using the InfoStat software package. A significance level of $p = 0.05$ was used.

3. Results and discussion

3.1. Results of applied wetlands

The average value of the parameters monitored biweekly for three months in the influent and effluent of the wetlands is presented in Table 3.

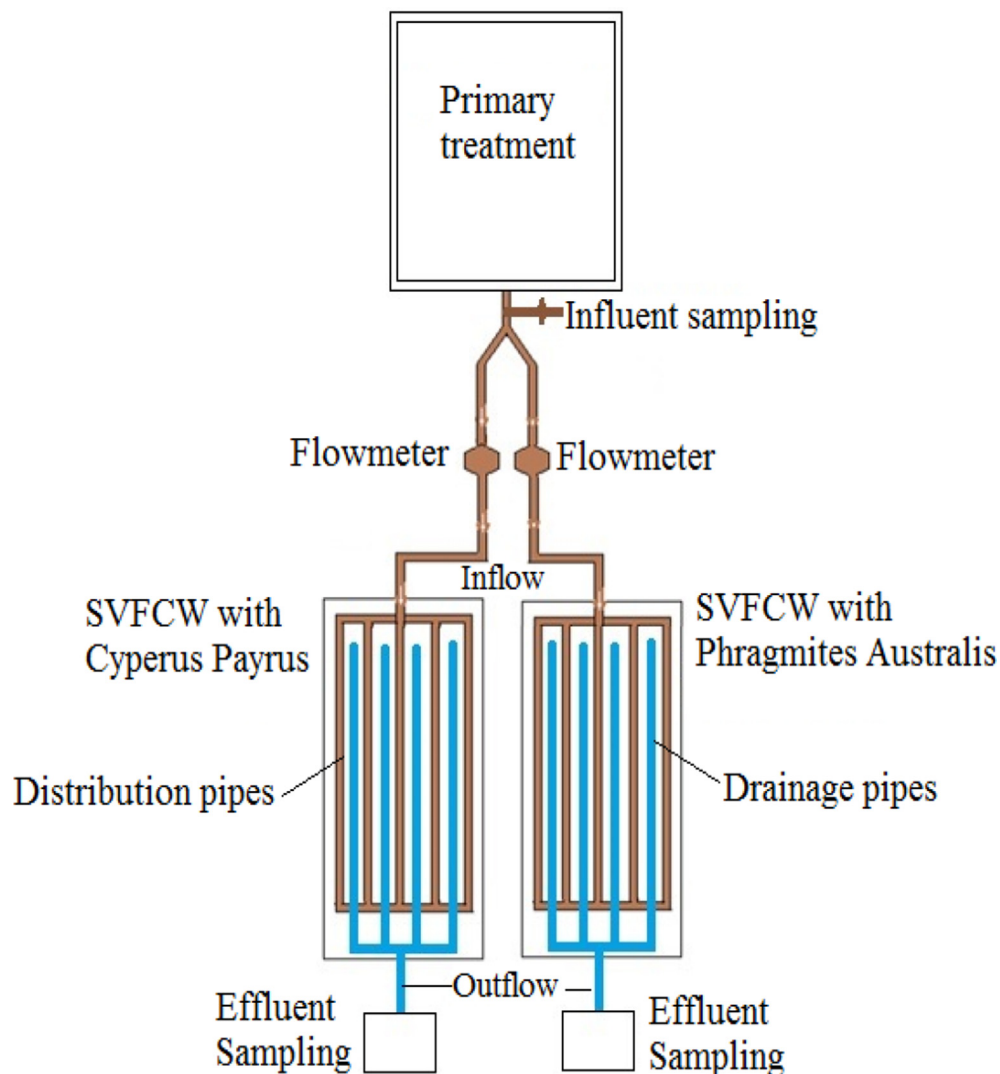


Fig. 2. Location of sampling points, water inlet and outlet pipes.

Table 3
Influent and Effluent wastewater characterization.

Parameter	Units	Influent	Effluent <i>Cyperus Papyrus</i>	Effluent <i>Phragmites Australis</i>
pH		6.95 ± 0.03	6.31 ± 0.1	6.21 ± 0.11
Temperature	°C	24.62 ± 0.55	24.37 ± 0.59	24.23 ± 0.54
Alkalinity	mg L ⁻¹ CaCO ₃	175.87 ± 11.93	68.8 ± 18.29	62.6 ± 12.12
Conductivity	μS cm ⁻¹	643.25 ± 27.23	633.67 ± 76.22	608.5 ± 44.2
Suspended solids	mg L ⁻¹	88.83 ± 14.74	60 ± 16.85	33 ± 9.54
BOD ₅	mg L ⁻¹	95.75 ± 13.18	18.49 ± 2.58	23.57 ± 5.25
COD	mg L ⁻¹	222.44 ± 16.99	67.02 ± 5.50	78.35 ± 4.75
Nitrates	mg L ⁻¹	0.84 ± 0.2	7.16 ± 1.41	8.35 ± 4.75
Ammoniacal Nitrogen	mg L ⁻¹	29.52 ± 2.56	8.95 ± 2.53	8.65 ± 1.98
Total Phosphorus	mg L ⁻¹	7.42 ± 1.09	3.71 ± 0.06	3.76 ± 0.19
Total Coliforms	MPN/100 ml	5.4E10 ± 2.22E9	1.038E9 ± 4.18E8	2.14E9 ± 5.3E8
Fecal Coliforms	MPN/100 ml	1.71E10 ± 4.72E9	7.52E8 ± 3.04E8	1.07E9 ± 4.1E8

3.2. Removal percentages obtained with the application of pilot wetlands

The average value of the purification efficiency of wastewater treated with *Phragmites Australis* and *Cyperus Papyrus* is presented in Table 4.

3.3. Role of hydraulic loading rate

To achieve an efficient pollutant removal rate (PRR), CWs require low HLR and longer hydraulic retention times (HRT) (Zhang et al., 2014). The European recommendations for HLR of VFCW for wastewater treatment are 50–60 mm d⁻¹ (Brix & Arias, 2005). Vegetation can increase nitrogen and phosphorus uptake by increasing HLR (eg, 18–68 mm d⁻¹), but decreases nutrient uptake if loading rates are too high (e.g., 135 mm d⁻¹) (Tripathi, Srivastava, & Misra, 1991). There is still discussion about the relationship between HLR and PRR, because there are no criteria that define the low and high values of HLR (Chang et al., 2007). Thus, Lin, Jing, Lee, and Wang (2002) considered a high HLR value of 0.135 m d⁻¹; while Lin et al. (2005) considered a high HLR value between 1.57 and 1.95 m d⁻¹. However, Schulz, Gelbrecht, and Rennert (2003) reported values up to 5410 m d⁻¹. Some variables such as the size and distribution of the medium; the growth rate of biofilm; the TSS load rate affect or determine the maximum HLR and therefore the existence of floods (Cooper, 2005). Weedon (2010) operated without floods in the range of 0.033–1.027 m d⁻¹, but was flooded at 1.27 m d⁻¹. On the other hand, Johansen, Brix, and Arias (2002) tested with HRL in the range of 0.20–1.20 m d⁻¹, but had floods at 0.80 m d⁻¹. Stefanakis and Tsihrintzis (2012) used three hydraulic load rates: 0.19, 0.26 and 0.44 m d⁻¹ in the long term in ten vertical flow wetlands, to treat synthetic wastewater. The authors showed that for each load increase, the systems achieved higher nitrogen removal performance. The high HRL and less retention times allow

Table 4
Comparison of percentages of purification.

Parameter	<i>Cyperus Papyrus</i> (%)	<i>Phragmites Australis</i> (%)
pH	9,25	10,69
Temperature	1,02	1,56
Alkalinity	60,87	64,40
Conductivity	1,49	5,40
Suspended solids	32,46	62,85
BOD ₅	80,69	75,39
COD	69,87	64,78
Nitrates	-88,67	-90,29
Ammoniacal Nitrogen	69,69	70,70
Total Phosphorus	50,00	49,38
Total Coliforms	98,08	96,02
Fecal Coliforms	95,61	93,74

the renewal of oxygen, allowing a better efficiency of the filter beds and long-term functioning (Molle, Liénard, Grasmick, & Iwema, 2006). According to the HRL values mentioned by these authors, the HRL value (0.2 m d⁻¹) used in this study is consistent with the HRL used in the aforementioned studies.

Table 5 shows the removal percentages of NH₄-N, TP, COD and BOD₅ for different HLR used in other studies. These HLR values are similar or close to the 0.2 m d⁻¹ value used in this study. It can be observed that the removal values of COD and BOD₅ (69.87%, 80.69%) obtained in this study for both types of vegetation were superior to the results obtained by Chang et al. (2007) and Yadav, Chazarenc, and Mutnuri (2018), who used HRL of 0.2 and 0.22 m d⁻¹ respectively. While, the NH₄-N removal values (69.69%) obtained in this study were lower than those reported by Chang et al. (2007), but higher than those reported by Yadav et al. (2018). Weedon (2010) using an HLR of 0.34 m d⁻¹ obtained higher PRR compared to this study. The removal percentages of NH₄-N, TP, COD and BOD₅ in this study were greater than those obtained by Yadav et al. (2018) when they tested an HLR of 0.15 m d⁻¹. The removal values of TP (50%) of this study were lower than those reported by Chang et al. (2007); but higher than those reported by Dąbrowski et al. (2017) and Maina, Mutua, and Oduor (2011) who tested with HLR of 0.1 and 0.174 m d⁻¹, respectively.

3.4. Results analysis

3.4.1. On-site parameters

The pH values shown in (Fig. 3a) indicate a slight decrease of the pH in the effluent with respect to the tributary, which may be related to the production of organic substances that acidify the medium, generated inside the wetland during the growth and death of the plant, and by the mineralization of organic matter (Coleman et al., 2001). These pH variations could be due, to that during the day (morning and afternoon) ammonia is produced as a product of the decomposition of nitrogen compounds, contributing to the increase in pH, but during the night, due to the release of carbon dioxide, the pH decreases. Therefore, the pH of the effluent can be acidic during the night and basic during the day (Vymazal, 2014); in this study all the samples were taken at times from 8 a.m. to 9am, so there are remnants of water that have rested at night. These results indicate the ability of macrophytes such as *Phragmites Australis* and *Cyperus Papyrus* to slightly modify the pH conditions in the rhizosphere, which are consistent with the results of Hussein and Scholz (2017) and Kyambadde et al. (2004) respectively. Nitrification can be considered as the main cause of the pH decrease in VFCW, because during this process H⁺ ions are released (Vymazal, 2007) and alkalinity is consumed (Paredes et al., 2007). The average value of the alkalinity in the influent was 175.84 mg L⁻¹ CaCO₃, reducing on average to 68.8 and 62.6 mg L⁻¹

Table 5
Percentages of contaminants removal for different HLR used in other studies.

HLR (m d^{-1})	$\text{NH}_4\text{-N}$ (%)	TP (%)	COD (%)	BOD_5 (%)	Author
0.2	91.77	71.79	36.46	47.5	Chang et al. (2007)
0.34	90.9	56.4	93.8	98.3	Weedon (2010)
0.012	95.1	85.1	92.4	99.6	Weedon (2010)
0.15	54		64	65	Yadav et al. (2018)
0.22	58		61	62	Yadav et al. (2018)
0.1	89.2	30.2	84.5	88.1	Dąbrowski et al. (2017)
0.16–0.32	88	92	68	72	Dan, Quang, Chiem, and Brix (2011)
0.174	97.8	47.1			Maina et al. (2011)

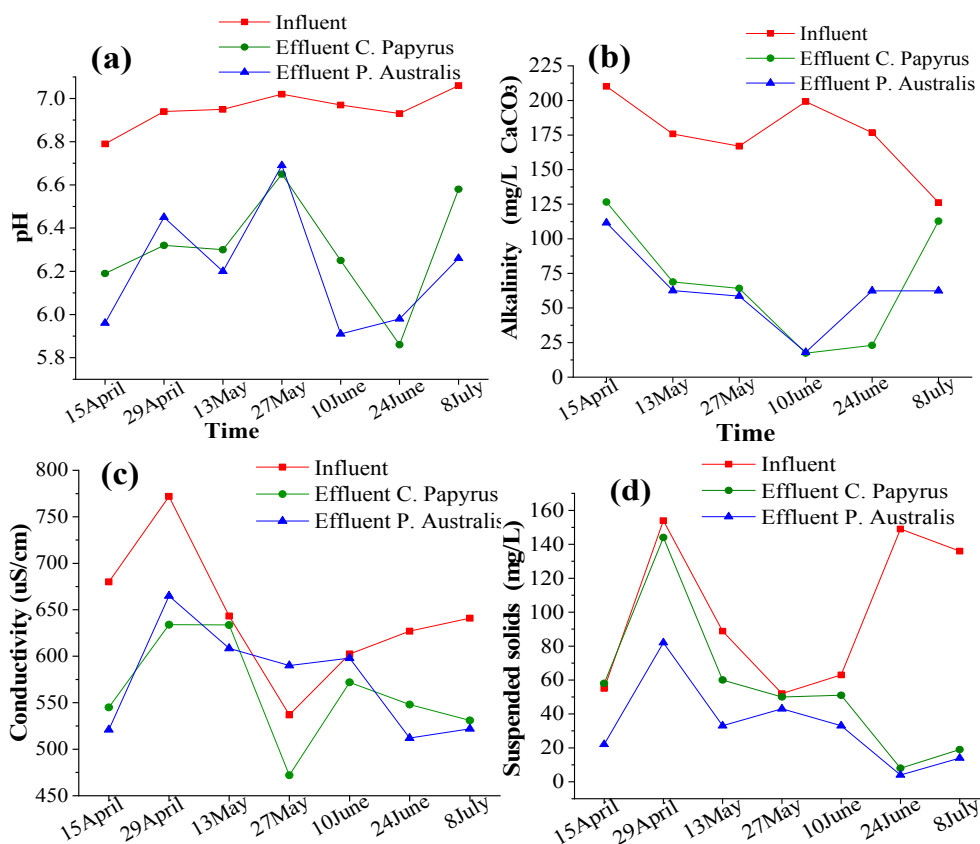


Fig. 3. Concentration of pH (a), alkalinity (b), electrical conductivity (c) and suspended solids (d) in the influent and the VSSFCW planted with *Cyperus Papyrus* and *Phragmites Australis*.

CaCO_3 for *Cyperus Papyrus* and *Phragmites Australis* respectively (Fig. 3b). The reduction of alkalinity justifies the reduction of pH (García-Ávila, Ramos-Fernández, & Zhindón-Arévalo, 2018b). In addition, reactions of degradation of organic matter where heterotrophic bacteria produce acetic acid, butyric acid and lactic acid can also explain the decreases in pH (Paredes et al., 2007).

The effluent temperature was between 21.8°C and 26.7°C ; this temperature is suitable for the efficient removal of organic matter (Bohórquez et al., 2017; Pérez Villar et al., 2012).

The conductivity is directly proportional to the concentration of dissolved solids, the conductivity values are slightly reduced both in the WC with *Cyperus Papyrus* and *Phragmites Australis* (Fig. 3c). Confirming that vertical flow wetlands have a relatively low capacity to eliminate conductivity (Bohórquez et al., 2017; Hussein & Scholz, 2017).

3.4.2. Totals suspended solids

With reference to totals suspended solids, the removal in the wetland with *Phragmites Australis* had a value of 62.85%, it was

higher with respect to the depuration value of the WC with *Cyperus Papyrus* of 32.46% (Fig. 3d).

According to Crites and Tchobanoglous (1998), solids in suspension mainly by physical processes are eliminated, such as sedimentation and filtration. Filtration occurs by impaction of particles in the roots and stems of macrophytes or in gravel particles in subsurface flow systems (Sanmuga Priya & Senthamil Selvan, 2017). The voids and media grain structure which is clearly visible in gravel, has remarkable impact on suspended solids and trapping during the flow path (Abdelhakeem et al., 2016; Knowles, Dotro, Nivala, & García, 2011; Tsihrintzis, 2017; Zidan et al., 2015).

In the elimination of TSS, according to the statistical analysis, there is no significant difference between the primary treatment and wastewater treatment with wetlands ($p > 0.05$).

3.4.3. Organic matter abatement

For the BOD_5 parameter, there was a reduction in the concentration in the two wetlands, the values of removal percentages in

BOD₅ are consistent with that reported by Vera, Martel, and Marquez (2013). However, it can be shown that *Cyperus Papyrus* showed a removal of 80.69% compared to 75.39% of *Phragmites Australis*, therefore *Cyperus Papyrus* has a higher efficiency than *Phragmites Australis* (Fig. 4a). Organic material biodegradation takes place in the biofilm (attached microbial population) along the plant roots and stems and the surface of the substrate grains, which allows to reduce the BOD₅ concentration (Stefanakis et al., 2014).

It is the same case, for chemical oxygen demand COD, *Cyperus Papyrus* presents a removal of 69.87% and *Phragmites Australis* 64.78% (Fig. 4b); therefore, with *Cyperus Papyrus* slightly higher removal yields are obtained. The results are a bit lower than the concentration reductions reported by Vymazal (2002) for HSSF-CW, and Abdelhakeem et al. (2016) for a VSSF-CW (88%). The transformation of the COD is essentially affected by the microorganisms whose presence and activity is carried out by the presence and processes mediated by the wetland plants (Barco & Borin, 2017; Bruch, Alewell, Hahn, & Hasselbach, 2014).

In the reduction of BOD₅ and COD, according to the statistical analysis, it was significantly different ($p < 0.05$) between the primary treatment and the water treatment with wetlands. In as much, that the treatment between *Phragmites Australis* and *Cyperus Papyrus*, shows that there is no significant difference between both ($p > 0.05$).

3.4.4. N and P concentrations and contents

The concentration of nitrites NO₂⁻ in the effluent was greater than in the influent for both cases (Fig. 5a). These data show that the concentration of NO₃⁻ in the effluent of the VSSF-CW seeded with the two species of plants increased slightly, producing negative removal percentages. This reflects that conditions existed for nitrification. These low results for the elimination of NO₃⁻ reflect the absence of favorable conditions for their elimination by the well aerated VSSF-CW wetland. Nitrates by reducing them to nitrogen gas due to the denitrification process are eliminated. This process occurs in the presence of an organic substance available only in anaerobic and anoxic conditions, where nitrogen is used as an electron acceptor instead of oxygen (Abdelhakeem et al., 2016; Kyambadde et al., 2004). The anaerobic conditions required for the initiation of NO₃⁻ reduction are not met in the VSSF-CW. These results are consistent with those obtained by Vymazal (2010) and Stefanakis et al. (2014), who found that constructed vertical flow wetlands successfully eliminate ammonia N, but very limited denitrification occurs. The VSSF-CW offers good oxygen requirements for the nitrification of NH₄ but unfavorable conditions for the denitrification of NO₃⁻. Permanently flooded wetlands have

higher denitrification rates (Hernandez & Mitsch, 2007), which is why VFSSCW have a low denitrification process. The absence of the denitrification process is the main obstacle in vertical flow wetlands to achieve greater nitrogen elimination (Abdelhakeem et al., 2016). Negative efficiencies were registered in the elimination of NO₃⁻, this was also reported by Tsihrintzis (2017). The increase of the NO₃⁻ levels indicated the nitrification of ammonium to nitrate, which could be favored by the lower concentrations of organic matter and the consequent greater growth of autotrophic bacteria (nitrifying). The system did not remove nitrates, probably because anaerobic conditions predominated in some system area.

For removal of ammoniacal nitrogen (NH₄-N), the average values of the percentages obtained in each wetland, provides the following results: for *Cyperus Papyrus* 69.69% and for *Phragmites Australis* 70.70% (Fig. 5b). In this case there is no significant difference between the two plant species, these results are similar to those obtained by Bohórquez et al. (2017); Barco and Borin (2017). These results confirm that the VFCW have been used mainly for the treatment of municipal and domestic wastewater, due to their greater capacity of nitrification, they have also been used for the treatment of other types of wastewater, mainly those with a high concentration of ammoniacal nitrogen (Stefanakis et al., 2014).

The Total phosphorus (TP) data obtained in the effluent show that the reduction of phosphorus is effective using both plants, observing very similar results between *Cyperus Papyrus* 50% and *Phragmites Australis* 49.38% (Fig. 5c).

These values are somewhat lower than those obtained by (Kyambadde et al., 2004; Vymazal, 2010). Phosphate removal efficiencies were significantly higher in surface flow constructed wetland (SFCW) than subsurface flow constructed wetland (SSFCW) (Hernández, Galindo-Zetina, & Hernández Hernández, 2017). The phosphorus transformations in the wetlands are the adsorption and absorption by the substrate, being the mechanism that governs the removal of this pollutant, without ruling out the chemical precipitation, fragmentation and leaching, mineralization and burial (Vymazal, 2011).

In the reduction of NH₄-N and TP, according to the statistical analysis, it was significantly different ($p < 0.05$) between the primary treatment and the water treatment with wetlands. In as much, that the treatment between *Phragmites Australis* and *Cyperus Papyrus*, shows that there is no significant difference between both ($p > 0.05$).

3.4.5. Microbiological analysis

In relation to total coliforms (TC), the percentages of removal of the wetland with *Cyperus Papyrus* were 98.08% and *Phragmites*

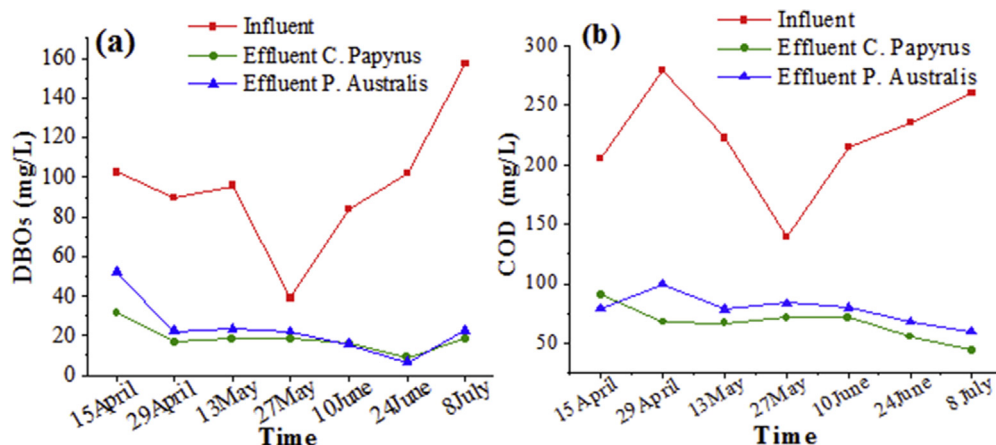


Fig. 4. Concentration of BOD₅ (a) and COD (b) in the influent and the VSSF-CW planted with *Cyperus Papyrus* and *Phragmites Australis*.

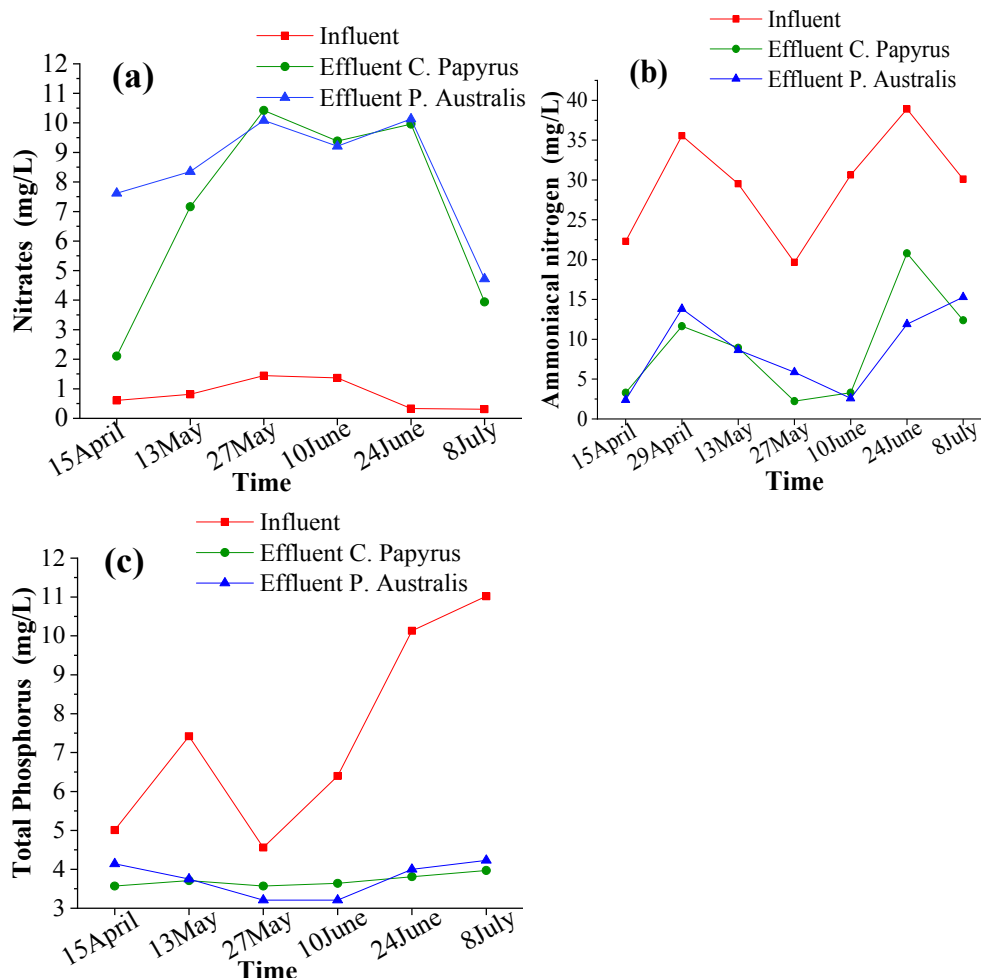


Fig. 5. Concentration of Nitrates (a), ammonia nitrogen (b) and total phosphorus (c) in the influent and the VSSF CW planted with *Cyperus Papyrus* and *Phragmites Australis*.

Australis 96.02%. Of the two species, the greatest removal of total coliforms was slightly greater with *Cyperus Papyrus* (Fig. 6a). The decline of the coliform population is due to filtration, sedimentation, adsorption, absorption of vegetation. In reference to fecal coliforms (FC), *Cyperus Papyrus* presents 95.61% and *Phragmites Australis* 93.74% removal (Fig. 6b), results consistent with that obtained by Kivaissi (2001). The CW are efficient and profitable systems to eliminate pathogens, the extraction mechanisms are not well understood, but may include: physical (filtration, sedimentation); chemical (oxidation, UV irradiation, adsorption to OM, etc.); and biological (degradation by other microorganisms, retention in biofilm, natural death, etc.) (Tsihrintzis, 2017).

The elimination efficiency of TC and FC was significantly different ($p < 0.05$) between the primary treatment and the treatment with wetlands. There is no significant difference between both wetlands ($p > 0.05$).

For compliance with Ecuadorian regulations, in the case of TC and FC does not comply with the regulations, due to the high microbial load in the tributary. However removal percentages greater than 90% are evident (Fig. 7). It is observed that for parameters pH, conductivity, total dissolved and suspended solids, BOD₅, COD, nitrates and total phosphorus if it complies with the Ecuadorian regulations.

It was obtained a significant reduction of pollutants in the effluent wastewater after eight weeks of operation of the vertical subsurface wetland. The percentages of reduction of all the physical-chemical and microbiological parameters are presented in

Fig. 7. The values obtained for all parameters are according to the Ecuadorian regulations for the disposal.

In this study, native species *Phragmites Australis* and *Cyperus Papyrus* tolerated very well the treatment conditions in the constructed wetlands, showing high growth and biomass, allowing the plants to reach their potential to improve the treatment.

In relation to BOD₅ and COD, *Cyperus Papyrus* showed greater removal compared to *Phragmites Australis*; discarding the hypothesis of this study, that the wetlands with *Phragmites Australis* would have greater efficiency than the wetlands with *Cyperus Papyrus*. This greater removal is due to the structure of the stems, the *Cyperus Papyrus* possesses the porous structure; while in the *Phragmites Australis*, the greater stems do not have parenchyma.

Due to the type of root, *Cyperus Papyrus* does not develop deep roots and forms a kind of networks that helps a greater coverage of the root area, which in turn allows a greater oxygenation of the system. In the *Phragmites Australis* case, the roots develop vertically, as they do not occupy a greater horizontal area, increasing the hydraulic conductivity compared to *Cyperus Papyrus*; what makes the purification in deeper layers therefore does not guarantee removal of contaminants due to the size of the granular material.

4. Conclusions

- The elimination efficiency of BOD₅, COD, total coliforms, fecal coliforms, ammonia nitrogen and phosphates was at 80.69, 69.87, 98.08, 95.61, 69.69 and 50.0% for *Cyperus Papyrus*

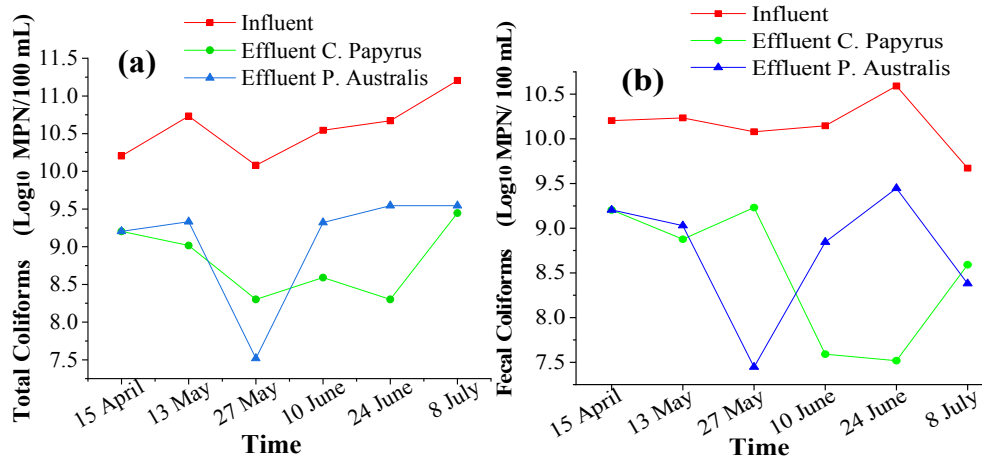


Fig. 6. Logarithm (base-10) of the most probable number (MPN/100 mL) of coliforms in the VSSFCW planted with *Cyperus Papyrus* and *Phragmites Australis*, determined in the influent and effluent for each treatment: (a) total coliforms, (b) fecal coliforms.

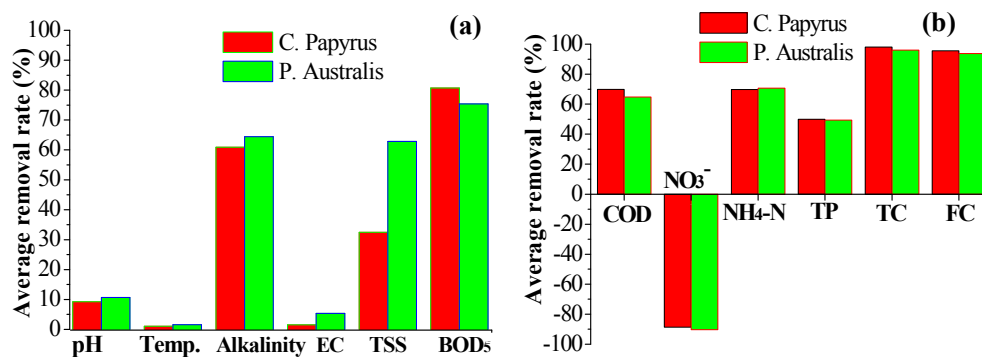


Fig. 7. Treatment performance of VSSFCW a) Percentages of pH removal, Temperature, Alkalinity, Conductivity, Suspended solids, BOD₅ b) Percentages of COD removal, Nitrates, Ammoniacal Nitrogen, Total Phosphorus, Total Coliforms, Fecal Coliforms.

elimination and 75.39, 64.78, 96.02, 93.74, 70.70 and 49.38% for *Phragmites Australis* respectively.

- *Cyperus Papyrus* was slightly more efficient in eliminating these parameters, while *Phragmites Australis* was more efficient in removing suspended solids. These results allow us to infer that *Cyperus Papyrus* could be a species of macrophyte ideal for wetlands built on a large scale, due to its high elimination efficiencies. The system did not remove nitrates, probably because anaerobic conditions predominated in some system area.
- According to the results obtained, the construction of VFCW planted with *Cyperus Papyrus* is recommended because it achieves high yields in the elimination of both physicochemical and biological pollutants present in urban/domestic wastewater especially for small communities.
- Another advantage of this plant species is that it can withstand temperatures between 20 and 33 °C, that is, it can adapt to warm zones; additionally by its capacity of reproduction, it does it in a shorter time in comparison to the *Phragmites Australis*, presents facility to carry out the pruning, and an important contribution for its landscape value. Under the conditions of this study, a slight disinfection is required to eliminate the residual pathogen in the treated effluent. No odors or insects were detected during the entire VSSFCW operating time.
- Effective solutions for wastewater treatment have been presented using promising VSSFCW technology planted with *Phragmites Australis* and *Cyperus Papyrus* that have low construction cost and low operating requirements, and has

application in the treatment of a variety of contaminants produced through various human activities.

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