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INFLUENCE OF SUBSTRATE AND SPECIES ARRANGEMENT OF CULTIVATED GRASSES ON THE EFFICIENCY OF HORIZONTAL SUBSURFACE FLOW CONSTRUCTED WETLANDS

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KEYWORDS

wastewater, elephant grass, Tifton 85 bermudagrass, plants, porosity.

ABSTRACT

We aimed to evaluate the efficiency of six different horizontal subsurface flow constructed wetlands (HSSF-CWs), with different substrates (gravel and crushed PET bottles), which also varied in relation to the presence and arrangement of plant species (elephant grass and Tifton 85 bermudagrass) in the removal of pollutants from a bulk milk cooling tank (MTWW). Each bed was fed at a flow rate of 0.18 m^3 d^{-1} and average organic load rate (OLR) of 318 kg ha^{-1} d^{-1} of BOD_5, with hydraulic detention time (HRT) of 1.84 days in the gravel-filled HSSF-CWs (CW_S-G) and 2.97 days in the PET-filled HSSF-CWs (CWs-P). The CWs-P were as efficient as the CWs-G in the removal of BOD_5, COD, Total-P, and K-Total, being in some cases even more effective (turbidity, TS, TSS and Na). The gravel, on the other hand, provided greater removals of Total-N from the MTWW. In the non-cultivated CWs and those cultivated with elephant grass, in its first half and Tifton 85 grass in its second half, there were higher average efficiencies in COD and TSS removal sand, in the latter, the highest average removal of Total-N.

INTRODUCTION

The bulk milk cooling tank, also known as the expansion tank, is an equipment that receives and stores milk in bulk (without the use of brass) and provides its direct cooling (Melo & Reis, 2007), as required by law. These tanks, located mostly in rural areas, are subject to the same sanitation operations that occur in the dairy industry, consuming large volumes of water, chemicals and, consequently, generating an effluent with polluting potential. As aggravating this effluent has been discarded without any previous treatment in water bodies. Therefore, the study of ways of treating this wastewater has become necessary.

In order to treat wastewater generated in a rural area that is difficult to access and does not have operators, it is necessary to use low cost installation and operation techniques and among these constructed wetlands systems stand out.

The Constructed Wetlands Systems (CW_S) can be a promising technical alternative, since it is a simple, low cost and easy operation and maintenance technology for the treatment of a wide variety of wastewater, such as domestic (Avelar et al., 2015), of dairy products (Matos et al., 2012; Mendonça et al., 2015), of swine farming (Sarmento et al., 2012; Fia et al., 2015), of textile industry (Saeed & Sun, 2013) and drugs industry (Zhang et al., 2014).

The CWs can be defined as reactors filled with porous materials that provide hydraulic conductivity and support for microbial growth and plant species. The system formed by the filtering substrate, the adhered biofilm and the plants, favors the degradation of part of the organic matter in the solution, the removal of sediment and suspended solids, of nutrients and other contaminants, through physical, chemical and biological processes, providing the debugging of wastewater (Prata et al., 2013). The removal efficiencies of the CWs depend on the choice of the type of liquid inlet and outlet configurations, feed rate, applied loads, environmental conditions, type of substrate, and the plant species used (Kantawanichkul & Wannasri, 2013; Wang et al., 2014a).

According to Dordio & Carvalho (2013), gravel, sand, gravel and soil are the materials most used as means of plant support, microorganism fixation and solid

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filtration. On the other hand, the placement of these filter media provides a fast clogging either by the size of their pores or by possible wear over the operating time (Pedescoll et al., 2009). Thus, the use of materials that are inert and easy to acquire, can become an interesting solution in terms of the associated costs (purchase and replacement). In this context, PET bottles (polyethylene terephthalate), whose destination has increasingly become an environmental liability, requiring recycling and reuse, have the potential to serve as an alternative means of support for HSSF-CWs. On the other hand, the availability of scientific articles related to their use is still little, therefor, requiring more studies to evaluate, in practice, the performance of CWs filled with this material.

As for plant species, several plants have already been used, from macrophytes present in aquatic environments, ornamental flowers and forage grasses (Chang et al., 2012; Zacarkim et al., 2014; Borges et al., 2015; Calheiros et al., 2015). The last ones are notable for their high extractive capacity, easy adaptation and rapid growth, such as the elephant grass (*Pennisetum purpureum* Schum) and Tifton 85 (*Cynodon* ssp.). As plant species have different nutrient requirements and environmental development conditions, intercropping can be another important variable to increase the efficiency of these systems. To make the technique even more attractive, grasses of interest were evaluated in the dairy chain, since they can be supplied for feeding dairy cattle.

In the light of the above, the objective of this study was to evaluate the extractive capacity of elephant grass, Napier cv. (*Pennisetum purpureum* Schum) and Tifton 85 grass (*Cynodon* spp), when treated in different ways in HSSF-CWs filled with different types of substrates, in the treatment of a community bulk milk cooling tank (MTWW).

MATERIAL AND METHODS

In order to carry out this study, gross wastewater generated in the process of cleaning the community bulk milk cooling tank was used, located in the municipality of Silveirânia-MG, located in Zona da Mata Mineira, where the pilot scale experiment was implemented and conducted. This wastewater is composed of milk residues, detergent-sanitizers, waste from bovine livestock (cleaning of brass) and sand.

The experimental infrastructure consisted of six HSSF- CW_s, in the dimensions of 0.6 m height x 1.0 m width x 2.5 m in length, positioned on the ground, having the bottom (level) and the sides waterproofed with 0.5 mm thick PVC tarpaulin, mounted parallel and delimited by masonry walls.

The distribution of the affluent was given at the central point at the entrance of each HSSF-CW_S through a ½-inch plastic tap where the flow control was made. The HSSF-CW_S effluent drainage system consisted of a 32 mm diameter PVC pipe, perforated, installed in the bottom of the exit zone. The height of the water level was 0.35 m, in both types of substrate (gravel # 0 and crushed PET bottles).

In Figure 1, a sketch is presented with the general scheme of the experiment.

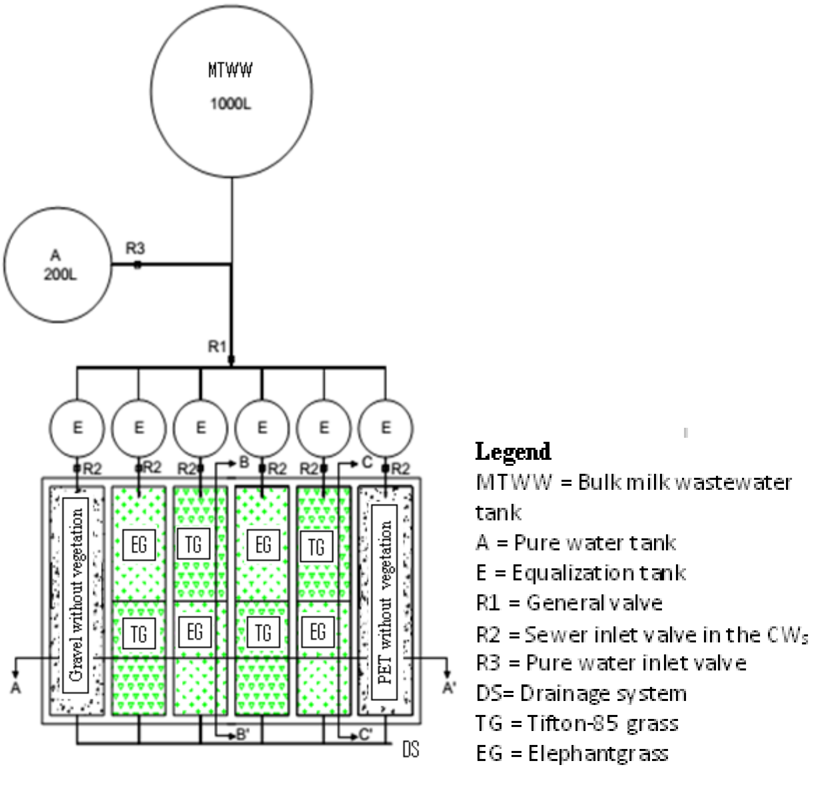


FIGURE 1. Sketch detailing the general scheme of the experiment.

The HSSF-CWs 1, 2 and 3 (CW_S-GWV, CW_S-GET and CW_S-GTE) were filled with gravel # 0 (D_{60} = 9.1 mm, coefficient of uniformity – CU D_{60}/D_{10} = 3.1 and initial void volume, n = 0.398 m^3 m^{-3}), while the HSSF-CWs 4, 5 and 6 (CW_S-PET, CW_S-PTE and CWs-PWV) were filled with PET bottles (250 and 500 mL) (n = 0.642 m^3 m^{-3}). Figure 2 shows the equipment used to knead the PET bottles and the material ready to be used as a substrate in the HSSF-CWs.

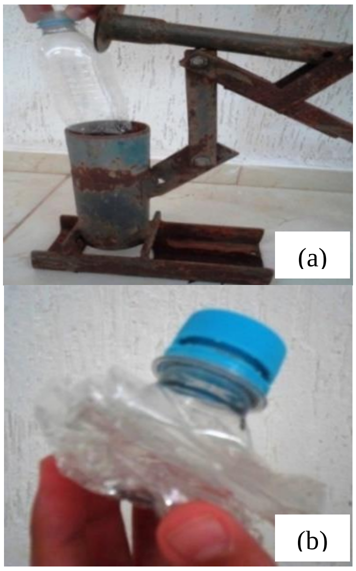


FIGURE 2. Equipment used to knead the bottles (a), bottle used as substrate (b).

To determine the porosity (n) of the medium consisting of the kneaded PET bottles, a glass vessel (aquarium-like) of known volume (Vr) was used, which was filled with the material, where, then, was added water, with the aid of a test tube, until filling the entire porous space of the material. With the required volume of water (Vw) to fill the container, the void ratio or porosity of the substrate was determined using [eq. (1)], and the value obtained was 64.22%. In relation to the gravel porosity # 0, the value obtained by Ferres et al. (2017).

$$n = {}^{Vw}/_{Vr} \tag{1}$$

In the HSSF-CWs where the substrate used was gravel # 0, the material depth was 0.45 m. In the HSSF-CWs whose substrate was composed of crushed PET bottles, the depth of the layer of this material was 0.35 m, being placed above that layer, another of 0.10 m of gravel # 3, to avoid that the bottles floated when the CWs-P were fed with MTWW. As the water level was maintained at 0.10 m below the surface of both HSSF-CWs (gravel# 0 and crushed PET bottles + gravel layer # 3), it is important to point out that even with this change in the disposal of the filling material, the waste water flow in the HSSF-CWs filled with PET bottles remained only by the layer of this material, so the wet height in both systems was 0.35 m. In Figure 3, an overview of the six HSSF-CWs used in this study.

FIGURE 3. Overview of the six HSSF-CW_S, three filled with PET bottles; and three filled with gravel # 0 (a) and after placement of the cover layer with gneissic gravel # 3 (b).

In CW_S-GET (2) and CW_S-PET (4) elephant grass was cultivated in the first half and Tifton grass 85 in the second half. The CW_S-GTE (3) and CW_S-PTE (5), in turn, were planted in reverse arrangement of the plant species, with Tifton 85 grass in the first half and elephant grass in the second half. In these units, the planting of each plant species with density of 19 propagules per square meter was carried out. On the other hand, the CW_S-GWV (1) and CW_S-PWV (6) were kept uncultivated.

Each bed was fed with 0.18 m^3 d^{-1} and average organic load (OLR) of 318 kg ha^{-1} d^{-1} of BOD_5, with hydraulic detention time of 1.84 days in the CWs filled with the gravel (CW_S-G) and 2.97 days in the CWs with PET (CW_S-P).

Evaporation water losses (CW_S-GWV and CW_S-PWV) and evapotranspiration (CW_S-GET, CW_S-GTE, CW_S-PET and CW_S-PTE) were calculated based on the difference between the inflow and effluent flows in the systems.

In the calculation of the efficiency in the removal of pollutants / nutrients was considered the evaporation / evapotranspiration in each of the HSSF-CWs, as presented in [eq. (2)].

$$EF = \frac{[(QAf \ . \ CAf) - (QAf - Ev) \ . \ CEf)]}{(QAf \ . \ CAf)} \tag{2}$$

Where,

EF = efficiency in removal of the pollutant (%);

C_{Af} = input concentration (variable unit);

Q_{Af} = affluent flow (m^3 d^{-1});

C_{Ef} = effluent concentration (variable unit);

Ev = evaporation / evapotranspiration in HSSF-CWs (m^3 d^{-1}).

To evaluate the performance of the systems, 12 samples of the wastewater and effluent from each HSSF-CW_S were carried out, measuring the electrical conductivity, turbidity and pH and quantifying the concentrations of BOD_5, COD, TSS, Total-N, Total-P, K and Na. The experiment was monitored for 8.5 months.

The procedure of collection, preservation of samples and analyzes was carried out according to the recommendations of the Standard *Methods for Examination of Water and Wastewater* (APHA, 2012) and Matos (2015). The analyzes were carried out in the Laboratory of Water Quality (LWQ), Department of Agricultural Engineering of the Federal University of Viçosa (UFV).

The efficiencies in the removal of the studied variables were analyzed statistically in the 2 x 3 factorial scheme, a total of 6 treatments. The first factor, support material, with two levels (gravel # 0 and crushed PET bottles) and the second factor, combination of cultivation, with three levels (without vegetation - WV, elephant grass and Tifton 85 - ET and Tifton 85 and elephant grass - TE). The type of design was randomized blocks (DRB) with the number of blocks for each variable as a function of the number of samples.

The means were submitted to Analysis of Variance (ANOVA, p = 0.05) and, when significant, the *Tukey* (p = 0.05) among averages; having a significant interaction between the factors, the unfolding was performed. For verifications of the normality and homogeneity of variance assumptions, it was applied the *Lilliefors* Test and the Tests of *Cochran Bartlett*, respectively. For the data processing and the statistical analyzes, the *software Assistat* Beta Version7.7.

RESULTS AND DISCUSSION

The mean values and standard deviation of the surface application rate of BOD_5, nitrogen (Total-N), phosphorus (P), potassium (K), sodium (Na) and total solids (TS) were applied during the monitoring period were, respectively, 318 kg ha^{-1} d^{-1}, 20 kg ha^{-1} d^{-1}, 7.7 kg ha^{-1} d^{-1}, 7.1 kg ha^{-1} d^{-1}, 8.2 kg ha^{-1} d^{-1} and 421 kg ha^{-1} d^{-1}.

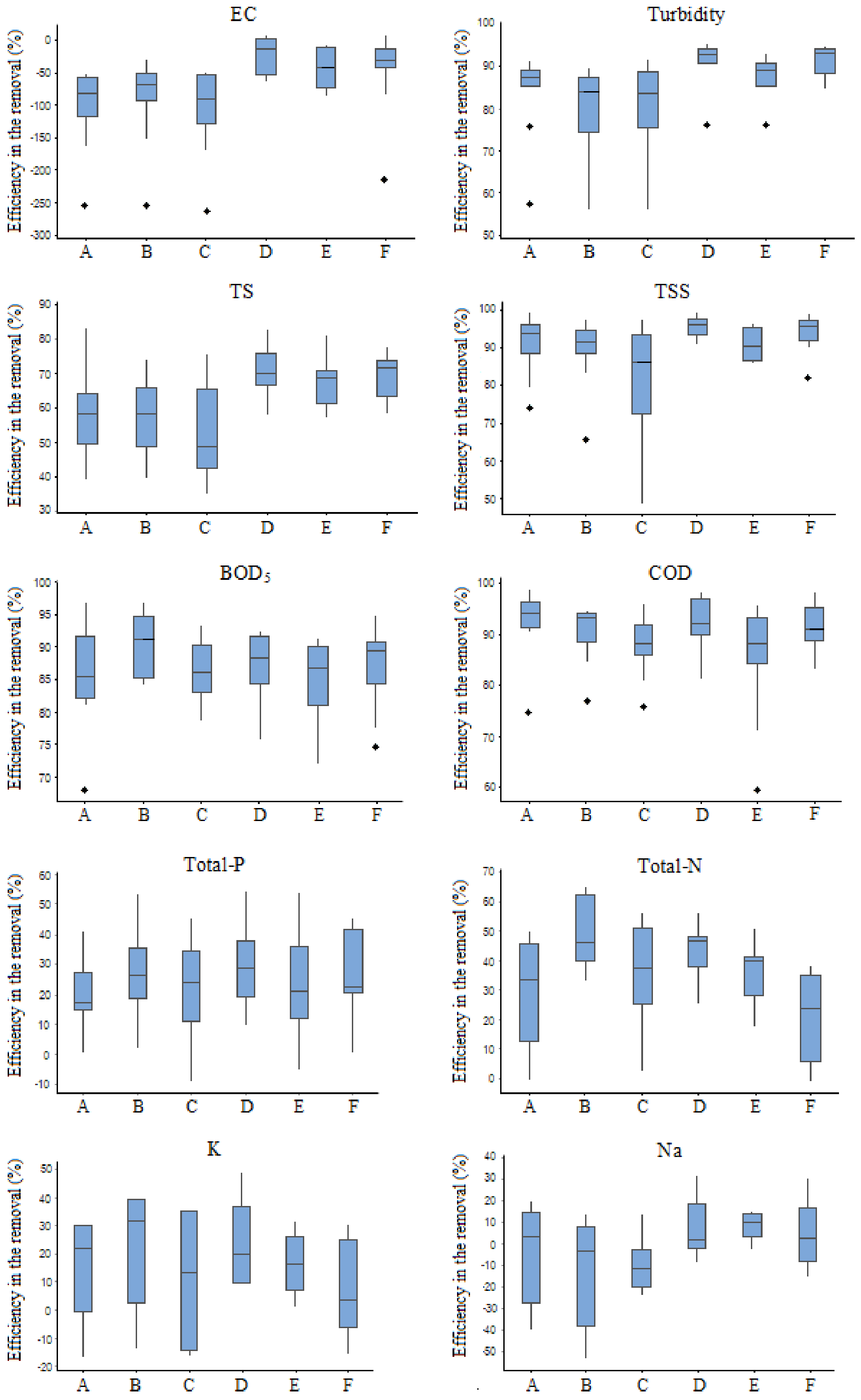
Table 1 shows the mean values of turbidity, BOD_5, COD, TS, TSS, Total-N, Total-P, K and Na and effluent and their removal efficiencies in HSSF-CWs.

TABLE 1. Mean values of turbidity, EC, BOD_5, COD, TS, TSS, Total-N, Total-P, K and Na affluent and effluent and the efficiencies in their removals in HSSF-CWs.

Treatment	**Turbidity (UNT)**			**EC (µS cm^{-1})**			**pH**		
	Afl.	**Efl.**	**Efi. (%)**	**Afl.**	**Efl.**	**Efi. (%)**	**Afl.**	**Efl.**	**Efi. (%)***
CW_S-GWV	125	20	84	217	416	-94	5.6	6.7	----
CW_S-GET	125	30	78	217	414	-79	5.6	6.7	----
CW_S-GTE	125	25	81	217	419	-98	5.6	6.7	----
CW_S-PET	125	11	91	217	272	-17	5.6	6.3	----
CW_S-PTE	125	16	87	217	296	-37	5.6	6.2	----
CW_S-PWV	125	11	91	217	298	-40	5.6	6.6	----
Treatment	**TS (mg L^{-1})**			**TSS (mg L^{-1})**			**BOD_5 (mg L^{-1})**		
	Afl.	**Efl.**	**Efi. (%)**	**Afl.**	**Efl.**	**Efi. (%)**	**Afl.**	**Efl.**	**Efi. (%)**
CW_S-GWV	570	250	58	201	15	91	403	63	87
CW_S-GET	570	259	57	201	26	90	403	70	87
CW_S-GTE	570	287	52	201	38	82	403	71	85
CW_S-PET	570	183	70	201	9	96	403	60	87
CW_S-PTE	570	200	67	201	19	91	403	85	82
CW_S-PWV	570	186	69	201	10	94	403	65	87
Treatment	**COD (mg L^{-1})**			**Total-N (mg L^{-1})**			**Total-P (mg L^{-1})**		
	Afl.	**Efl.**	**Efi. (%)**	**Afl.**	**Efl.**	**Efi. (%)**	**Afl.**	**Efl.**	**Efi. (%)**
CW_S-GWV	702	64	93	26	20	30	10	9	20
CW_S-GET	702	74	91	26	15	50	10	8	27
CW_S-GTE	702	108	88	26	18	38	10	9	22
CW_S-PET	702	79	92	26	17	43	10	8	29
CW_S-PTE	702	133	86	26	18	37	10	9	23
CW_S-PWV	702	83	92	26	23	21	10	8	27
Treatment	**K-Total (mg L^{-1})**			**Na-Total (mg L^{-1})**					
	Afl.	**Efl.**	**Efi. (%)**	**Afl.**	**Efl.**	**Efi. (%)**			
CW_S-GWV	11	10	15	12	14	6			
CW_S-GET	11	9	23	12	15	-14			
CW_S-GTE	11	11	11	12	15	-12			
CW_S-PET	11	10	23	12	13	8			
CW_S-PTE	11	10	16	12	12	-3			
CW_S-PWV	11	11	7	12	13	6			

In which, Afl. is the measure in the affluent, Efl. in the effluent (after treatment), and Efi. (%), the percentage of removal.

Variations in the removal efficiencies of the analyzed variables, over the period of experimental monitoring, are presented in Figure 4.



Where, A: CWs-GWV; B: CWs- GET; C: CWs-GTE; D: CWs-PET; E: CWs-PTE; and F: CWs-PWV.

FIGURE 4. Variation of efficiency in the removal of EC, Turbidity, TS, TSS, BOD_5, COD, Total-P, Total-N, K and Na, over the period of experimental monitoring.

The results of the statistical analysis of the mean efficiencies in the removal of the studied variables related to the type of support material and the combination of culture in the HSSF-CW_S are presented in Table 2.

TABLE 2. Average efficiency in the removal of BOD_5, COD, turbidity, EC, TS, TSS, Total-P, Total-N, K and Na in the different material supports and crop combination.

*Factor	Factor level	Efficiency in removal(%)				
		EC	**Turbidity**	**TS**	**TSS**	**COD**
Substrate	**G**	-92b	81b	56b	88b	91a
	P	-32.0a	90a	69a	94a	90a
Combination of cultivation	**WV**	-67.5a	88a	63a	93a	92a
	ET	-57.0a	85a	64a	93a	91a
	TE	-63.0a	84a	60b	87b	86b
		BOD_5	**Total-P**	**Total-N**	**K**	**Na**
Substrate	**G**	86a	23a	39a	16a	-6b
	P	85a	26a	34b	15a	4a
Combination of cultivation	**WV**	87a	24a	26c	11a	6a
	ET	87a	28a	47a	23a	-3a
	TE	84a	23a	38b	14a	-8a

(ET) (cultivated in the first half with elephant grass and in the second half with Tifton grass 85), TE (cultivated in the first half with Tifton 85 grass, and in the second half with elephant grass).

* Means followed by the same lowercase letter, vertical, and upper case, horizontal, do not differ statistically by the Tukey test, at a level of 5% significance.

The pH values in the effluents were between 6.2 and 6.7. Matos et al. (2012) found mean values of pH between 6.6 and 6.9, evaluating HSSF-CW$_S$ filled with gravel and cultivated with Tifton 85 grass and elephant grass, in the treatment of MTWW (pH affluent of 3.7 to 4.9). The different crop combinations influenced the pH values of the effluent only in the CWs-P, being the highest values obtained in the CWs-PWV. The release of acids by plants (Pavinato & Rosolem, 2008), with the purpose of mineralizing organic compounds, and the greater removal of nutrients by plants, reducing to a lesser magnitude of the H^+ ion activity, may explain the fact that pH was higher in the outflow of non-vegetated CW$_S$ (Matos, 2015). Regarding the different media, the mean pH values were higher in the CWs-G in all crop combinations. As described by Fia et al. (2010), the release of some cations from the gravel material used as support material resulted in a decrease in the activity of the H^+ ions in the medium, an unexpected reaction in a chemically inert carrier medium (PET bottle). This justifies, in part, the differences obtained in the effluent pH values of the HSSF-CWs filled with the different support materials.

The average efficiency of the removal of EC showed significant differences for different support media, and the gravel provided higher effluent values (mean negative efficiencies of greater absolute value) than those obtained in CWs-P. On the other hand, the different crop combinations had no effect on the average efficiency in EC removal from MTWW. This evidence corroborates the hypothesis of release of ions in solution in a medium containing gravel, which also influenced the pH variable. As discussed, gneissic gravel can release ions into the system, as described by Fia et al. (2010), which is no longer expected in the carrier material made up of PET bottles, since they are chemically inert materials.

The mean values of turbidity in the effluent and the efficiency in its removal varied, respectively, from 11 to 30 NTU and 78 to 91%. Colares & Sandri (2013) evaluated the efficiency of HSSF-CWs cultivated with *Typha sp* and obtained turbidity removal efficiency of 66.8%, when the support medium was natural gravel, 72% with gravel # 2 and 71% when washed gravel was used.

There was a statistical difference between treatments in turbidity removal, with the best results obtained in CWs-P (filled with PET bottles). According to Fia et al. (2010), gravel, as a chemically reactive material, wears out over time with release of solids in wastewater under treatment, which explains the higher efficiencies in CWs-P in this variable. In addition, the higher HRT, Table 1, in the CWs-P, may have provided greater solids removal, since lower velocity in the medium allows for greater sedimentation of solids in the treated wastewater, which would also explain the best results achieved in the filled with PET bottles.

The mean values of the concentration of BOD_5 in the effluent and of the efficiencies in its removal varied, respectively, from 60 to 85 mg $L^{-1}$and from 82 to 87%, while those of COD were between 64 to 133 mg L^{-1} and 88 to 93%. Chagas et al. (2013), used gravel #0 in HSSF-CW$_S$ cultivated in yellow-lily (HRT of 3.9 days), obtaining 90.1% efficiency in the removal of BOD_5 sanitary sewage and Çakir et al. (2015) evaluating the performance of HSSF-CWs cultivated with reeds, in the treatment of domestic sewage, obtained removal efficiencies of 49.1 and 64.9% of BOD. Matos et al. (2012), evaluating HSSF-CWs used in the treatment of MTWW, with OLRs between 66 and 570 kg ha^{-1} $d^{-1}BOD_5$, HRT of 4.8 days, cultivated with Tifton 85 grass and elephant grass, obtained efficiencies in the removal of BOD_5in the range of 79 to 96%.

Akratos & Tsihrintzis (2007), evaluating HSSF-CWs cultivated with cattail, filled with gneissic gravel # 0, obtained efficiencies in the removal of BOD_5 of 88%, 94%, 91% and 90%, with HRT of 6, 8, 14 and 20 days, respectively, and suggested that the higher the HRT the greater the removal capacity of BOD_5 of wastewater under treatment in CWs. However, in this study, the different HRT in CWs-G and CW$_S$-P did not influence the efficiency in the BOD_5 removal, since there was no significant difference in the efficiency in the BOD_5 removal.

When comparing different crop combinations, combinations WV (no vegetation) and ET (elephant grass followed by Tifton 85) provided COD removals higher than those obtained by TE combination (Tifton 85 grass and elephant, in sequence), which are assumed to be associated with the root type of the cultivated plant species. Tifton 85 grass has a deeper root, with vertical growth, and elephant grass, the shortest and most superficial root, presenting horizontal growth (Matos et al., 2010a). Thus, it is speculated that when Tifton 85 grass was cultivated in the second half of the CWs, it provided the formation of a more efficient physical barrier in the removal of the particulate organic matter, also reducing the passage of solids released by the plant itself. In these conditions, it would also justify the reason why the CW_S without vegetation (absence of vegetal debris) is as efficient as the CW_S with ET.

The mean values of TS concentration in the effluent and efficiency in its removal were respectively between 186 to 287 mg L^{-1} and 52 to 70% and, of TSS, of 9 to 38 mg L^{-1} and 82 to 96%. The crushed PET bottles provided mean efficiencies higher than those obtained by the gravel support medium, which can be explained by the wear suffered by the gravel and the higher HRT in the CWs-P, as already explained for the variable turbidity.

Corroborating with the observed trend for COD removal, the mean efficiencies in TS and TSS removal in the WV and ET crop combinations were higher than those obtained in the TE combination, which, however, were equal to each other. Thus, the hypothesis of the importance of filtration in the root environment is reinforced.

The mean concentrations of Total-P in the effluent and the efficiency in its removal varied, respectively, from 8 to 9 mg L^{-1} and 20 to 29%. The different media and crop combination of the treatments did not interfere in the removal of Total-P. Mendonça et al. (2012) verified that there was no significant difference between the cultivated and non-cultivated HSSF-CWs in the removal of phosphorus, with average efficiencies varying from 19 to 34%. Amorim et al. (2015), worked with loading rates of P higher than those evaluated in this study, between 13 and 240 kg ha^{-1} d^{-1}, obtained efficiency in the removal of P, which varied between 20 and 30%.

There are no limits of phosphorus concentration in effluents to be released in water bodies, in the Brazilian environmental legislation, therefore, there is nothing to compare in relation to this. But it can be said that HSSF-CWs, in general, presented satisfactory removal efficiencies of P from MTWW, when comparing the values to those obtained in other processes and systems used in the treatment of wastewater.

In the effluent the mean values of Total-N concentration and efficiency in their removal varied, respectively, from 15 to 23 mg $L^{-1}$and from 21 to 50%. CWs-G provided higher mean efficiencies in Total-N removal than the CWs-P.

Higher N concentrations in the dry matter of the aerial part of the grasses and nitrogen extraction capacity via aerial part were obtained in the CWs filled with gravel, which is an indication of the possibility of obtaining better efficiencies in the removal of N from MTWW in these systems. Another plausible explanation is that the lower porosity of the CW_S-G has provided less redox potential, and possible greater denitrification, compared to the PET-filled bed.

The ET crop combination presented an average higher than those obtained in the WV and TE combinations. This result can be explained by the differences between the roots of the plant species, as already explained for the differences obtained in the crop combination in the removal of COD. This time, the planted unit differed from the uncultivated bed, due to the presence of plant species contributing to nutrient extraction.

In view of this, it is clear the importance of the correct choice of plant species and, in the case of polyculture, the positioning of cultivation within the system. More resistant species, superficial roots, with greater capacity of insertion of oxygen in the bed (through structures like aerenchyma) and more resistant to high organic loads, should be cultivated in the initial part of the CWs, being a predominantly anaerobic zone. Already species with greater need of nutrients, should be positioned after the region of more intense organic matter removal, being able to associate other species for reduction of coliforms, for example. Some studies have shown this positive relationship between plant diversity and the system’s ability to remove pollutants, such as those conducted by Kouki et al. (2012); Turkera et al. (2013) and Wang et al. (2014b).

According to the literature review by Vymazal (2007), the mean efficiencies in nitrogen removals are between 40 and 55%, when loads of 6.8 and 17 kg ha^{-1} d^{-1} are applied of N in different configurations of constructed wetlands systems, vertical and horizontal. Thus, having an average load of 20 kg ha^{-1} d^{-1} of N, with removals varying from 21 to 50%, the results were close to those commonly found.

The mean values of K concentration in the effluent and removal efficiency varied, respectively, from 9 to 11 mg L^{-1} and of 7 to 23% and; to Na, the values, in the same sequence, ranged from 12 to 15 mg L^{-1} and -14 to 8% (CW_S-PET).

Matos et al. (2010b) treating swine wastewater (SWW) in HSSF-CWs cultivated with alternanthera, cattail and Tifton 85 grass and in uncultivated CW_S, which received loads of 36.3 kg ha^{-1} d^{-1} of K and 11.3 kg ha^{-1} d^{-1} of Na, resulted in removal efficiencies of K between 29 and 46% and Na between 18 and 28%, values higher than those obtained in this study. Fia et al. (2015) obtained efficiencies in the removal of 15 to 27% of K and 2 to 24% of Na in the treatment of swine wastewater (SWW) in HSSF-CWs cultivated with cattail and Tifton 85 grass, when applying loads between 21 to 80 kg ha^{-1} d^{-1} of K and 15 to 56 kg ha^{-1} d^{-1} of Na. It is observed, that there are species with greater potential of removal of certain nutrients.

Fia et al. (2015) studied the influence of nutrient load and cultivated species on the removal of K, Na, Cu and Zn in the treatment of swine wastewater (SWW) in HSSF-CWs cultivated with cattail and Tifton 85 grass, and verified that greater removal of this element was obtained in treatments which received lower loads, corroborating what was observed in this study.

Different support materials and cultivation combinations did not influence the removal of K. Already, in the Na removal, the support medium consisting of crushed PET bottles provided mean efficiencies higher than those obtained by the gravel support medium and the different vegetal combinations did not influence the removal of this variable.

It is observed that the efficiencies provided by the evaluated treatments were low, in relation to the studies demonstrated. However, the applied load should be taken into account, which was higher than those that generated the best results, the HRT and the evaluated cultures. Since Na and K are highly soluble in water, there is the possibility of adsorption to the carrier medium, until saturation or incorporation into the biofilm occurs. However, given the great mobility, the absorption seems to be the most sensitive factor to the change in the concentration of chemical elements in the effluent.

Finally, it is interesting to use PET bottles as a support medium in HSSF-CW$_S$, providing efficiencies similar to that beds filled with gravel, and with less risk of wear over the operating time, which could generate solids of clogging.

CONCLUSIONS

The substrate composed of crushed PET bottles was as efficient as gravel in the removal of some variables (BOD$_5$, COD, Total-P and Total-K) being, in some situations, even more effective (turbidity, TS, TSS and Na). Gravel, on the other hand, provided greater removals of Total-N from MTWW.

In the HSSF-CWs without culture and those cultivated with elephant grass in its first half and Tifton 85 grass in its second half, there were higher mean efficiencies in COD and TSS removals, and in the latter, higher mean removal of Total-N. Thus, the use of PET bottles as a support medium has shown promising, and that the choice and disposal of plant species can provide gain in removal efficiency.

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