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UASB TECHNOLOGY AND CONSTRUCTED WETLAND ALTERNATIVE TO SEWAGE TREATMENT OF SMALL COMMUNITIES

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Abstract

In Brazil, there is a deficit of access in relation to sewage services for a large part of the population, and this fact is the one responsible for the contamination of watercourses. This is because to the lack of investment in infrastructure in the country, since sewage treatment plants require high costs and high area demand for deployment, besides the installation of the collection system, which directs the wastewater to treatment. This paper describes the behavior of wetlands as a post-treatment step for anaerobically treated sewage in order to the removal of organic matter, suspended solids, nitrogen compounds, total phosphorus and faecal coliforms. Raw sewage was treated in a Upflow Anaerobic Sludge Blanket (UASB) reactor with a retention time of 8 h and the effluent was used in two biosystems of constructed wetlands (CWs), vertical flow and horizontal flow, operated in series, with crushed stone of different diameters and continuous flow. The UASB reactor with post-treatment with CWs has achieved high overall removal efficiencies for chemical oxygen demand (COD), total suspended solids (TSS) and faecal coliforms: 96.5%, 98.4% and 98.2%, respectively. In relation to total Kjeldahl nitrogen (TKN), ammonia and phosphorus the results were similar to those described in the literature, with mean values of 67.1%, 63.4% and 63.5, respectively. The anaerobic pretreatment reduced the area of the CW by about 60%, and appeared to be helpful to prevent CW clogging and odor problems.

Key words: constructed hybrid wetlands, organic matter removal, nitrogen removal, phosphorus removal, rural areas

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1. Introduction

Brazil has more than 209 million inhabitants, about 46% of wastewater receiving treatment. Despite the lack of sewage collection and treatment, the quality of the effluent from the stations is not sufficient to avoid environmental problems, such as surface water eutrophication (De Sousa et al., 2001). Currently in the country, the process configurations of the Wastewater Treatment Plant (WWTP) are undergoing modification and optimization processes along with the introduction of new operations at the unit, such as polishing effluents to reduce nitrogen (N), phosphorus (P) and resource recovery (Fedler, 2017). However, complex operations and rising operating costs are still some challenges in these WWTP. In this sense, simplified treatment systems in the country can be considered as alternatives to provide a desired level of treatment at a much lower cost compared to conventional mechanical systems.

Many studies have shown good results as treatment ponds. These can be anaerobic, aerobic or facultative. More recently, studies with Pond-In-Pond (PIP) lagoons have been considered an alternative system for the treatment of wastewater with good results of integrated nitrogen removal, however the design criteria must be taken into consideration (Adhikari and Fedler, 2019).

Another system that can be considered is the Upflow Anaerobic Sludge Blanket (UASB) reactor.

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Specifically, in Brazil the use of UASB reactors for sewage treatment was introduced in the 1980s, when projects were initiated by several national research groups and engineers working in the sector. In fact, WWTP composed of UASB reactors followed by aerobic post-treatment generally allow a reduction of the implantation costs (capital expenditure - CAPEX) between 20-50% and of operation (operational expenditure - OPEX) above 50%, when compared to conventional activated sludge stations (Chernicharo, 2018). This is one of the facts responsible for the increase in the levels of sewage treatment in Latin America (Chernicharo et al., 2015).

In this context, it is noted that UASB reactors are the main trend in sewage treatment in Brazil, as single units, or followed by some form of post-treatment. According to a survey conducted by Chernicharo (2018), about 40% of the WWTPs located in the South, Southeast and Midwest regions of Brazil use UASB reactors in their treatment flowchart. This also allows us to state that the country has the largest park of anaerobic reactors in the world, considering the application of technology for the treatment of wastewater.

UASB reactors usually remove around 70% of the influent biochemical oxygen demand (BOD), which usually requires a polishing stage in order to enhance organic matter removal (De Sousa et al., 2001). While this removal may be insufficient for complying with discharge standards in most countries, it does provide an important contribution, reducing land requirements for the post-treatment stage (Da Costa et al., 2013). Constructed Wetlands (CW) are low-cost and eco-friendly technologies that take advantage of natural processes to remove pollutants from the water, generally avoiding the use of chemical products and the input of external energy.

Among the different types of CW, vertical flow (VF) CW have predominant aerobic conditions and often high nitrification rates but low denitrification rates which impair total nitrogen (TN) removal. Combining horizontal flow and vertical flow CW in hybrid systems (El-Khateeb and El-Gohary, 2003). Removal efficiencies in CWs will mostly depend on the hydraulic conductivity of the support matrix, type and amount of microorganisms, oxygen supply for the microorganisms, the substrate chemical characteristics, as well as the region climate and latitude (Saeed and Sun, 2012). Temperature can play an important role on the CWs treatment performance, especially between vertical flow and horizontal flow systems. Horizontal flow systems show a better insulation capacity being less sensitive to temperatures fluctuations. In contrast, vertical flow systems are more sensitive to solar radiation that can promote higher degradation rates (Zhang et al., 2011).

In Brazil there is a large deficit of investments in sanitation, especially in relation to WWTP, which require large investments, high cost of deployment and maintenance, this situation is more critical for isolated communities (Gonzalo et al., 2017; Pelissari et al., 2014; Wu et al., 2011;). On the other hand, the use of constructed wetland systems for wastewater treatment is increasingly accepted as an alternative that is both technically and economically feasible, especially for populations of small communities and isolated areas. These systems are attractive because of their low costs for investment, operation, maintenance and the possibility of removing nutrients (Okurut et al., 1999; Vymazal, 2007). Within this context and the potential that the country has for the application of simplified wastewater treatment systems, the objective of the study was to evaluate the performance of a wastewater treatment system composed of a UASB reactor combined with a small-scale hybrid wetland.

2. Material and methods

The experimental investigation was carried out at demonstration scale. The treatment plant was constructed and operated at the Technological Center of Hydraulics (TCH) located in the Polytechnic School of the University of São Paulo, (São Paulo, Brazil). Wastewater that fed the system came from the housing complex and from the polytechnic school of São Paulo University (CR-USP, São Paulo, Brazil). First, the wastewater received a preliminary treatment (screening and grit removal) then was pumped into the UASB reactor and was subsequently sent to CWs.

UASB reactor is of cylindrical type, with flow Q = 3.0 m^3/h, hydraulic retention time (HRT) of 8 h and useful volume V = 24.5 m^3. The effluent of the UASB reactor was treated in the second part of the system composed of 2 artificial wetlands (VFCW and HFCW), each with a length of 3 m and a width of 1.2 m so that the superficial area was 3.6 m^2.

The wetlands units were constructed in plastered brickwork and operated in series (vertical flow and horizontal flow). Figure 1 shows a schematic representation used in the investigation. VFCW was filled with crushed stone in the following conditions: 20 cm (d_{10} = 31.5 mm, non-uniformity coefficient $d_{60/10}$ = 2.4 and porosity = 0.52), 20 cm (d_{10} = 22.0 mm, non-uniformity coefficient $d_{60/10}$ = 1.2 and porosity = 0.45) and 20 cm (d_{10} = 14.3 mm, non-uniformity coefficient $d_{60/10}$ = 1.2 and porosity = 0.40), resulting in a useful height of 60 cm; length/width ratio = 2.5; surface area = 3.6 m^2; wet volume V= 2.2 m^3; flow Q= 0.2 m^3/d; surface hydraulic loading rate = 0.06 m^3/m^2.d; hydraulic retention time (HRT) (V.porosity/Q) = 5±1 days.

HFCW was filled with crushed stone in the following conditions: 60 cm (d_{10} = 22.0 mm, non-uniformity coefficient $d_{60/10}$ = 1.2 and porosity = 0.45), resulting in a useful height of 60 cm; length/width ratio = 2.5; surface area = 3.6 m^2; wet volume V= 2.2 m^3; flow Q= 0.2 m^3/d; surface hydraulic loading rate = 0.06 m^3/m^2.d; hydraulic retention time (HRT) (V.porosity/Q) = 5 days. They were obtained through donation by the Itubanaiá® nursery, located in the city of Miracatú, state of São Paulo, Brazil. For this study, five emerging macrophyte species were chosen according to the recommendations of Souza and

Lorenzi (2008) (*Pontederia cordata* L.; *Hedychium coronarium; Cyperus prolifer* Lam. and *Colocasia esculenta var. aquatilis*). The seedlings were planted with bare roots, directly in the filling material, planted at density of 4 rhizomes/m^2 (Crites and Tchobanoglous, 1998).

The wetland units were fed continuously with the UASB reactor effluent. At the start of the operation water was added to the system with the aim of removing organic material that might be present in the sand. Table 1 shows the physical and operational characteristics of the wetland systems. The system was operated under constant flow conditions. The UASB reactor was operated at retention times of 8 h and each of the wetlands received a constant flow of the UASB effluent. The UASB effluent was introduced sub-superficially over the entire width of the units by the inlet device shown in Fig. 1.

The performance of the wetlands was evaluated over a period of three months of acclimatization (June to August, winter period in the southern hemisphere) of the macrophytes to wastewater and six months of the experimental investigation stage (August to January, spring and summer in the southern hemisphere.). In the acclimation stage, a cut was performed on the macrophytes (20 cm) after 30 days of system operation. Wetland went into operational stability in 90 days (Fig. 2). The pH and the mean temperature during acclimation were 7.5 ± 3.0 and 22±8 ° C, respectively. After this stage, the experimental investigation was started, presented next. The analyzed parameters were: temperature, pH, COD, NH_4-N, TKN, Total P, faecal coliforms, turbidity, Series of solids and Helminth eggs. Laboratory analyzes were performed at the Sanitation Laboratory of the University of São Paulo (EPUSP, Brazil).

All parameters were determined by methods in accordance with *Standard Methods for the Examination of Water and Wastewater* (APHA, 2005). The differences of the treatment efficiencies of the UASB reactor and Wetland Systems over the nine months operational period were statistically analyzed (ANOVA).

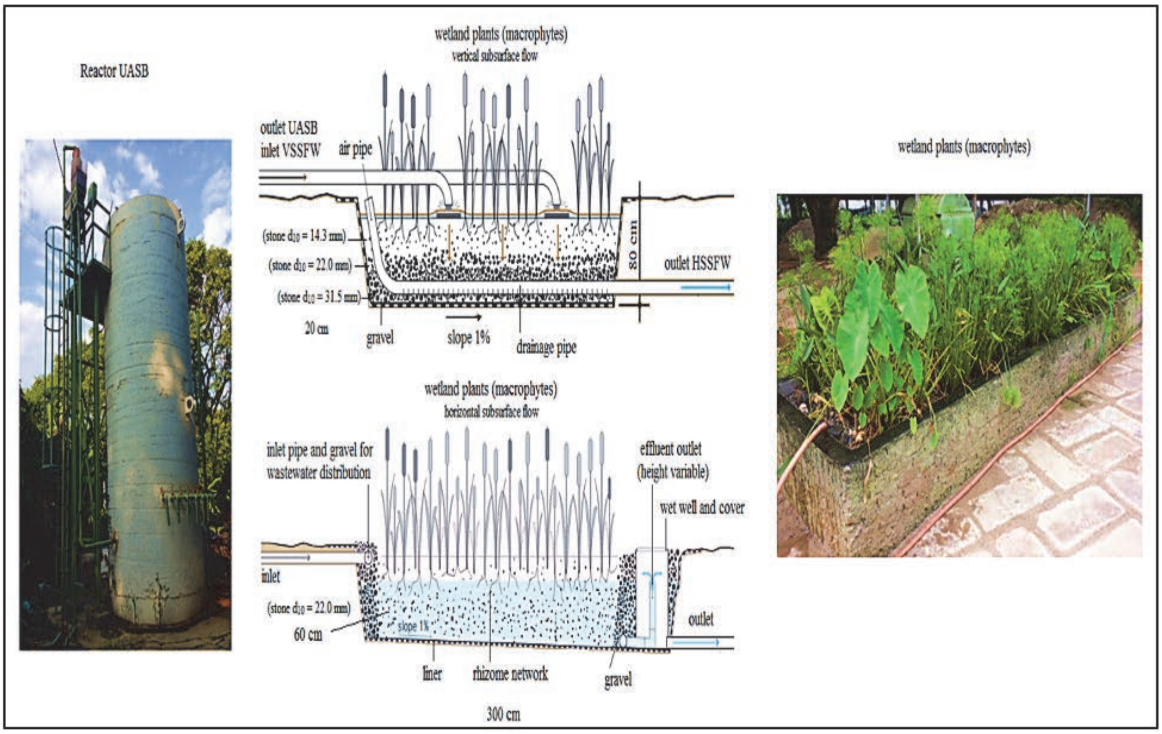

Fig. 1. Schematic layout of the treatment plant

Table 1. Physical and operational characteristics of the UASB reactor and the CWs

UASB reactor		***VFCW***		***HFCW***	
Volume (useful)	24.5 m^3	Constructed Height	1.0 m	Constructed Height	1.0 m
Diameter	2.5 m	Height of the medium	0.6 m	Height of the medium	0.6 m
Height	5.7 m	Length	3.0 m	Length	3.0 m
Hydraulic retention time (V.porosity/Q)	8 h	Width	1.2 m	Width	1.2 m
Volumetric organic load	2.0 kgCOD/m^3.dia	Porosity (void fraction)	0.45±5	Porosity (void fraction)	0.44±2
Flow rate	3.0 m^3/h	Uniformity coefficient	1.2-2.4	Uniformity coefficient	1.2

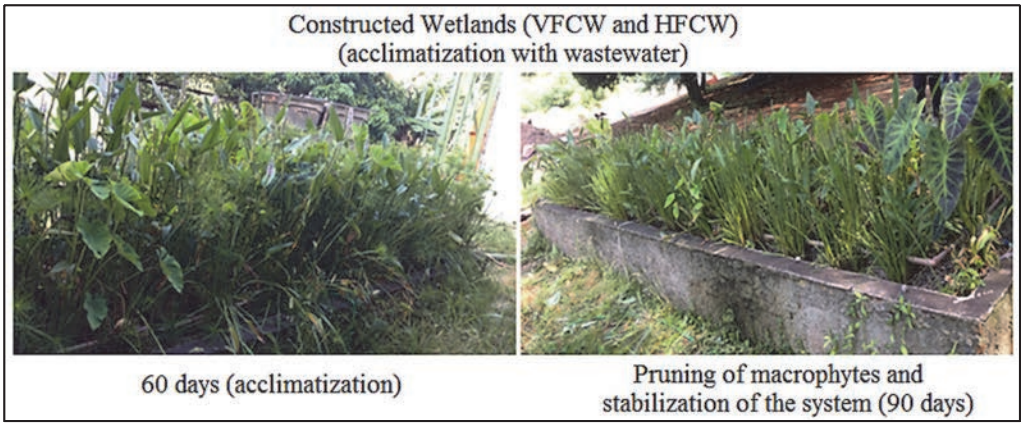

Fig. 2. Acclimatization and pruning of macrophytes CWs

3. Results and discussion

3.1. Performance evaluation and organic matter removal

During the nine months of monitoring, the UASB reactor was operated at a retention time of 8 hours (0,33 d). The applied organic and hydraulic loads on the different wetlands were continuous, as was the liquid retention time in these. Table 2 shows the hydraulic loading rate as well as the loads of organic material, nitrogenous compounds and phosphorus in the two wetlands (VFCW and HFCW). It can be observed that the effluent flow rates are lower than the influent ones. The water losses are due to the evaporation and the transpiration that occurs in the VFCW and HFCW.

The data in Table 3 characterize the digested sewage (effluent from the UASB reactor), which was applied to the wetlands, as well as the effluents of the different wetland units during the six months of operation (Total study time was 270 days).

Table 2. Flow rates, hydraulic loading rate, load of organic and nitrogenous material and applied phosphorus load during the operational period of 06 months for CWs

Parameters	*Units*	*Wetland (VFCH)*	*Wetland (HFCH)*
Surface hydraulic loading rate	m^3/m^2.d	0.06	0.06
Flow rate (influent)	m^3/d	0.20	0.18
Flow rate (effluent)	m^3/d	0.18	0.15
Mean water loss	%	10±5	20±5
Hydraulic retention time	d	5±1	5±0.5
Average organic loading rate	gCOD/m^2.d	13.0	5.2
Average nitrogen loading rate	gTKN/m^2.d	4.0	2.0
Average phosphorus loading rate	gP/m^2.d	0.3	0.2

The UASB reactor was operated at a wastewater temperatures varying from 16°C during winter time to 35°C during summer time. Average percentage removal values of COD and TSS were 65.9% and 68.4%, respectively. In most cases the removal of faecal coliforms in the reactor did not exceed 3 log units. The average percentage removal was 98%. Residual TSS was around 95 mg/L. The values of the nutrient concentrations in the digested sewage (72±12 mg N/L and 5.6 ± 1.8 mg P/L) indicate that the sewage was of medium strength (Tchobanoglous et al., 2003).

The use of UASB reactors for small communities is attractive, as observed, we can obtain a significant removal of organic matter, with low cost of implantation, operation and maintenance when compared to activated sludge processes. However, in the present study and corroborating with the literature, problems with odor and an effluent with a high concentration of solids were observed, on average 95±30 mg TSS/L and virtually no removal of nitrogen and phosphorus, which justifies the need for a after treatment.

The data in Table 3 show that the pH in all the wetlands units tended to increase slightly but always remained in the range of 6.9 to 7.8. Alkalinity increased during the passage through the wetland units, possibly due to the elimination (oxidation) of part of the volatile fatty acids and ammonification of organic nitrogen that escaped from the UASB reactor (166 - 250 mg $CaCO_3$/L). With respect to the TSS, there was clearly a decrease of the concentration in the final effluent, 38 mg/L (VFCH) and 5 mg/L (HFCH).

The influent COD average concentration was 234 mg/L (Output of the UASB reactor) and effluent COD of CWs was 93 mg/L and 24 mg/L, VFCH and HFCH, respectively. The combined UASB + CWs system resulted in an average efficiency of COD 96.5% and 98.4% TSS, a result very close to the aerobic treatment processes. Hybrid wetland contributes a COD removal of 89.7%. The Fig. 3 show the values of the COD concentration during the 6 months operation period of the treatment system and Fig. 4 show the values of the TSS.

Table 3. Average concentrations and standard deviations of the effluent from the UASB reactor (digested sewage) and the effluents from the two wetland units (environmental temperatures ranged from 16 to 35°C)

Parameters	***Unit***	***n number of tests***	***Raw wastewater***	***UASB***	***Wetland (VFCH)***	***Wetland (HFCH)***
COD	mg/L	21	687±107	234±56	93±47	24±13
TSS	mg/L	21	303±100	95±30	33±10	5±4
Alkalinity	mgCaCO$_3$/L	21	199±42	215±33	236±55	266±47
Nitrite	mgN/L	12	< 0.1	< 0.1	0.2±0.1	< 0.1
Nitrate	mgN/L	12	< 0.1	< 0.1	1.8±0.5	< 0.1
TKN	mgN/L	21	82±12	72±12	35±11	27±9
Ammonia	mgN/L	21	63±10	55±8	27±9	23±8
P-total	mgP/L	12	7.4±1.6	5.6±1.8	4.0±1.4	2.7±1.0
pH	-	12	6.9±0.2	6.9±0.2	7.2±0.3	7.5±0.3
Turbidity	NTU	21	150±15	80±22	10±5	3±1
Sedimentantary residue	mL/L	12	32±10	15±5	1.0±1.0	< 1.0
Faecal Coliforms	CFU/100mL	12	9.2x10^5	2.0x10^5	1.6x10^4	4.5x10^3
Helminth eggs	Eggs/100mL	6	n.d.	n.d.	n.d.	n.d.

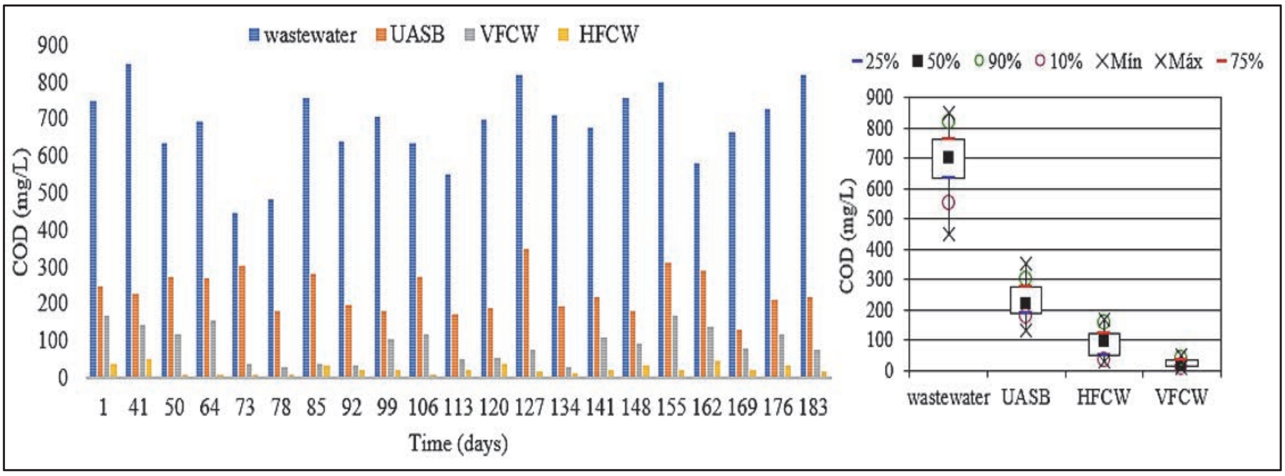

Fig. 3. Values of the COD concentration during the 6 months operation period of the treatment system, composed of UASB reactor + CWs

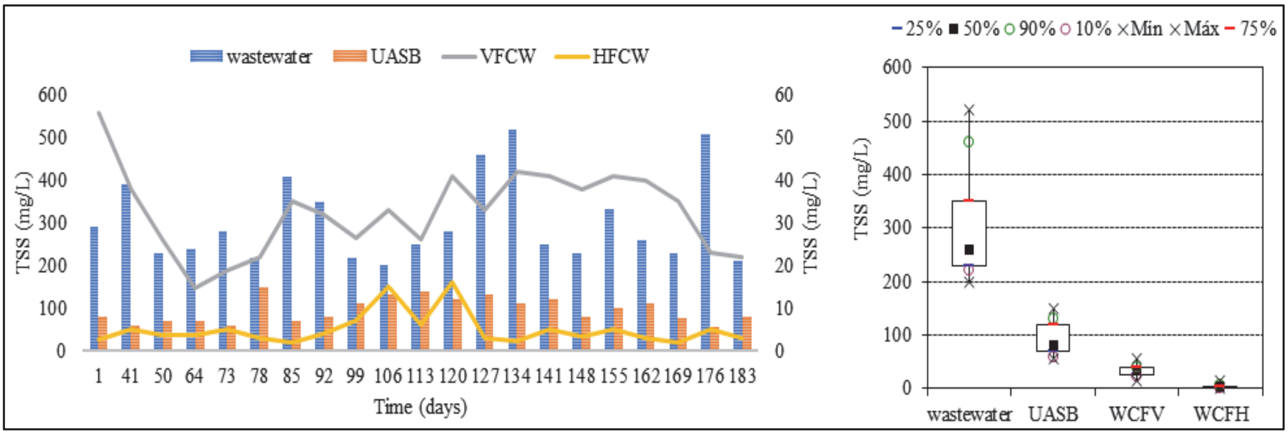

Fig. 4. Values of the TSS concentration during the 6 months operation period of the treatment system, composed of UASB reactor + CWs

The excellent performance of the system as well as the important contribution of the hybrid wetlands to the polishing of the UASB reactor effluent are clearly seen, complementing the evaluation based on effluent concentrations. The large contribution of the wetlands in reducing organic matter from the effluent of the UASB reactor is noted and an important reduction of COD, TSS and sedimentary residue concentrations in the final effluent can be observed.

3.2. Nitrogen removal

The evolution of nitrogen removal in the UASB and CWs systems is shown in Fig. 5 and Fig 6.

The concentrations of nitrite and nitrate in the effluent after the VFCH wetland was 0.2 ± 0.1 mgN/L and 1.8 ± 0.5 mgN/L, respectively. When analyzing these values together with the TKN concentrations, it is observed that there was no accumulation of nitrite and nitrate in the process. With respect to TKN and ammonia, significant removal took place in the unit with vertical flow (VFCW) but much less in the unit Horizontal flow (HFCW). The removal efficiency was 67.1% (UASB+CWs), results of TKN removal in the range of 50-60% and ammonia of 50-80% are reported in the literature (Brix and Arias, 2005; Molle et al., 2006). The nitrogen removal in the VFCW with plants must be attributed to two basic factors: assimilation by microorganisms and macrophytes present in the systems and nitrification due to the probable transport of atmospheric oxygen by the plants, which permits the distribution of oxygen to the rhizomes and plant roots (Cooper et al., 1999). Despite obtaining a high nitrogen removal using the combined vertical and horizontal flow wetland, the nitrogen concentrations in the final effluent are still considered to be quite high, in the process of nitrification activated sludge it is possible to obtain effluents with concentrations below 3.0 mg N/L of ammonia. However, with higher cost of implementation, operation and maintenance.

3.3. Phosphorus removal

Fig. 7 shows the behavior of the UASB reactor effluent and of the CWs (VFCW and HFCW) over the six-month monitoring period. As shown in the figure, the effluent produced by the CWs contained concentration average of phosphorus was 2.7 mgP/L. The average efficiency of removal of total phosphorus was 63.5% (This gain in efficiency evidences the benefit of the association of vertical and horizontal flows of CWs).

The values found are compatible with those described in the literature. Moraes (2012) operated a Hybrid CWs in the French model applied to domestic sewage treatment and obtained phosphorus efficiencies in the range of 45 to 50%. The removal of phosphorus is due to utilization by the plants and microorganisms as well as adsorption and precipitation on the medium (Gonzalo et al., 2017). Therefore, this study did not detect phosphorus uptake by macrophytes. Since traditional wetland systems employ gravel and/or sand as the substrate, P removal was often poor due to their limited P adsorption capacity. Removal of P in all types of constructed wetlands is low unless special substrates with high sorption capacity are used (Vymazal, 2007).

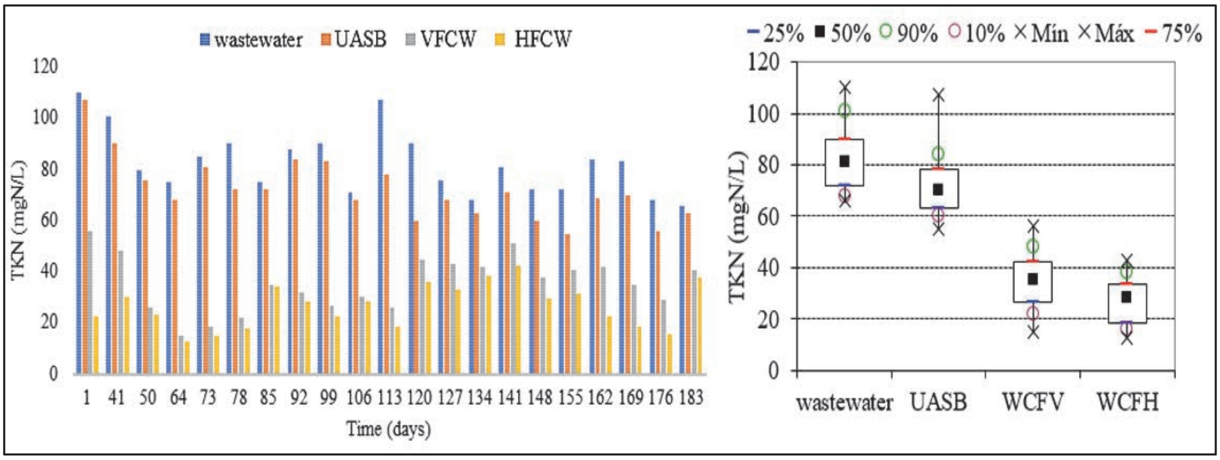

Fig. 5. Values of the TKN concentration during the 6 months operation period of the treatment system, composed of UASB reactor + CWs

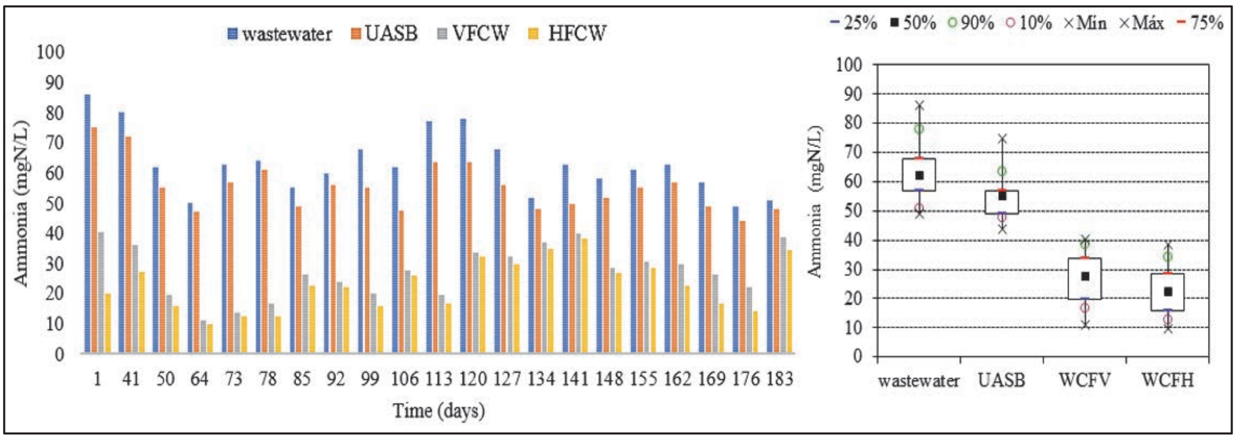

Fig. 6. Values of the ammonia concentration during the 6 months operation period of the treatment system, composed of UASB reactor + CWs

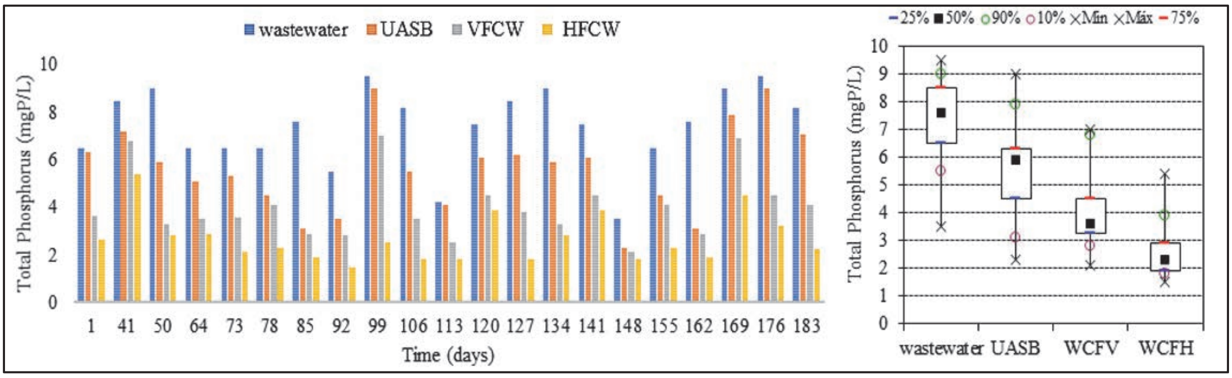

Fig. 7. Values of the total phosphorus concentration during the 6 months operation period of the treatment system, composed of UASB reactor + CWs

4. Conclusions

It is concluded that the system composed of UASB followed by hybrid-built wetlands is in fact an alternative to be considered for the treatment of wastewater. The removal of organic material obtained in the study was similar to mechanized processes such as activated sludge. The nitrogen removal in the proposed arrangement (vertical flow followed by horizontal flow) occurred mostly in the vertical flow system.

The introduction of the horizontal flow system contributed decisively to the removal of organic material, the removal of pathogens and solids, which resulted in a very clarified effluent. In this case, we highlight the potential of the system for applying non-potable water reuse. We consider new studies to be important, with longer evaluation periods, variations in the rates of application of organic and nitrogen loads and new arrangements.

References

Adhikari K., Fedler C.B., (2019), Pond-in-Pond: An alternative system for wastewater treatment for reuse, *Journal of Environmental Chemical Engineering*, **1**, 1-42.

APHA, (2005), American Public Health Association, Standard methods for the examination of water and wastewater, Washington, DC, USA.

Brix H., Arias C.A., (2005), Danish guidelines for small-scale constructed wetland systems for onsite treatment of domestic sewage, *Water Science and Technology*, **51**, 1-9.

Chernicharo C.A.L., (2018), Contribution to the improvement of the design, construction and operation of UASB reactors applied to the treatment of sanitary sewage - Part 1: Topics of interest, *Revista DAE*, **66**, 5-16.

Chernicharo C.A.L., van Lier J.B., Noyola A., Bressani R.T., (2015), Anaerobic sewage treatment: State of the art, constraints and challenges, *Reviews in Environmental Science and Bio/Technology*, **14**, 649-679.

Cooper P., Griffin P., Humphries S., Pound A., (1999), Design of a hybrid reed bed system to achieve complete nitrification and denitrification of domestic sewage, *Water Science and Technology*, **4**, 283-289.

Crites R., Tchnobanoglous G., (1998), *Small and Decentralized Wastewater Management Systems*, McGraw-Hill, New York.

Da Costa J.F., De Paoli A.C., Seidl M., von Sperling M., (2013), Performance and behaviour of planted and unplanted units of a horizontal subsurface flow constructed wetland system treating municipal effluent from a UASB reactor, *Water Science and Technology*, **68**, 1495-1502.

De Sousa J.T., Van Haandel A.C., Guimaraes A.A.V., (2001), Post-treatment of anaerobic effluents in constructed wetland systems, *Water Science and Technology*, **44**, 213-219.

El-Khateeb M.A., El-Gohary F.A., (2003), Combining UASB technology and constructed wetland for domestic wastewater reclamation and reuse, *Water Science and Technology: Water Supply*, **3**, 201-208.

Fedler C.B., (2017), *Water Stewardship: A Sustainable Perspective for Agriculture*, American Society of Agricultural and Biological Engineers, Saint Joseph, Michigan.

Gonzalo O.G., Ruiz I., Soto M., (2017), Integrating pretreatment and denitrification in constructed wetland systems, *Science of the Total Environment*, **584**, 1300-1309.

Tchobanoglous G., Burton F.L., Stensel H.D., (2003), *Wastewater Engineering: Treatment and Reuse*, 4th Edition, Metcalf and Eddy Inc., New York.

Molle P., Liénard A., Grasmick A., Iwema A., (2006), Effect of reeds and feeding operations on hydraulic behaviour of vertical flow constructed wetlands under hydraulic overloads, *Water Research*, **40**, 606-612.

Moraes D.C., (2012), *Influence of seasonality and plants on the reduction of the concentration of pollutants in a system of floods constructed of vertical flow, with pulse feed, used to treat raw domestic sewage*, MSc Thesis, Federal University of Minas Gerais, Minas Gerais/Brazil, On line at: http://www.smarh.eng.ufmg.br/defesas/999M.pdf.

Okurut T.O., Rijs G.B.J., Van Bruggen J.J.A., (1999), Design and performance of experimental constructed wetlands in Uganda, planted with *Cyperus papyrus* and *Phragmites mauritianus*, *Water Science and Technology*, **40**, 265-271.

Pelissari C., Sezerino P.H., Decezaro S.T., Wolff D.B., Bento A.P., Carvalho J.O., Philippi L.S., (2014), Nitrogen transformation in horizontal and vertical flow constructed wetlands applied for dairy cattle wastewater treatment in southern Brazil, *Ecological Engineering*, **73**, 307-310.

Saeed T., Sun G., (2012), A review on nitrogen and organics removal mechanisms in subsurface flow constructed wetlands: dependency on environmental parameters, operating conditions and supporting media, *Journal of Environmental Management*, **112**, 429-448.

Souza V.C., Lorenzi H., (2008), *Systematic Botany - illustrated guide for identifying families of native and exotic phanerogams in Brazil*, Plantarum Institute, São Paulo.

Vymizal J., (2007), Removal of nutrients in various types of constructed wetlands, *Science of the Total Environment*, **380**, 48-65.

Wu S., Austin D., Liu L., Dong R., (2011), Performance of integrated household constructed wetland for domestic wastewater treatment in rural areas, *Ecological Engineering*, **37**, 948-954.

Zhang L., Xia X., Zhao Y., Xi B., Yan Y., Guo X., Zhan J., (2011), The ammonium nitrogen oxidation process in horizontal subsurface flow constructed wetlands, *Ecological Engineering*, **37**, 1614-1619.