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Scientific Paper

Partially saturated vertical flow constructed wetland for urban wastewater treatment

Wetland construído com fundo parcialmente saturado para o tratamento de esgoto sanitário

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ABSTRACT

Partially saturated vertical flow constructed wetland (SVF) emerge as an alternative to classical vertical flow due to improved carbonaceous organic matter removal and nitrogen transformations without of the need for external energy source. In this context, the main objective of this study was to show the long-term evaluation of a SVF wetland with 7.5 m^2 of surface area (filter media depth of 0.75 m where 53% of total depth was saturated) and planted with *Typha domingensis* macrophyte, operated as secondary/advanced treatment unit of urban wastewater under subtropical climate conditions. Sampling and analysis of conventional wastewater quality parameters, oxygen consumption rate estimation, and assessment of bacterial dynamics were conducted over 6 years, which allowed inferring that operating the SVF wetland with a specific hydraulic load around 4 $L{\cdot}m^{-2}{\cdot}min^{-1}$ and hydraulic regime with feeding and resting cycles of 3.5 d, all wastewater quality parameters met local legislation release standards in river water bodies. Saturated zone of the wetland favors the presence of denitrifying bacteria representing a potential of 44% of TN removal due to simultaneous nitrification-denitrification.

Keywords: constructed wetlands; saturated bottom layer; treatment performance; bacterial dynamics.

RESUMO

Wetlands construídos verticais de fundo saturado (SVF) despontam como uma alternativa aos clássicos wetlands verticais devido ao aumento da remoção de matéria orgânica carbonácea e as transformações de nitrogênio sem a necessidade de fonte de energia externa. Neste contexto o objetivo principal deste estudo foi mostrar a avaliação a longo prazo de um SVF com 7,5 m^2 de área superficial (meio filtrante profundidade de 0,75 m onde 53% da profundidade total estava saturada) e plantado com macrófita *Typha domingensis*, sendo operado como unidade de tratamento secundário/avançado esgoto sanitário em condições de clima subtropical. Amostragens e análises de parâmetros de qualidade de tratamento de efluentes, estimativa da taxa de consumo de oxigênio e avaliação da dinâmica bacteriana foram realizadas ao longo de 6 anos, o que permitiu inferir que operando o SVF *wetland* com uma carga hidráulica específica em torno de 4 $L{\cdot}m^{-2}{\cdot}min^{-1}$ e regime hidráulico com ciclos de alimentação e descanso de 3,5 d, todos os parâmetros de qualidade no efluente tratado atenderam aos padrões de lançamento da legislação local em corpos d'água superficiais. A presença de bactérias desnitrificantes na zona saturada do SVF *wetland* infere a um potencial de 44% de remoção de NT devido à ocorrência da nitrificação-desnitrificação simultâneas.

Palavras-chave: wetlands construídos; saturação de fundo; desempenho de tratamento; dinâmica bacteriana.

INTRODUCTION

Constructed wetlands (CW) are an ecotechnology widely used worldwide. Nowadays, an operational strategy has been employed in the classical vertical flow constructed wetland to maximize treatment performance, especially for nitrogen transformations. This strategy is known as partially saturated vertical flow constructed wetland (SVF), which is provided by a partial saturation bottom

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layer through the outlet level controller elevation, providing aerobic, anoxic/anaerobic environments in the same unit. Such an approach has shown higher total nitrogen removal efficiency due to nitrification and denitrification processes (PELISSARI *et al.*, 2017), likely for providing oxidative and reducing environments inside the same unit, greater adaptation of the macrophytes, and lower final effluent volume due to high evapotranspiration capacity.

The feeding mode usually employed in the SVF wetland is composed of different variables linked to the hydraulic regime, such as number of pulses, specific pulse volume (SPV), time interval between pulses, specific hydraulic loading rate applied (SHLR), and hydraulic loading rate (HLR).

The optimization of the feeding mode is related to the amount of oxygen supplied and the minimum contact time required for oxidation reactions (Torrens *et al.*, 2009). The interdependence between contact time and oxygen supply can be assessed by the oxygen consumption rate (OCR) that occurs inside the filter medium (NIVALA *et al.*, 2013). If the contact time and the oxygen supply are adequate, there will be a higher yield in the oxidation reactions. This implies a high OCR, resulting in higher carbon and nitrogen removal performance (Torrens *et al.*, 2009). Thence, it is clear that the relationship between oxygen supply, OCR, hydraulic retention time (HRT), and the feeding mode directly influences the treatment performance of the SVF wetland (BOLLER *et al.*, 1993; FORQUET *et al.*, 2009).

The main objective of this study was to show the long-term evaluation in SVF wetland applied as secondary/advanced treatment unit of urban wastewater under subtropical climate conditions.

METODOLOGY

The wastewater treatment system was implemented in southern Brazil (27°35'48" latitude 48°32'57" longitude), under subtropical climate. The treatment plant was comprised of a septic tank (3 m^3 of volume) as primary treatment, followed by SVF wetland as a secondary and advanced treatment. The wetland module was commissioned in June 2015 and was designed to treat urban wastewater equivalent to 5 populations (Figure 1).

The operational module of SVF wetland had a 7.5 m^2 of surface area and was planted with *Typha domingensis* macrophyte (4 plants per m^2). The module had a filter media depth of 0.75 m, with an upper layer of 0.05 m composed by gravel, followed by 0.60 m of sand (effective diameter d_{10} of 0.29 mm, d_{60} of 1.16, and uniformity of 4.05), and 0.10 m of drainage layer composed by gravel at the bottom. The last 0.40 m (53% of total depth of the treatment unit) were kept saturated with the effluent (Figure 2).

Operational characteristics

Flow rate was continuously measured throughout the monitoring period. Inflow rate measurement was made during each sampling for water quality monitoring. The inflow was measured by using a container of known volume (approximately 40 L). The time needed to fill it was measured and then the flow rate was calculated (Figure 3). The outflow from the SVF wetland was measured continuously. A tilting device with 3 L volume was set up at the outlet of the SVF wetland. Each time the device was filled up, it tilted and emptied itself, returning immediately to its primary position. At this moment, the time and classification of the tilting was recorded in a data logger, allowing the measurement of total treated volume and the discharge rate for the wetland module (Figure 4).

Tracer tests with saline tracers were conducted to determine the HRT of the SVF wetland, as described by Fechine *et al.* (2020). Two tracer tests were performed as follows: the first one after 38 operational months and the second one after 42 operational months.

Pruning of the plants was carried out every 3 months, as shown in Figure 5.

Sampling and analysis of conventional wastewater quality parameters

Influent and effluent wastewater samples were collected twice a month throughout the study period, just after the 8 a.m. feeding pulse in the superficial area of the wetland module. Samples were taken to the adjacent laboratory for analysis of the following water quality parameters: pH, total suspended solids (TSS), chemical oxygen demand (COD), biochemical oxygen demand (BOD_5), total nitrogen (TN), ammonium nitrogen (NH_4^+-N), and orthophosphate phosphorus (PO_4^{3-}-P), according to standard methods (APHA *et al.*, 2005) and Vogel (1981) for NH_4^+-N analysis.

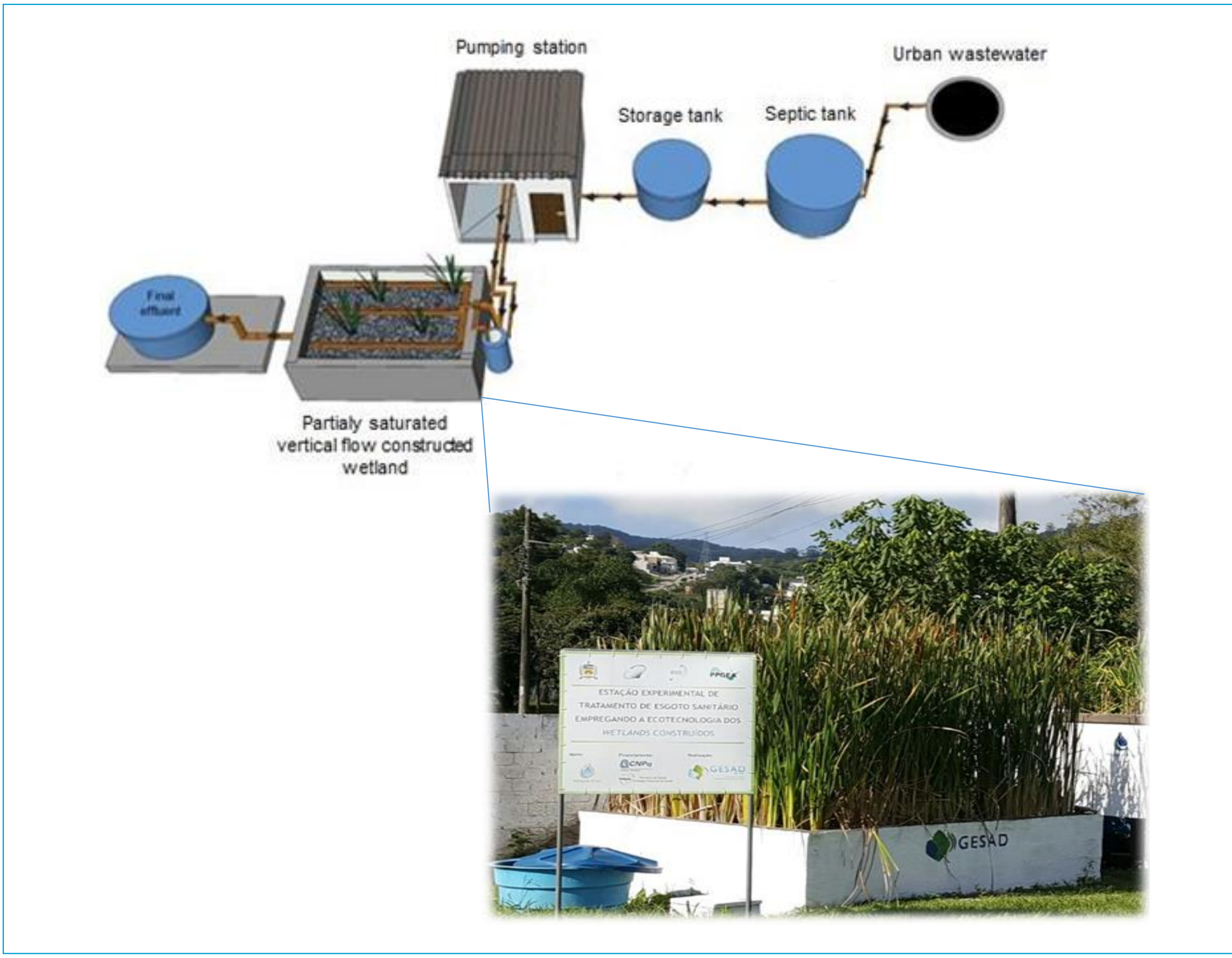


Figure 1 – Diagram and image of a partially saturated vertical flow constructed wetland (SVF wetland) of a wastewater treatment pilot plant.

Oxygen consumption rate estimation

OCR was estimated based on the oxygen consumption utilized for organic matter and NH_4^+-N oxidations that occurred in SVF wetland during the operational period, considering 4 pulses per day of feeding. The OCR was calculated by means of the model proposed by Platzer (1999), as described in Equation 1.

$$OCR = 0.7\ (\Delta\ COD) + 4.86\ (\Delta\ TKN) - 2.89\ (\Delta\ TN) \quad (1)$$

Where:

OCR: oxygen consumption rate ($g \cdot m^{-2} \cdot d^{-1}$);

Δ COD: COD load removal efficiency ($g \cdot m^{-2} \cdot d^{-1}$);

Δ TKN: TKN load removal efficiency ($g \cdot m^{-2} \cdot d^{-1}$);

Δ TN: TN load removal efficiency ($g \cdot m^{-2} \cdot d^{-1}$).

Sampling and assessment of bacterial dynamics

Quantitative essay of functional genes from total eubacteria (16S rRNA), nitrifying (*amoA*), and denitrifying bacteria (*nosZ*) were determined through quantitative polymerase chain reaction (qPCR) analyses. During the first year of SVF wetland operation (from July 2015 to July 2016), filter media samples from top (0 – 15 cm depth) and bottom (60 – 70 cm depth) layers were collected at two points in each layer. Samples were collected every two months totalizing seven sampling campaigns throughout the study (first, third, fifth, seventh, ninth, eleventh, and twelfth month of operation). All samples were collected with the aid of a properly sterilized soil sampler. Moreover, filter media collection and storage of the samples followed the recommendations of Pelissari *et al.* (2017, 2018). qPCR analysis was performed at all sampling campaigns.

DNA extraction of approximately 0.25 g of filter media was carried out in triplicate at each sampling campaign using the Power Microbiome™ DNA Isolation kit protocol (MOBIO Laboratories, Inc.; Carls- bad, CA). DNA extracts were kept frozen at -80°C until further analysis. qPCR analysis of eubacteria was conducted in the V3 hypervariable region of 16S rRNA following Prenafeta-Boldú *et al.* (2012). Nitrifying population was quantified by ammonia monooxygenase α-subunit encoding gene (*amoA*), as recommended by Rotthauwe et al. (1997), by ammonia oxidizing bacteria (AOB). The denitrifying population were quantified by noz Z (Clade I) gene that accesses the enzyme nitric oxide reductase (Braker and Tiedje, 2003; CALDERER *et al.*, 2014). All samples were analyzed in triplicate by means of three independent samples. The standard curve of each target gene was designed by using the FunGene database (http://fungene.cme.msu.edu/) five gBlocks® Gene Fragments (IDT, Integrated DNA Technologies). Serial dilutions (10^{10} to 10^{2} gene copies μL^{-1}) from synthetic genes were subjected to qPCR assays in duplicate. qPCR reactions fitted quality standards efficiency between 95 and 110% and R^2 above 0.995. The qPCR reaction was analyzed using the 7500 real-time PCR system (Applied Biosystems, The Netherlands).

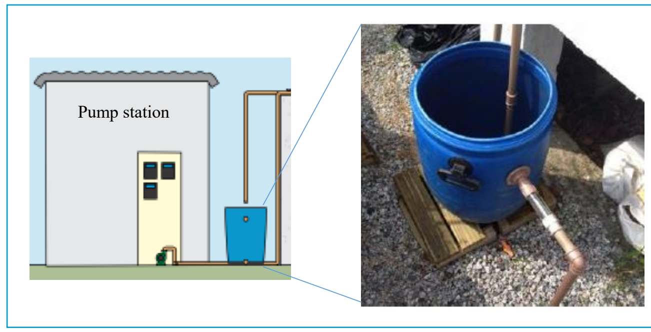


Figure 3 – Illustrative pump station and image of the inflow meter device.

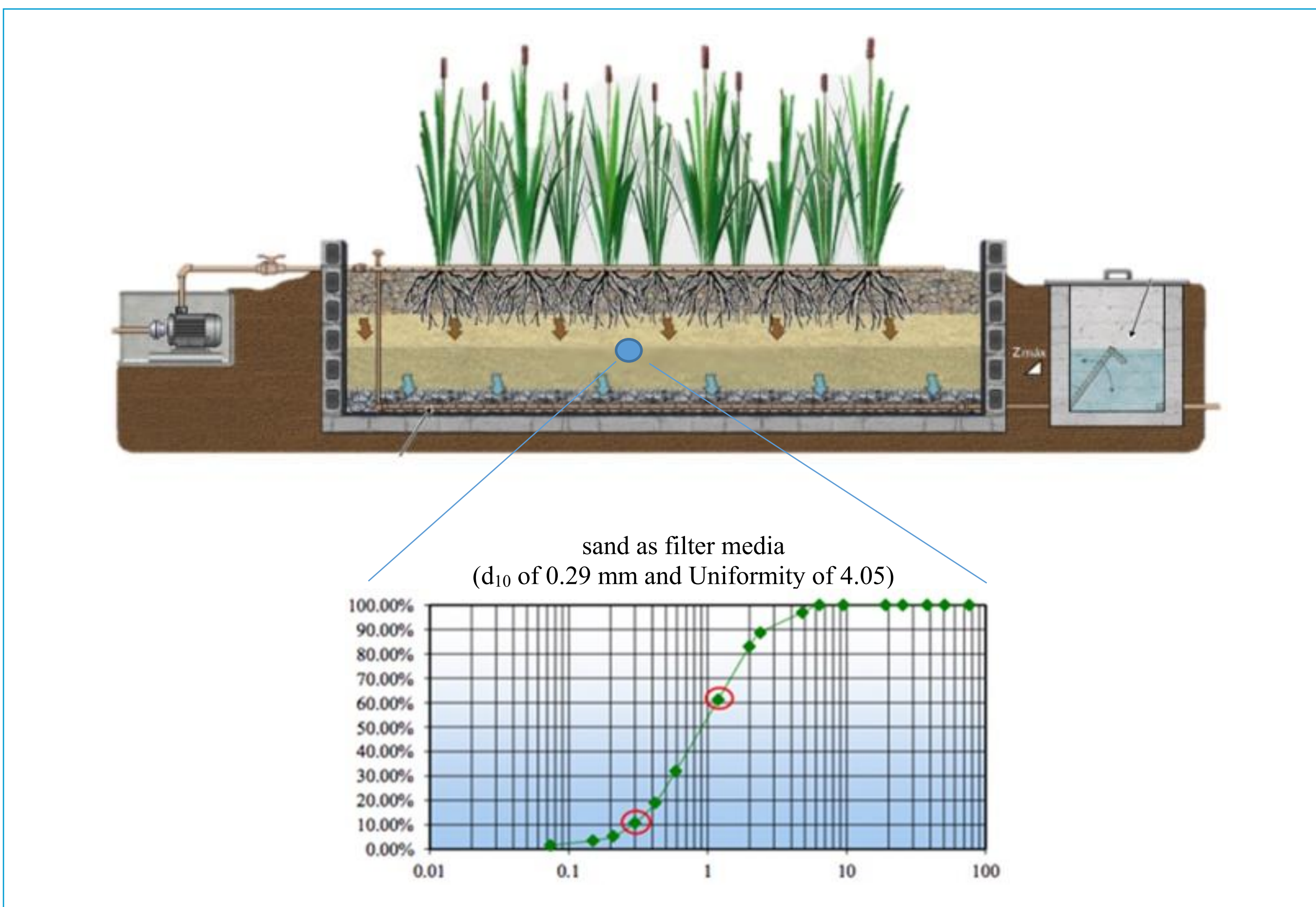


Figure 2 – Cross-section profile of partially saturated vertical flow constructed wetland (SVF wetland) and sand characterization.

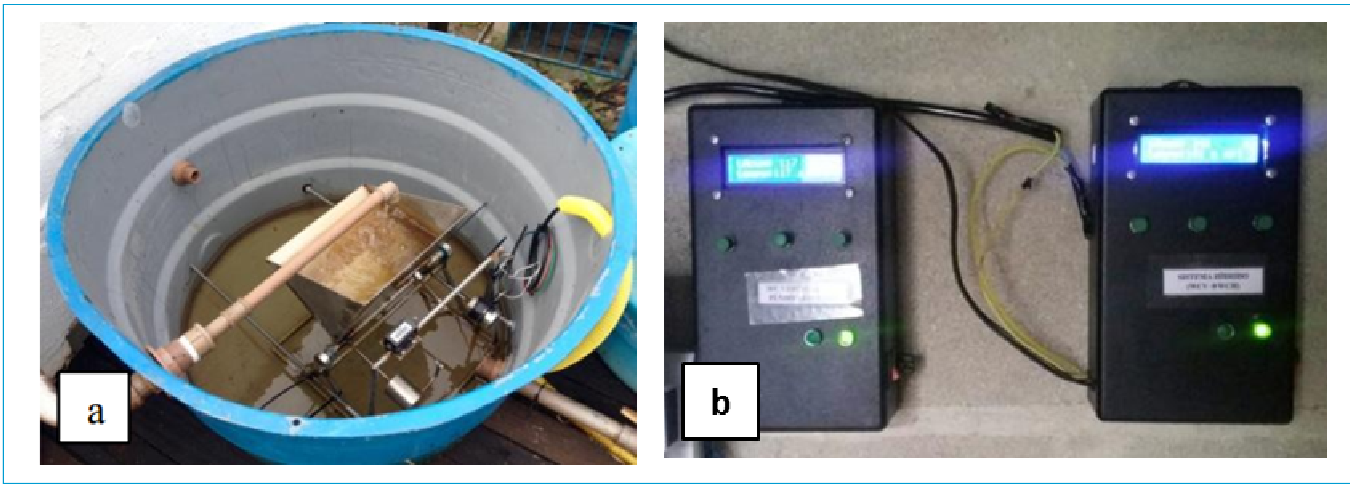


Figure 4 – Flow meter device installed at the outlet of wetland module. (a) Image of the tilting device. (b) Image of data logger.

Figure 5 – Images of *Typha domingensis* macrophytes pruning.

RESULTS AND DISCUSSION

The SVF wetland was operated with hydraulic regime with feeding and resting cycles of 3.5 days. For each feeding day, the average flow applied was 560 L·d^{-1} by means of 3 and 4 pulses throughout the day. The average HLR applied was 85 mm d^{-1} and the average organic load rate (OLR) was 40 g COD m^{-2}·d^{-1}.

Regarding the tracer tests, a tracer recovery of 74% was observed for test 1 and 86% for test 2 (Table 1), which is considered a good recovery in the literature, where values around 80% are accepted (HEADLEY and KADLEC, 2007).

Based on the treatment performance identified over three years of operation (Table 2), the SVF wetland performed better than conventional vertical flow constructed wetlands (see BASSANI *et al.*, 2021) and is configured as a technological alternative with great potential for promotion of urban wastewater treatment in the context of decentralization.

Table 1 – Values obtained by tracer tests applied in the saturated vertical flow constructed wetland module.

Parameter	Test 1	Test 2
Average air temperature (°C)	22	25
Maximum air temperature (°C)	29	30
Minimum air temperature (°C)	16	20
NaCL$^-$ loading applied (g/L)	10	10
Inflow volume (L)	186	179
NaCL$^-$ mass applied (g)	1821	1780
HRT theoretical (d)	1.6	1.6
HRT real (d)	1.4	1.6
Average TSS loading rate (g·m^{-2}·d^{-1})	5	6
Tracer recovery (%)	74	86
ITMD = HRT real / HRT theoretical	0.9	1.0

HRT: hydraulic retention time; TSS: total suspended solids; ITMD: average detention time index.

MeanThe range of values for the OCR identified in the SVF wetland ranged from 37 to 42 g O_2 m^{-2}·d^{-1} during the operational period. Based on the OCR values obtained and considering an average 4 $L \cdot m^{-2} \cdot min^{-1}$ of SHLR applied, it results in a reduction of approximately 33% of the SHLR value recommended by the German standard (6 $L \cdot m^{-2} \cdot min^{-1}$) (NIVALA *et al.*, 2018), without negatively affecting the oxygen supply.

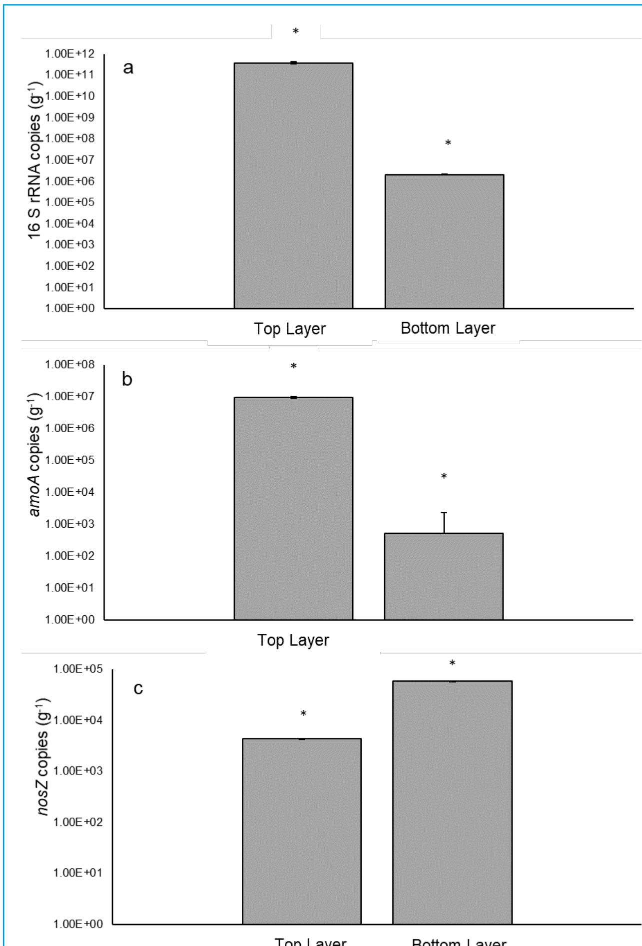
The eubacterial population showed a different behavior between the layers of SVF wetland (Figure 6a). In the upper layer (10^{11} 16S rRNA copies gene g^{-1}), bacterial abundance was five orders of magnitude compared to the bottom layer (10^{6} 16S rRNA copies gene g^{-1}). Greater bacterial abundance at the top may be associated with higher oxygen and carbon availability in this layer.

Regarding AOB, the *amoA* gene showed the same behavior as eubacteria populations (Figure 6b). Greater abundance was identified in the upper layer (10^{6} *amoA* copies gene g^{-1}) rather than in the bottom layer (10^{2} *amoA* copies gene g^{-1}). These results suggest that nitrification occurred in a larger magnitude in the first 15 cm of this wetland. On the other hand, it is important to note that even in a region with lower oxygen concentration, such

Table 2 – Average values of concentrations and loads (standard deviation) of wastewater quality parameters in the influent (after septic tank) and effluent of the partially saturated vertical flow constructed wetland for the 6-year evaluation period (valid samples were 80 for all parameters, except to total nitrogen, which were 50 samples).

Parameter	Influent	SVF effluent
pH	7.2 (0.4)	6.1 (0.4)
TSS ($mg \cdot L^{-1}$)	**82 (52)**	**1 (2)**
ALR ($g \cdot m^{-2} \cdot d^{-1}$)	8 (5)	
LRR ($g \cdot m^{-2} \cdot d^{-1}$)		7 (4)
RE (%)		99
COD ($mg \cdot L^{-1}$)	**440 (170)**	**34 (33)**
ALR ($g \cdot m^{-2} \cdot d^{-1}$)	40 (10)	
LRR ($g \cdot m^{-2} \cdot d^{-1}$)		38 (9)
RE (%)		92
BOD_5 ($mg \cdot L^{-1}$)	**270 (98)**	**25 (7)**
ALR ($g \cdot m^{-2} \cdot d^{-1}$)	30 (5)	
LRR ($g \cdot m^{-2} \cdot d^{-1}$)		27 (5)
RE (%)		91
TN ($mg \cdot L^{-1}$)	**78 (24)**	**47 (20)**
ALR ($g \cdot m^{-2} \cdot d^{-1}$)	8 (2)	
LRR ($g \cdot m^{-2} \cdot d^{-1}$)		3 (1)
RE (%)		44
NH_4^+-N ($mg \cdot L^{-1}$)	**66 (15)**	**19 (10)**
ALR ($g \cdot m^{-2} \cdot d^{-1}$)	6 (2)	
LRR ($g \cdot m^{-2} \cdot d^{-1}$)		5 (1)
RE (%)		71
PO_4^{3-}-P ($mg \cdot L^{-1}$)	**37 (8)**	**7 (2)**
ALR ($g \cdot m^{-2} \cdot d^{-1}$)	4 (1)	
LRR ($g \cdot m^{-2} \cdot d^{-1}$)		3 (1)
RE (%)		81

TSS: total suspended solids; ALR: Applied Load Rate; LLR: Load Rate Removal; RE: Removal Efficiency; COD: chemical oxygen demand; BOD_5: biochemical oxygen demand; TN: total nitrogen.



Presented values are the mean and standard deviation of independent triplicates from seven sampling campaigns. * Differences statistically between the top and bottom layers of the wetland ($p < 0.05$).

Figure 6 – Nitrogen functional genes average from seven sampling campaigns, identified in the biofilm from top (0 – 15 cm) and bottom (60 – 70 cm) layers of the partially saturated vertical flow constructed wetland, during the first year of operation. (a) Average of 16S rRNA copies abundance; (b) Average of *amoA* copies abundance; (c) Average of *nosZ* (clade I) copies abundance.

as the saturated layer of the wetland, the presence of AOB was identified.

Furthermore, the denitrifying population was identified throughout the vertical profile of SVF wetlands (Figure 6c). The *norZ* gene (10^3 and 10^4 g^{-1} copies in top and bottom layers, respectively) was higher in the bottom layer. This behavior suggests that the saturated zone of the wetland favors denitrification.meanMeanMeanMeanstandard deviation Statistical dif

CONCLUSION

Based on 6 years of operation in a partially SVF wetland applied as secondary/advanced treatment unit of urban wastewater under subtropical climate conditions, the main conclusions were as follows:

The SVF wetland presented itself as a potential intensification of the classic modality of vertical flow constructed wetland, highlighting the efficiency in carbonaceous organic matter (mean of 92% for COD and 88% for BOD) and 71% NH_4+-N.

The saturated conditions of the SVF wetland influenced the AOB populations, suggesting that nitrification occurred in a larger magnitude in the first 15 cm of this wetland.

The saturated zone of the wetland favors the presence of denitrifying bacteria representing a potential of 44% of TN removal due to simultaneous nitrification-denitrification.

Operating SVF wetlands with a specific hydraulic loading around 4 $L{\cdot}m^{-2}{\cdot}min^{-1}$ and hydraulic regime with cycles of feed and rest periods of 3.5 d, all wastewater quality parameters meet local legislation release standards in river water bodies. It is important to emphasize that at least two SVF wetland modules in parallel are required for full-scale operation.

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