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ABSTRACT

## EXTRACTION CAPACITY OF GRASSES GROWN IN CONSTRUCTED WETLAND SYSTEMS USING DIFFERENT ARRANGEMENTS AND SUBSTRATES

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## **KEYWORDS**

wastewater, forage, nutrients, built wetlands. Constructed wetland systems have been used to treat different wastewater; among their essential components are the cultivated plant species and type of substrate used to fill them. The choice of plant species and type of substrate are important for the good system performance in wastewater treatment. This study aimed to evaluate the extraction capacity of Napier and Tifton 85 grasses when cultivated at different positions in horizontal subsurface flow constructed wetlands (HSSF–CWs) filled with different substrates in wastewater treatment from a bulk milk-cooling tank (MTWW). The experimental unit consisted of four HSSF–CWs, in which an average surface organic loading rate of 318 kg ha<sup>-1</sup> d<sup>-1</sup> of BOD<sub>5,20</sub> was imposed and a hydraulic holding time of 1.8 and 3.0 d, respectively, in units filled with gravel or crushed PET bottles. Tifton 85 grass presented the highest Na extraction capacity, with better results obtained in HSSF–CWs filled with crushed PET bottles, while Napier grass was more effective in extracting N, K, and P from MTWW. When cultivated in the second half of gravel-filled HSSF–CWs, both grasses provided higher average N extractions when compared to those filled with crushed PET bottles, which had higher extractions in the first half of HSSF–CWs.

#### INTRODUCTION

In recent decades, there has been a growing interest in constructed wetlands (CWs) systems, as they are simple, low cost, and easy to operate and maintain for treating a wide range of wastewater, such as domestic (Avelar et al., 2015), dairy products (Matos et al., 2012; Mendonça et al., 2015), pig farming (Fia et al., 2015), textile industry (Saeed & Sun, 2013), and compounds from pharmaceutical industries (Zhang et al., 2014).

These systems consist of a filtering medium (substrate), growth biofilm adhered to the filling material, and plants. Together, these components of CWs favor the degradation of part of the organic matter in solution, removal of sedimentable and suspended solids, nutrients, and other contaminants through physical, chemical, and biological processes, providing wastewater purification (Prata et al., 2013).

In the literature, there are controversies on the real contributions of macrophytes in wastewater treatment in CWs, and some authors have reported that these systems do not contribute significantly to pollutant removals. However, most studies have demonstrated the relevant role of plants and that, according to Kadlec & Wallace (2009), their cultivation in CWs is essential for the good performance of these treatment systems since many studies have proven a higher efficiency in removing pollutants when they are present.

Differences in the behavior of evaluated systems may be associated with the hydraulic holding time (HHT), organic load or applied nutrient load (notably nitrogen and phosphorus), characteristics of system (flow direction), and the cutoff frequency of plant shoot (Wang et al, 2015; Zheng et al., 2015). As plants have limited nutrient absorption capacities, higher efficiencies can be obtained by applying lower loads than their removal capacity and thus a larger surface area should be available in HSSF–CWs.

Several plant species have been used in CWs, such as *Eichhornia crassipes* (Zacarkim et al., 2014), *Typha latifolia*, *Chrysopogon zizanioides* (Borges et al., 2015), *Canna flaccida*, *Zantedeschia aethiopica*, *Canna indica*, *Agapanthus africanus*, and *Watsonia borbonica* (Calheiros et al., 2015). However, there is still little scientific data on their behavior when grown intercropped in horizontal subsurface flow constructed wetlands (HSSF–CWs). Button et al. (2016) observed an influence of the cultivation of

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intercropping species (two by two) on the microbial community in CWs, and thus there could be an effect on the unit performance, which was demonstrated by Saraiva et al. (2018). Consequently, there may be a higher extraction capacity for species cultivated under this condition.

As plant species and cultivation form, the type of substrate used to fill CWs also decisively interferes with system performance and useful life. The most commonly used substrates are gravel, crushed stone, sand, and soil, but there has been an increased interest in alternative materials, such as solid waste discarded by other activities, with low acquisition costs and characteristics that minimize the speed of clogging of the porous medium of these systems (Yin et al., 2017; Matos et al., 2017). Alternative materials of an inert nature available are PET (polyethylene terephthalate) bottles, which is easily available because it is a solid waste produced in large quantities in urban areas whose final destination is increasingly becoming a problem of great magnitude.

Therefore, study aimed to evaluate the extraction capacity of Napier (*Pennisetum purpureum* Schum.) and Tifton 85 (*Cynodon* spp.) grasses when grown intercropped in sequence in HSSF–CWs filled with crushed PET bottles and gneiss gravel in the treatment of wastewater from a community bulk milk-cooling tank (MTWW).

#### **MATERIAL AND METHODS**

Raw wastewater generated in the cleaning process of a community bulk milk-cooling tank of Silveirânia, MG, located in Zona da Mata, where the experiment was implemented and conducted, was used in this study. PVC canvas. Regarding the type of substrate used as a support medium, two HSSF–CWs were filled with gneiss gravel #0 ( $D_{60} = 9.1$  mm, uniformity coefficient – UC  $D_{60}/D_{10} =$ 3.1, and initial void volume = 0.40 m<sup>3</sup> m<sup>-3</sup>) and other two with previously crushed 250 and 500 mL PET bottles. The choice for gneiss gravel #0 was because it is the type of substrate most commonly used in HSSF–CWs, while PET bottles were chosen as they are a low-cost substrate alternative for filling these systems.

and masonry sides waterproofed with a 0.5-mm thick

Affluent distribution was performed at the central point at the entrance of each HSSF–CW using a 0.5-inch plastic tap to control the applied wastewater flow. HSSF–CW effluent drainage system consisted of a 32-mm diameter PVC pipe, perforated and installed at the bottom of the HSSF–CW exit area. Wastewater level control in the system was performed by adjusting the height of the pipe connected externally to the HSSF–CW drainage system, maintaining a saturated height of 0.35 m in both types of support material (gravel #0 and crushed PET bottles).

Lids and labels were removed from PET bottles before being crushed to facilitate it and then capped again. Figure 1 shows the equipment used to crush PET bottles and the material ready to be used as substrate in HSSF–CWs.



FIGURE 1. Equipment used to crush PET bottles and detail of the final condition of the bottle used as support material.

The porosity of the medium composed of crushed PET bottles was quantified using a glass container (similar to that of an aquarium) of known volume (Vr), which was filled with the material and then water was added until filling all porous space. The water volume (Vw) used to fill the porous space in the container was used to determine the void index or substrate porosity (n) using [eq. (1)]. The value found was 0.64 m<sup>3</sup> m<sup>-3</sup>. The value of 0.40 m<sup>3</sup> m<sup>-3</sup> was used for porosity of the gravel #0, as in Ferres et al. (2017).

$$\mathbf{n} = \mathbf{V}\mathbf{w}/\mathbf{V}\mathbf{r} \tag{1}$$

In HSSF–CWs, where the substrate used was gravel #0, substrate depth was 0.45 m, whereas in those filled with crushed PET bottles, layer depth was 0.35 m. Another 0.10 m gravel #3 was placed above this layer to give weight and thus prevent this material from floating when MTWW was applied to HSSF–CWs. Thus, keeping MTWW level at 0.10 m below the surface of both HSSF–CWs (gravel #0 and crushed PET bottles + gravel #3 layer), the wet height in both systems was 0.35 m.

Plant species planted in HSSF–CWs were Napier (*Pennisetum purpureum* Schum.) and Tifton 85 (*Cynodon* spp.) grasses, whose seedlings were collected in a production area of the Department of Animal Science of the Federal University of Viçosa (UFV).

Regarding cultivation arrangement, two HSSF– CWs received Napier grass in the first half and Tifton 85 grass in the second half. In the other two HSSF–CWs, cultivation arrangement of these plant species was inverted, i.e., Tifton 85 grass was grown in the first half and Napier grass in the second half. Thus, considering different cultivation arrangement and substrates, operating conditions of HSSF–CWs were established as follows:

CW–GNT – substrate consisting of gravel #0, with Napier grass (*Pennisetum purpureum* Schum.) grown in the first half and Tifton 85 grass (*Cynodon* spp.) grown in the second half.

CW-GTN – substrate consisting of gravel #0, with Tifton 85 grass (*Cynodon* spp.) grown in the first half and Napier grass (*Pennisetum purpureum* Schum.) grown in the second half.

CW–PNT – substrate consisting of crushed PET bottles, with Napier grass (*Pennisetum purpureum* Schum.) grown in the first half and Tifton 85 grass (*Cynodon* spp.) grown in the second half.

CW–PTN – substrate consisting of crushed PET bottles, with Tifton 85 grass (*Cynodon* spp.) grown in the first half and Napier grass (*Pennisetum purpureum* Schum.) grown in the second half.

Tifton 85 grass was planted using 4- to 5-knot stem segments, while Napier grass was planted through 2- to 4knot vegetative propagules. Seedlings were inserted into the surface layer of the HSSF–CW bed by means of small pits of approximately 100 mm in diameter and 100 mm deep, which were subsequently covered with gravel or crushed PET bottles. Spacing between pits was triangular, totaling 48 pits per CW. Rooting and the fast seedling establishment were provided by a daily and briefly water level raising of the local supply network added to HSSF– CWs. After the seedling establishment, which occurred at 50 days after planting, MTWW was applied without dilution, and the experiment lasted 8.5 months.

The variables biochemical oxygen demand (BOD<sub>5.20</sub>), chemical oxygen demand (COD), total solids (TS), total suspended solids (TSS), total dissolved solids (TDS), total Kjeldahl nitrogen (TKN), total phosphorus (Ptotal), potassium (K), sodium (Na), turbidity, pH, and electrical conductivity (EC) were analyzed from MTWW samples collected fortnightly during the first three months and monthly during the remaining period. Laboratory analyses were performed at the Laboratory of Water Quality of the Department of Agricultural Engineering of UFV, in accordance with the Standard Methods recommendations (APHA, 2012). The evaluated variables and the respective methods are described as follows: BOD<sub>5,20</sub> - quantification of dissolved oxygen by the iodometric method (Winkler Process); COD – chemical oxidation by the open reflux method; TS and TSS - gravimetric method; TDS difference between TS and TSS; and TKN - Kjeldahl semimicro process. Total P concentrations (spectrophotometry) and Na and K (flame photometry) were quantified after nitric-perchloric digestion of the sample. The main physical, chemical, and biochemical characteristics of MTWW are shown in Table 1.

TABLE 1. Physical, chemical, and biochemical characteristics of wastewater from the cleaning process of a community bulk milk-cooling tank.

	Mean and standard deviation				
variable	Unit	Value			
pН	_	5.6 ± 0.1 (12)			
CE	$\mu S cm^{-1}$	$217 \pm 44.3(12)$			
BOD <sub>5,20</sub>	$mg L^{-1}$	$403 \pm 128$ (11)			
COD	$mg L^{-1}$	702±186 (12)			
Turbidity	UNT	125±39 (11)			
TSS	$mg L^{-1}$	202±46 (12)			
TDS	$mg L^{-1}$	366±292 (11)			
TS	mg L <sup>-1</sup>	570±125 (11)			
N-total	mg L <sup>-1</sup>	26±6 (12)			
P-total	$mg L^{-1}$	$10\pm 2(11)$			
Potassium	$mg L^{-1}$	$11\pm 2(6)$			
Sodium	$mg L^{-1}$	$12\pm 2$ (6)			
Oils and greases	$mg L^{-1}$	115±107 (11)			

\*In parentheses is the number of samples considered when calculating the means.

Table 2 shows the mean and standard deviation values of the theoretical HHT mean, and surface application rate of BOD<sub>5,20</sub>, nutrients and sodium applied during the monitoring period.

TABLE 2. Hydraulic holding time (HHT) and surface organic (OLRs), nutrient (N-total, P-total, and K) and sodium (Na) loading rates applied during HSSF–CW monitoring period.

Treatment	HHT	Flow	DOD <sub>5,20</sub>	N-total	P-total	Κ	Na	TS
	(d)	$(m^3 d^{-1})$	TAS (kg ha <sup>-1</sup> d <sup>-1</sup> )					
CW-GNT	1.85	0.187±0.004	318±104	20±5	7.7±1.2	7±2	8±2	421±94
CW–GTN	1.84	$0.187 {\pm} 0.006$	318±104	20±5	7.7±1.2	7±2	8±2	421±94
CW-PNT	2.97	$0.186 \pm 0.005$	318±104	20±5	7.7±1.2	7±2	8±2	421±94
CW–PTN	2.97	$0.187{\pm}0.005$	318±104	20±5	7.7±1.2	7±2	8±2	421±94

\*CW–GNT (gravel #0, with Napier grass grown in the first half and Tifton 85 grass grown in the second half); CW–GTN (gravel #0, with Tifton 85 grass grown in the first half and Napier grass grown in the second half); CW–PNT (PET, with Napier grass grown in the first half and Tifton 85 grass grown in the second half); CW–PTN (PET, with Tifton 85 grass grown in the first half and Napier grass grown in the second half).

Extraction capacity of cultivated plant species was determined from the quantification of dry matter yield and nutrient and sodium contents in the plant shoot. For this, four cuts were performed in the crops on August 1 (winter), September 17 (spring), November 3 (spring), and December 15 (summer), 2015, in periods from 40 to 60 days after the previous cut, depending on plant development stage.

The grass was cut at the height of 0.10 m from the HSSF–CW surface and plants positioned at the edges (0.5 m on the sides and ends) of each HSSF–CW were eliminated. The harvested material was placed in paper bags and weighed to determine the green mass yield (GMY), and then taken to an air circulation oven for drying at 65 °C until constant mass when the dry matter yield was quantified (Equation 2). Plant capacity to extract nutrients and sodium was obtained by [eq. (3)], according to Matos (2015).

$$PMS = \frac{(GMY \times DM)}{100}$$
(2)

$$NEC = \frac{(NC \times PMS)}{10}$$
(3)

Where,

GMY is the green mass yield (Mg ha<sup>-1</sup>);

DM is the dry matter content (dag  $kg^{-1}$ );

NEC is the nutrient extraction capacity (kg ha<sup>-1</sup>), and

NC is the nutrient content (dag  $kg^{-1}$ ).

Laboratory analyses of plant tissue were performed at the Laboratory of Soil and Solid Waste of the Department of Agricultural Engineering of UFV, in accordance with the recommendations of Kiehl (1985) and Matos (2015). Phosphorus, potassium, and sodium concentrations were quantified after nitric-perchloric digestion of the sample and measurement using

spectrophotometer and flame photometer, respectively. The statistical analysis of the data was arranged in a 2 x 2 x 2 factorial scheme, totaling eight treatments. Factors, with two levels each, consisted of plant species (Napier and Tifton 85 grass), support material (gravel #0 and crushed PET bottles), and cultivation arrangement (first and second half). The experimental design used was a randomized block design, in which cuts were considered as blocks.

The assumptions of normality and homogeneity of variance were verified by the Lilliefors and Cochran Bartlett tests, respectively. In case of the normal distribution of the data, the means of variables obtained in each treatment were submitted to analysis of variance (ANOVA, p = 0.05) and, when significant, to the Tukey test (p = 0.05). A slicing regarding arrangement, cultivation species and type of support material was carried out when a significant interaction was observed between factors. The software Assistat v. 7.7 Beta was used for data processing and statistical analysis.

## **RESULTS AND DISCUSSION**

The values of sodium and nutrient extraction capacity (NEC) and dry matter yield (DM) of the shoot of Napier and Tifton 85 grasses are shown in Table 3.

HSSF-CW	Plant species	$\frac{\text{NEC}}{(\text{kg ha}^{-1} \text{ d}^{-1})}$				$\frac{\rm DM}{\rm (Mg\ ha^{-1})}$
		Ν	Р	K	Na	
	Т	2.9±1.10	0.2±0.10	1.0±0.70	0.02±0.01	2.2±1.0
CW–GNT	Е	$5.6 \pm 1.80$	$0.5 \pm 0.30$	$3.3 \pm 2.50$	$0.01 \pm 0.01$	$4.0{\pm}1.8$
	Т	2.1±1.00	0.2±0.10	$0.9{\pm}0.60$	$0.03{\pm}0.02$	2.0±1.2
CW-GTN	E	$6.8 \pm 4.70$	$0.4{\pm}0.30$	$2.5 \pm 1.90$	$0.01 {\pm} 0.01$	3.1±2.3
CW-PNT	Т	$1.6{\pm}0.80$	0.2±0.10	$0.4{\pm}0.20$	$0.03{\pm}0.02$	$1.4{\pm}0.6$
	Е	4.5±2.50	$0.5 \pm 0.10$	$1.2{\pm}0.70$	$0.01 {\pm} 0.01$	3.1±1.2
CW-PTN	Т	2.7±1.00	0.3±0.10	0.7±0.30	0.06±0.03	1.9±0.4
	Е	3.0±1.60	0.3±0.20	$1.1 \pm 0.80$	$0.01 \pm 0.01$	$1.7{\pm}0.7$

TABLE 3. Mean and standard deviation values of sodium (Na) and nutrient extraction capacity (NEC) (N, P, and K) and dry matter yield (DM) through the shoot of Napier (E) and Tifton 85 (T) grown in different HSSF–CWs.

\*CW–GNT (gravel #0, with Napier grass grown in the first half and Tifton 85 grass grown in the second half); CW–GTN (gravel #0, with Tifton 85 grass grown in the first half and Napier grass grown in the second half); CW–PNT (PET, with Napier grass grown in the first half and Tifton 85 grass grown in the second half); CW–PTN (PET, with Tifton 85 grass grown in the first half and Napier grass grown in the second half).

The first shoot cut of grasses was carried out at 56 days after planting, the second cut at 46 days after the first cut, the third cut at 47 days after the second cut, and the fourth cut at 42 days after the third cut.

The mean dry matter yield of the shoot of grasses ranged from 1.4 to 4.0 Mg ha<sup>-1</sup>, which are close to those

found by Andrade et al. (2000), who observed values from 2.6 to 4.7 Mg  $ha^{-1}$  when evaluating different nitrogen doses on Napier grass yields.

Matos et al. (2009), on the other hand, obtained from 20 to 34 Mg ha<sup>-1</sup> of dry matter of Tifton 85 in HSSF– CWs used in the treatment of swine wastewater (SWW) in three different cuts at an interval from 100 to 120 days between cuts, with a mean application of 93 and 22 kg ha<sup>-1</sup> d<sup>-1</sup> of nitrogen and phosphorus, respectively. The values of dry matter yield obtained by these authors were higher when compared to those found in this study, which can be attributed to the shorter period between cuts of the grass shoot (40 to 60 days) and, mainly, the lower nitrogen

(20.0 kg  $ha^{-1}\ d^{-1})$  and phosphorus (7.7 kg  $ha^{-1}\ d^{-1})$  application rates.

The mean values of the extraction capacity of N, P, K, and Na through cuttings of Napier and Tifton 85 shoots when cultivated in different substrates and arrangements in HSSF–CWs, as well as the interactions between factors, are shown in Tables 4 and 5, respectively.

TABLE 4. Mean values of sodium and nutrient extraction capacity (NEC) through the shoot of grasses grown under the different substrate, plant species, and cultivation positions in HSSF–CWs.

Factor	Factor level	NEC (dag $kg^{-1}$ )					
		Ν	Р	К	Na		
Plant species	E	228.58a	19.75a	Ι	Ι		
	Т	107.57b	11.09b	Ι	Ι		
Substrate	В	Ι	15.63a	Ι	Ι		
	Р	Ι	15.21a	Ι	Ι		
Cultivation position	First half	Ι	17.54a	70.56a	1.00a		
	Second half	Ι	13.30a	56.96a	0.84a		

E – Napier grass; T – Tifton 85 grass; B – gneiss gravel #0; P – crushed PET bottles; I – positive interaction between factors. \*Means followed by the same letter do not differ statistically from each other by the Tukey test at 5% significance level.

TABLE 5. Mean extraction capacity (NEC) of nutrients (N and K) and sodium (Na) regarding the interaction between plant species, cultivation position, and filling substrate of HSSF–CWs.

	NEC (kg ha <sup>-1</sup> )						
Source of variation	K		Na				
Plant species	Substrate						
	В	Р	В	Р			
E	132.71aA	53.50aB	0.668bA	0.527bA			
Т	44.20bA	24.61aA	1.046aB	1.454aA			
	N-total						
Cultivation position	Substrate						
	В		Ι	Р			
First half	176.3	168.2	168.29aA				
Second half	220.36aA 107.29bB						

E - Napier grass; T - Tifton 85 grass; B - gneiss gravel #0; P - crushed PET bottles.

\*Means followed by the same letter lowercase letter in the column and uppercase letter in the row do not differ statistically from each other by the Tukey test at 5% significance.

The average N extraction capacities obtained by Napier grass shoots were higher than those obtained by Tifton 85 grass, which justifies the higher mean yields achieved by this plant species.

Hunt et al. (2003) verified, under load of 3.0 kg ha<sup>-1</sup> d<sup>-1</sup> of N provided by SWW application, extractions of 1.17 kg ha<sup>-1</sup> d<sup>-1</sup> in a HSSF–CW under mixed cultivation of *Sparganium americanum* and *Typha*, and 0.97 kg ha<sup>-1</sup> d<sup>-1</sup> in a mixed cultivation of *Juncus effusus* and *Scipus*. Matos et al. (2009) found that Tifton 85 grass was able to extract, in different sections of its shoot, between 5.00 and 6.00 kg ha<sup>-1</sup> d<sup>-1</sup> of N when cultivated in an HSSF–CW used in SWW treatment, which is higher when compared to the values obtained in this study (Table 2). In this case, a higher N application rate was used by Matos et al. (2009),

which justifies the differences found in this study. Costa et al. (2015) obtained removals of 1.99 kg ha<sup>-1</sup> d<sup>-1</sup> of N via absorption (present in biomass) in HSSF–CWs grown with *Typha latifolia*, which are lower than the values found in this study and the literature. These low values can be explained by the lower HHT (1.2 d) of units monitored by the authors, indicating it is another factor influencing the performance of species grown in HSSF–CWs.

Grasses grown in the second half of CWs filled with gneiss gravel extracted higher amounts of N when compared to those grown in CWs filled with PET bottles at the same cultivation position. The explanation probably lies in the dynamics of nitrogen in these reactors. In HSSF–CWs, organic material mineralization includes the conversion of organic nitrogen into ammonia nitrogen, which is made available and can be absorbed by plants or even oxidized due to an expected increase in the redox potential of the medium. Thus, nitrate is formed and assimilated by plants and/or incorporated into microbial cellular material. Because there is a higher O<sub>2</sub> availability in the medium in the second half of CWs (Dušek et al., 2008), higher availability of nitrate is expected in the medium. However, because it is a highly soluble ion, it may leave the system more easily in support media with higher porosity. On the one hand, the highest N extractions were obtained in the first half of CWs filled with PET bottles, on the other hand, SACs filled with gravel showed no difference for cultivation positions in relation to the extraction of this nutrient. The highest drainable or effective porosity, consisting of macropores, which is where the wastewater drains most easily into the substrate of crushed PET bottles, may have allowed for faster degradation of organic matter in the medium and higher N availability to grasses in the initial part of the system. It occurs because larger pores present a higher possibility of renewal and gas exchange, providing a more aerobic condition, thus accelerating the degradation of organic matter and availability of nutrients in the medium.

Thus, higher extractions would have happened in the first half of CWs filled with PET bottles when compared to those obtained in areas close to the exit of the system.

According to the mean values of daily contribution (Table 2) and extraction of N (Table 3), Tifton 85 and Napier grasses were able to extract from the system via shoot cuttings 14.5 and 28.0% (CW–GNT), 10.5 and 34.0% (CW–GTN), 8.0 and 22.5% (CW–PNT), and 13.5 and 15.0% (CW–PTN), respectively, applied to them. These values can be considered of high relevance in terms of wastewater treatment.

Napier grass showed higher mean P extraction capacity through the shoot when compared to that obtained by Tifton 85 grass. This result is mainly due to the higher mean yield obtained by Napier grass since grasses evaluated in this experiment had mean P contents equal. Regarding the different substrates and plant cultivation positions, mean extraction capacities were statistically equal.

Garcia et al. (2015) obtained P extraction capacity ranging from 0.31 to 0.82 kg ha<sup>-1</sup> d<sup>-1</sup> in Tifton 85 grass fertigated with treated domestic sewage for 30 days under greenhouse conditions. In this case, P extraction capacity increased as wastewater dose increased.

The mean P extraction capacity of plant shoot obtained in this study, taking into account only the mean daily value of contribution to the system (Table 1), was 3.0 and 6.6% in CW-GNT, 2.5 to 5.8% in CW-GTN, 2.7 to 6.2% in CW-PNT, and 4.3 and 3.9% in CW-PTN, respectively, by the Tifton 85 and Napier. Matos et al. (2010) evaluated the extraction capacity of different plant species and observed that Tifton 85 grass removed, on average, 3.2% of the total P made available to the system, which is close to the value obtained in this study. Also, according to these authors, P is an element difficult to remove with conventional wastewater treatment systems and this value can be considered significant, mainly considering that this removal is only due to plant absorption. Nutrient release, especially N and P, is the main factor responsible for the eutrophication of water bodies and can result in the process of algal proliferation in the aquatic environment, thus harming the beneficial uses

of this water. Therefore, any removal of P provided to the effluent to be discharged into water bodies should be considered of high environmental value.

The different cultivation positions of plants provided no significant effect on K extraction capacity from MTWW, which is associated with the high solubility and therefore high mobility of this cation in the medium (Matos et al., 2010), which provide more homogeneous distribution in the porous medium of HSSF–CWs.

Napier grass grown in CWs filled with gravel was able to extract higher amounts of K through its shoot cutting when compared to those filled with PET bottles. However, for Tifton 85 grass, the different substrates did not have a significant effect on the extraction of this nutrient by plant shoot. Napier grass showed higher K extraction capacity when compared to Tifton 85 grass when cultivated in CWs filled with gravel, while CWs filled with PET bottles had K extraction capacity statistically equal between grasses.

Considering only the value of K supplied to the system (Table 2), Tifton 85 and Napier grasses were able to extract from the system 1.0 and 3.3 kg ha<sup>-1</sup> d<sup>-1</sup> (14 and 46% of the supplied value) in CW–GNT, 0.9 and 2.5 kg ha<sup>-1</sup> d<sup>-1</sup> (13 and 35% of the supplied value) in CW–GTN, 0.4 and 1.2 kg ha<sup>-1</sup> d<sup>-1</sup> (6 and 17% of the supplied value) in CW–PNT, and 0.7 and 1.1 kg ha<sup>-1</sup> d<sup>-1</sup> (10 and 15% of the supplied value) in CW–PTN. As discussed in relation to N extraction, the values obtained are relevant, considering the recognized difficulty in extracting soluble chemical elements in biological treatment systems.

Matos et al. (2009) evaluated K extraction capacity via plant shoot of *Typha latifolia* L., *Alternanthera philoxeroides*, and Tifton 85 grass (*Cynodon dactylon* Pers.) grown in HSSF–CWs with SWW and obtained removals of 12.7, 23.0, and 11.7%, respectively, in relation to the mass supplied to the system.

Regarding Na, Tifton 85 grass grown in HSSF–CWs filled with PET bottles provided higher mean extraction values when compared to those filled with gravel. For Napier grass, the different substrates did not provide any difference in Na extraction capacity from MTWW. The highest extraction capacities of this chemical element were obtained by shoot extraction of Tifton 85 grass, considering the same filling substrate of HSSF–CWs.

Matos et al. (2010) evaluated Na extraction capacity by Napier and Tifton 85 in HSSF–CWs used in the treatment of ARL and verified the better performance of Tifton 85 grass, corroborating the result obtained in this research.

Sodium, like potassium, is a chemical element of difficult removal in conventional wastewater treatments (Lo Monaco et al., 2009) and, therefore, plant species that can absorb significant quantities of this chemical element from the environment should be chosen when it is one of the targets of wastewater treatment. Applying a mean load of 8.2 kg ha<sup>-1</sup> d<sup>-1</sup> of Na (Table 2), Tifton 85 and Napier were able to extract from the system through shoot cuttings 0.02 and 0.01 kg ha<sup>-1</sup> d<sup>-1</sup> (0.24 and 0.12% of the supplied value) in CW–GNT, 0.03 and 0.01 kg ha<sup>-1</sup> d<sup>-1</sup> (0.37 and 0.12% of the supplied value) in CW–GTN, 0.03 and 0.01 kg ha<sup>-1</sup> d<sup>-1</sup> (0.73 and 0.01 kg ha<sup>-1</sup> d<sup>-1</sup> (0.73 and 0.12% of the supplied value) in CW–PNT, and 0.06 and 0.01 kg ha<sup>-1</sup> d<sup>-1</sup> (0.73 and 0.12% of the supplied value) in CW–PTN, respectively, of the MTWW applied to HSSF–CWs.

Queiroz et al. (2004), despite obtaining higher NEC for Na, found similar percentages of extractions through shoot cutting of Tifton 85 (0.3%) when compared to that supplied in this study.

#### CONCLUSIONS

Cultivation position of both grasses in HSSF–CWs did not influence their capacity to extract nitrogen, phosphorus, potassium, and sodium from MTWW. However, under the conditions the study was conducted, if the main purpose of wastewater treatment is sodium removal, the monoculture with Tifton 85 grass in HSSF–CWs filled with crushed PET bottles is recommended. On the other hand, if the major interest is the removal of N, P, or K, the recommendation is for Napier cultivation in HSSF–CWs filled with gneiss gravel.

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