Aerobic and anaerobic biodegradability of accumulated solids in horizontal subsurface flow constructed wetlands

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ABSTRACT

This study reports the rate of total solids accumulation (TS) and hydraulic conductivity (HC) in five units of horizontal subsurface (HSSF) flow constructed wetlands (CWs), including unplanted and planted units with four different macrophytes (*Juncus effusus*, *Iris pseudacorus*, *Thypha latifolia L.* and *Phragmites australis*). Two monitoring campaigns were carried out at 1.4 (I) and 2.4 (II) years of operation, at surface loading rates of 2.5 and 4.7 g BOD_5 m^{-2} d^{-1} respectively. Significant differences between units for TS density and most characteristics of accumulated solids were not found. On the contrary, significant differences existed between near inlet and outlet zones as well as between campaigns I and II. Approximately 35% of accumulated solids COD were biodegradable in aerobic conditions, but only 4% in anaerobic conditions, reaching maximum surface biodegradation rates of 4.4-12.0 and 0.2-0.8 g COD m^{-2} d^{-1}, respectively. Thus, promoting aerobic conditions allows a higher SLR without an increase in solids accumulation and prevents clogging. HC was approximately 16% lower in planted units than in the unplanted unit while a similar drainable porosity drop (13-18%) was registered. The results showed that the presence/absence of vegetation and plant species did not significantly affect clogging in HSSF CWs.

Keywords
Clogging, solids accumulation, solids biodegradation, plants, constructed wetlands

INTRODUCTION

Constructed wetlands (CWs) are engineered treatment systems showing a high sustainability potential when are properly designed and maintained. Macrophytes play several roles in engineered ecosystems helping to stabilise the surface of the beds, provide good conditions for physical filtration and insulate the surface against coldness. Plants caused nutrients uptake as well as root-zone oxygen and organic carbon release (Brix, 1997; Tanner, 2001; Vymazal, 2011). Plants also increase microorganism diversity and provide large surface areas for the development of biofilm which is responsible for most of the microbial processes occurring in the wetlands (Button et al., 2015; Chen et al., 2014).

One of the main problems related to the operation of subsurface flow (horizontal and vertical) CWs is clogging of granular media, due to the accumulation of different type of solids, leading to a reduction of the infiltration capacity and porosity of the gravel bed (Hua et al., 2010; Knowles et al., 2011; Pedescoll et al., 2011). Undegraded wastewater solids, microbial biofilm and plant detritus contribute to total solids accumulation in CW granular media. Problems arise when advanced clogging deteriorates the treatment efficiency and drastically reduces the system longevity (Caselles-Osorio et al., 2007; Knowles et al., 2011).

Efficient pre-treatment to reduce influent suspended solids and proper design and operation observing maximum surface loading rates are the main criteria to prevent premature clogging (Winter and Goetz, 2003; Zhao et al., 2009; de la Varga et al., 2013). Other factors such as the presence and type of plants as well as practices that favour oxygenation can affect the clogging process (Chazarenc et al., 2009; Pedescoll et al., 2011).

Plants presence increased the microbial activity (Brix, 1997; Stottmeister et al., 2003) but also increased solids accumulation due to rhizome system (Pedescoll et al., 2011) or to above-ground biomass decay and deposition (Tanner and Sukias, 1995). However the effect of plants on solids accumulation can depend on the species, its rhizome morphology and operational practices (Gagnon et al., 2007). Harvesting has been proposed to limit solids accumulation above-ground biomass and improve nutrient removal (Tanner and Sukias, 1995; Pedescoll et al., 2011; Březinová and Vymazal, 2014). On the other hand, Chazarenc et al. (2009) found that plants presence reduced accumulated solids by 26%, while higher solids accumulation and microbial activity in CWs planted with *Typha angustifolia* than in those planted with *Phragmites australis*. Pedescoll et al. (2013) reported a lower interstitial solids accumulation in CWs planted with cattails but also a lower hydraulic conductivity. Thus, contradictory results about the effect of plant presence on wetland clogging persist.

Anaerobic processes play an important role in horizontal subsurface flow (HSSF) CWs because oxygen transfer from the atmosphere or throughout plant rhizomes appeared to be very limited. Artificial aeration has been proposed in order to increase organics and nutrients removal rates (Wu et al., 2016; Chyan et al., 2016). Anaerobic degradation processes are particularly slow in the case of organic matter hydrolysis and suspended solids removal. However, the effect of enhanced aeration on clogging process remains unclear. Compared to continuous operation of HSSF CWs with permanent water saturation, Pedescoll et al. (2011) indicated similar solid accumulation rates and related clogging indicators in HSSF CWs operated in batch mode, alternating unsaturated and saturated phases, which increased oxygen transfer rates. This could be because a higher oxygen transfer rates may enhance biofilm growth in HSSF CWs (Chazarenc et al., 2009). However, these authors also reported that artificial aeration reduced the accumulation of total solids in non-planted CWs.

The aim of this work is to study the accumulation of solids in HSSF CWs planted with different macrophyte species and determine its biodegradability characteristics in both aerobic and anaerobic conditions, and to answer if promoting aerobic conditions increases or reduces clogging risk. Although HSSF CWs are considered mainly as anaerobic systems, to the best of our knowledge this is the first report in determining anaerobic biodegradability of accumulated solids and comparing it with aerobic degradation rates. The effect of several factors such as the presence or absence of vegetation, the plant species (*Juncus effusus*, *Iris pseudacorus, Thypha latifolia L.* and *Phragmites australis*) and the loading rate on solids accumulation and other clogging related parameters (hydraulic conductivity and drainable porosity) was assessed.

MATERIALS AND METHODS

Pilot plant description

The pilot plant was built in 2009 at the outdoors of the Science Faculty of the University of A Coruña, in A Coruña (Spain). The field pilot plant was constituted of five horizontal subsurface flow CW units in parallel, including an unplanted control unit while the others were planted with a different plant species each: HSSF1 (no vegetated), HSSF2 (*Juncus effusus*) HSSF3 (*Iris pseudacorus*), HSSF4 (*Thypha latifolia L.*) and HSSF5 (*Phragmites australis*). Each CW unit has an overall surface of 12 m^2 (3 m wide x 4 m long), depth of 0.35 m (0.3 m of water depth at the outlet) and 1% slope in the flow direction. The cells were filled with crushed granitic gravel of 6-12 mm in size, except for the inlet and outlet zones (0.5 m long) where large stones (60 mm) were used. Mean porosity resulted in 39.3% and effective (void) volume of 1.36 m^3. A peristaltic pump in combination with a flow distributor fed all the units with a similar influent flow.

The influent to the plant comes from a local sewer receiving wastewaters from one of the faculties of the University of A Coruña and surrounding houses and was pre-treated in an up-flow anaerobic sludge bed digester. After the day 650 of operation, the influent to the plant was supplemented with wine vinegar in order to increase influent concentration and SLR.

The amount and characteristics of accumulated solids were determined twice, after 1.4 (campaign I) and 2.4 (campaign II) years of operation (Table 1). Both campaigns were carried out in winter and lasted for about 2 months, starting with above-ground plant harvesting and characterization and following by sampling of gravel solids, solids characterization and batch biodegradability assays of accumulated solids, and finally determining hydraulic conductivity and drainable porosity.

Procedures for solid sampling and hydraulic conductivity and drainable porosity determination

At each wetland unit, four sample points were considered, two being placed near the inlet and the other two near the outlet. To obtain the samples, a 13 cm diameter steel cylinder was inserted into the gravel until the bottom. Subsequently, the gravel inside the cylinder was removed up using a gardening shovel and placed in a bucket containing oxygen depleted effluent from the corresponding unit. This aims to avoid solids aeration and facilitate solids extraction from the gravel to a water suspension, by stirring and brushing the gravel in water. At the same time, a sample of the liquid that remains in the cylinder was taken. A composite, representative sample was obtained from both gravel extracted solids and liquid fraction. Finally, left and right side samples from the same length position of each unit were mixed and concentrated by decantation in order to obtain a unique sample in which the characteristics were determined in duplicate assays. Samples of solids were characterized for total (TS) and volatile (VS) solids, chemical oxygen demand (COD), aerobic biodegradability by means of biological oxygen demand (BOD) assay, and anaerobic biodegradability (ABD) by means of methane production potential assay.

Hydraulic conductivity was measured in duplicate points near to that of solid measurements throughout the falling head method. A large metallic cylinder of 8 cm internal diameter perforated along the lower 15 cm was inserted into the gravel for a 15 cm depth below the water level. The gravel above the water surface was removed and the cylinder filled with clean water. The falling velocity of water was determined by means of a hydrostatic pressure probe (level transmitter TNS-119-Desin Instruments) and registered by Datataker DT50. Other details and data processing followed the methods described by Pedescoll et al. (2009).

The drainable porosity was determined by empting the beds at a slow flow (less than 4 L min^{-1}, for about 3 h) in order to avoid washout of solids, and measuring the initial and final heights of the water table in the bed and the drained water volume. The quotient between the drained water volume and the drained bed volume gives the drainable porosity. Measurements were carried out for two water table horizons as indicated in Section 3.

Batch biological assays

All BOD and ABD assays and analysis were carried out in duplicate at 20 ºC in a temperature controlled chamber. BOD assays consisted of measuring the pressure drop in the headspace of closed bottles. BOD bottles of 525 mL of total volume from VELP Scientifica were used. The system records the BOD value after each 24-hour period until day 5, provided that they do not exceed the range in the selected scale. If this happens it is necessary to restore the availability of oxygen by opening the system to air, reset the meter and re-start measurement. The same was done after each 5-days period, and the previously accumulated data of oxygen consumption were manually added to the new measurement. In this way, we obtained the BOD curve for a period of 44 days, given both the BOD_5, the ultimate BOD at 44 days (BOD_L) and the BOD profile in time. Nutrients were added to BOD assays as recommended by APHA (2005). Furthermore, 1 mL of the solution allylthiourea (ATU, 1 g L^{-1}) was also added as a nitrification inhibitor in order to suppress the potential of nitrogen oxygen demand.

Anaerobic biodegradability assays were carried out in bottles of 126 mL of volume and 50 mL of liquid volume with a VS concentration of 3 g L^{-1}, without adding any organic substrate but adding macro and micronutrients at a ratio of 1mL L^{-1} of the stock solutions defined by Ferreira et al. (2003). Anaerobic assays were monitored following the head-space gas analysis method (Soto et al., 1993). For this, the composition of duplicate gas phase samples (0.5 mL) was determined in a gas chromatograph equipped with a thermal conductivity detector (TCD). The incubation time was prolonged until the cumulative methane production stopped rising.

Above-ground biomass harvesting and determination

Total recovered biomass (fresh biomass), TS and VS (organic matter) content were determined. Harvesting was carried out by cutting all plants in each unit at about 5 cm from the ground. Following this, the size of harvested plants was reduced to approximately 20 cm long and the material was carefully mixed and divided by quartering, the produced amounts being weighted. Two of the quarters have been further subjected to cutting, quartering and finally grinding in order to obtain representative samples for TS and VS determination.

2.5 Analysis and calculations

Analytical methods were carried out in duplicate as described in Standard Methods (APHA, 2005). Calculations of mean and standard deviation values and regression analysis were carried out in Microsoft Excel, as well as one-way analysis of variance to compare sets of data by means of the probability (*p*) value.

RESULTS AND DISCUSION

Plant performance during measurement campaigns

The plant received a domestic wastewater pre-treated in an up-flow anaerobic sludge bed digester. After the day 650 of operation, the influent to the plant was supplemented with wine vinegar in order to increase influent concentration (COD and BOD were increased by approximately 182 and 127 mg L^{-1}, respectively) and SLR. As indicated in Table 1, campaign I corresponded to low SLR (2.5 g BOD_5 m^{-2} d^{-1}), while campaign II was carried out half a year after the increase of SLR to design conditions (4.4-5.0 g BOD_5 m^{-1} d^{-1}).

Table 1. Measurement campaigns and conditions of plant operation and efficiency.

Campaign (days)[a]	HLR[b] (mm d^{-1})	SLR[b] (g m^{-2} d^{-1})				Removal (%)[b]			
		TSS	COD	BOD_5	TN	TSS	COD	BOD_5	TN
I (495-543)	25.7±0.6	1.7±0.5	5.0±1.4	2.5±0.8	1.4±0.7	89-93	83-88	90-95	29-52
II (809-900)	22.5±0.8	0.8±0.3	7.2±0.7	4.7±0.4	1.0±0.0	65-86	67-88	69-94	16-35

[a] The period of the study went from March 11 2011 (day 495 of operation) to April 9 2012 (day 900 of operation). [b] Hydraulic loading rate (HLR), surface loading rate (SLR) and removal efficiencies correspond to the average values for low SLR conditions (up to 650 d of operation) and design SLR conditions (from day 650 forwards, as reported by Carballeira et al., 2016).

Total suspended solids (TSS) were effectively removed in the anaerobic digester pre-treatment, which delivered a pre-treated effluent with TSS concentration of 67 (I) and 37 (II) mg L^{-1}. Thus TSS SLR was reduced, being below 2 g TSS m^{-2} d^{-1}. Influent loads resulted in low SLR during campaign I while values close to usual design values of 5 g BOD_5 m^{-2} d^{-1} (Puigagut et al., 2007; Pedescoll et al., 2011) were applied during campaign II (Table 1). Both influent TSS concentration and TSS and BOD_5 SLR were below maximum required values for clogging prevention reported in literature (USEPA, 2000; Dahab and Surampalli, 2001; García et al., 2005; Ruiz et al., 2010).

Removal efficiency of BOD_5 and total nitrogen (TN) was typical of HSSF CWs and ranged from 69% to 95% and from 16% to 52%, respectively (Table 1). Other details of plant operation and performance have been reported elsewhere (Carballeira et al., 2016).

Solids accumulation and characteristics

Results for TS accumulation and its characteristics at campaigns I and II are given in Table 2. The obtained results were compared for spatial variation (inlet vs outlet zones), between units and between campaigns through one-way and two-way analysis of variance. In all cases, the amount of accumulated TS increased from campaign I (1.3-3.0 kg TS m^{-2}) to campaign II (3.3-5.3 kg TS m^{-2}) which reflects the impact of operation time and SLR increase. Thus, mean (from inlet and outlet zones) TS content significantly increased from 2.2 kg TS m^{-2} during campaign I to 4.3 kg TS m^{-2} during campaign II ($p<0.002$).

Table 2. Surface density of accumulated solids and main characteristics.

	TS (kg m^{-2})		VS (%)		COD (g g^{-1} VS)		BOD_5 (g g^{-1} VS)		BOD_L (g g^{-1} VS)		ABD (g COD-CH_4 g^{-1} VS)	
CW unit	I	O	I	O	I	O	I	O	I	O	I	O
Campaign I												
Average	2.34	1.98	8.7	7.0	1.80	1.26	0.12	0.13	0.58	0.56	0.112	0.043
	(0.46)	(0.64)	(1.0)	(0.7)	(0.32)	(0.12)	(0.01)	(0.04)	(0.07)	(0.06)	(0.045)	(0.025)
Probability (p) [a]												
Units	0.084		0.501		0.368		0.560		0.810		0.347	
I-O	0.163		0.028		0.018		0.519		0.786		0.029	
Campaign II												
Average	4.64	3.95	11.7	10.1	1.96	1.58	0.23	0.21	0.70	0.51	0.069	0.038
	(0.50)	(0.47)	(2.4)	(2.9)	(0.52)	(0.38)	(0.04)	(0.07)	(0.08)	(0.14)	(0.039)	(0.015)
Probability (p) [a]												
Units	0.866		0.021		0.036 (0.460[b])		0.046		0.163		0.174	
I-O	0.145		0.079		0.053		0.305		0.020		0.080	

I: inlet zone. O: outlet zone. Standard deviation is given in brackets. [a]ANOVA of two factors with only one data per group. [b]Excluding HSSF1 unit. [c]Excluding HSSF2 unit.

TS accumulation was usually higher near the inlet, where there was a TS content approximately 12% to 35% higher than near the outlet, except for HSSF2 unit (both campaigns) and HSSF1 unit in campaign II, which showed similar values in both zones. Instead of this lower TS accumulation near the outlet, overall differences between inlet and outlet zones were not significant (p 0.15). Only slight differences between units were found in campaign I (p 0.08) which completely disappeared in campaign II (p 0.68).

The content in organic matter of accumulated solids (%VS) was reduced and very uniform in campaign I (6-10% VS) in all units but increased to approximately 11-14% during campaign II in HSSF2, HSSF3 and HSSF4 units while remained the same in HSSF1 and HSSF5 units. Thus, significant differences existed between units in campaign II, as well as between inlet and outlet zones in both campaigns (Table 2). Low VS content in accumulated solids is usual although reported values in literature vary extensively from 3% to 89% (de la Varga et al., 2013). Lower values indicate more mineralized solids and are related to lower SLR and plant harvesting which is the case of the present study.

Data on TS accumulation and VS content gives a solids accumulation rate of 1.5 kg TS m^{-2} yr^{-1} **(**0.12 kg VS m^{-2} yr^{-1}) from the beginning of the operation to campaign I and a rate of 2.5 kg TS m^{-2} yr^{-1} **(**0.36 kg VS m^{-2} yr^{-1}) between campaigns I and II. This increase in TS accumulation rate reflects the influence of SLR on solids accumulation. Because of the increase in %VS between campaigns I and II in some units, the overall increase in VS accumulation rate was higher than that of TS accumulation rate. These TS accumulation rates were in the low range of reported values that range from 0.9 to 6.8 kg TS m^{-2} yr^{-1} (Caselles-Osorio et al., 2007; Pedescoll et al., 2011; de la Varga et al., 2013), being in accordance with the low to medium SLR applied in the present study.

The COD content of accumulated solids usually ranged from 1.1 to 2.2 g COD g^{-1} VS, being higher at the inlet zone than at the outlet zone, the differences being higher in campaign I (p 0.02) than in campaign II (p 0.05). Apart from the higher value obtained for HSSF1 unit during campaign II, significant differences in COD/VS ratio between units or between campaigns did not exist. Thus, the average value of COD/VS ratio was 1.6±0.3 g COD g^{-1} VS.

On the other hand, the BOD_5/VS ratio showed a distinctly behaviour as significant differences between inlet and outlet zones were not found ($p>0.3$) while it clearly increased from campaign I (0.13±0.03 g BOD_5 g^{-1} VS) to campaign II (0.22±0.06 g BOD_5 g^{-1} VS), at a significant level (p 0.03). In addition, differences between units appeared during campaign II, the BOD_5/VS ratio being higher for unit HSSF5 followed by unit HSSF1 and finally by the other units. Lower differences were found for BOD_L, the ratio BOD_L/VS showing mean values of 0.57±0.06 g O_2 g^{-1} VS and 0.61±0.14 g O_2 g^{-1} VS for campaigns I and II, respectively. Significant differences in BOD_L/VS between units, campaigns or inlet and outlet zones were not found ($p>0.16$) except between inlet and outlet zones during campaign II (p 0.02, Table 2). Both the ratios BOD_5/BOD_L and BOD_5/COD increased from campaign I (23% and 9%, respectively) to Campaign II (36% and 13%, respectively) which indicate a higher content in easily biodegradable matter in aerobic conditions during campaign II.

Accumulated solids showed higher ABD near the inlet than near the outlet ($p<0.08$) and correlated with COD/VS ratio in campaign I but not in campaign II. Among units, the highest values of ABD were found for HSSF2 unit in both campaigns. Excluding HSSF2 unit, significant differences in ABD between CW units were not found ($p>0.23$). While COD/VS increase from campaign I to campaign II, it is remarkable that ABD decreased from 0.078 to 0.054 g COD-CH_4 g^{-1} VS, at a significant level (p 0.012). Furthermore, while the aerobic biodegradability of the accumulated solids increased from campaigns I to II, as indicated above, the anaerobic biodegradability decreased.

These results indicate that differences in solids accumulation and solids characteristics among CW units were limited. Overall, only ABD appeared to be higher for HSSF2 unit (0.11 g COD-CH_4 g^{-1} VS) than for the other units (0.04-0.07 g COD-CH_4 g^{-1} VS). Other significant differences appeared only for campaign II in which the BOD_5/VS ratio was higher for unit HSSF5 (0.30 g BOD_5 g^{-1} VS) followed by unit HSSF1 (0.25 g BOD_5 g^{-1} VS) and finally by the other units (0.17-0.19 g BOD_5 g^{-1} VS). Significant differences between units for TS density, VS density, and COD/VS ratio were not found. On the contrary, significant differences between near inlet and outlet zones were found for most parameters, except for TS density and BOD_5/VS ratio. In addition, most parameters varied with operation time and SLR, showing significant differences between campaigns I and II, except COD/VS and BOD_L/VS.

Profiles of aerobic and anaerobic biodegradability of accumulated solids

Aerobic biodegradability of accumulated solids has been evaluated in section 3.2 throughout the standardized values of BOD_5 (5 days) and BOD_L (44 days). However, a detailed study of the BOD curve evolution is of great interest. BOD curves are presented in Fig. 1. During campaign I, the slope of BOD curves showed an inflection point at about 14 days of batch assay. BOD curves for near the inlet solids (Fig. 1A) showed a similar profile among them (except HSSF1) that appeared to be different from the profile of BOD curves for near the outlet solids (Fig. 1B). However, the inflection point occurred at approximately the same time in both inlet and outlet samples, as well as for campaigns I and II. The inflection point can be determined by linear regression that can be applied separately to both sides of the BOD curve, below and above the considered inflection point, and selecting the inflection point that gives the highest regression coefficient (R^2).

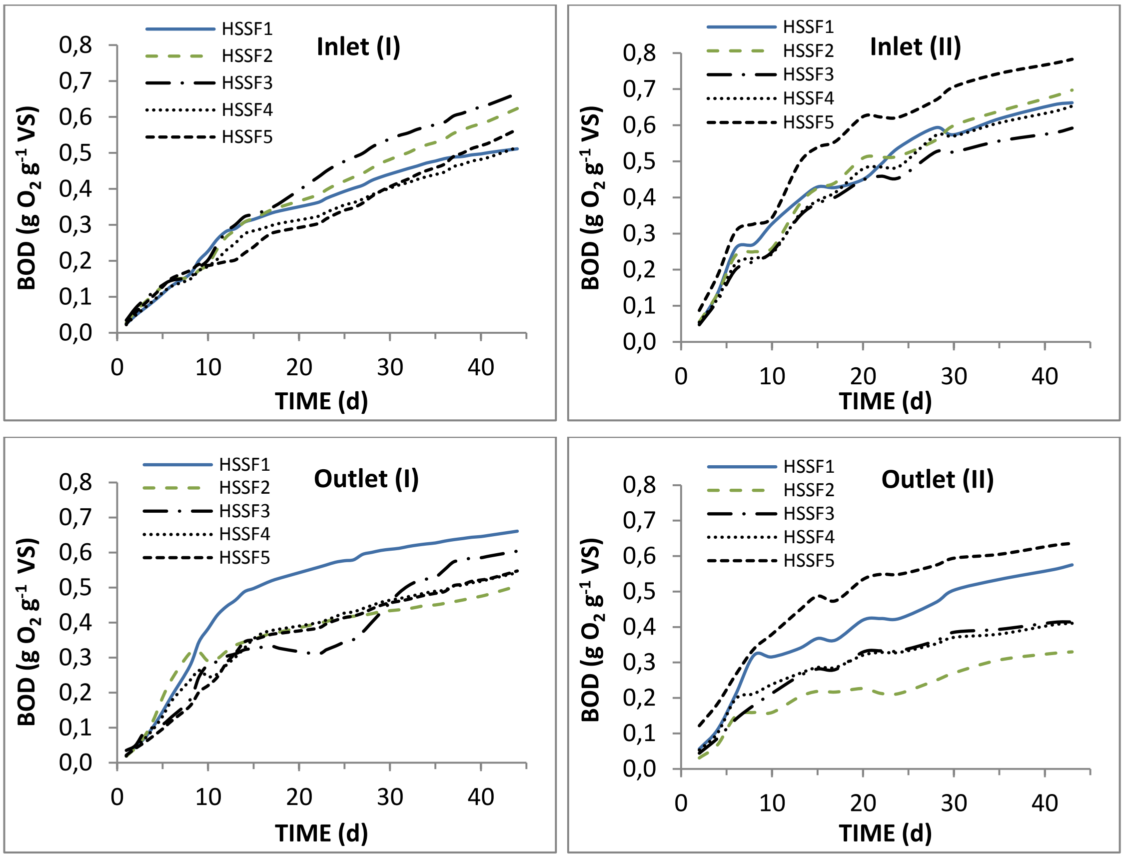

Fig. 1. Time profiles of aerobic biodegradability (BOD curves) of accumulated solids in HSSF units.

This means that the accumulated solids undergone aerobic biodegradation at a high rate over 14 days of batch digestion (initial high rate period), while it was clearly lower afterwards. In fact, while during the first 14 d of digestion the respiration rate was similar to BOD_5, after 14 d it decreased to less than half (25-36%) of the former.

A similar behaviour has been found for anaerobic biodegradation in batch conditions (Fig. 2), but the process took longer and to a less extension than in aerobic conditions. During the first campaign, a continuous methane production was found over at least 160 d of batch digestion. The inflection point was found to be approximately 60 d for near the inlet samples and 45 d for near the outlet samples (Fig. 2A and 2B). For campaign II samples, the inflection point appeared early, at approximately 21 d for both near the inlet and near the outlet solids. After the inflection point, anaerobic biodegradation rates of accumulated solids decreased to about half (43-45%) of that of the initial high rate period.

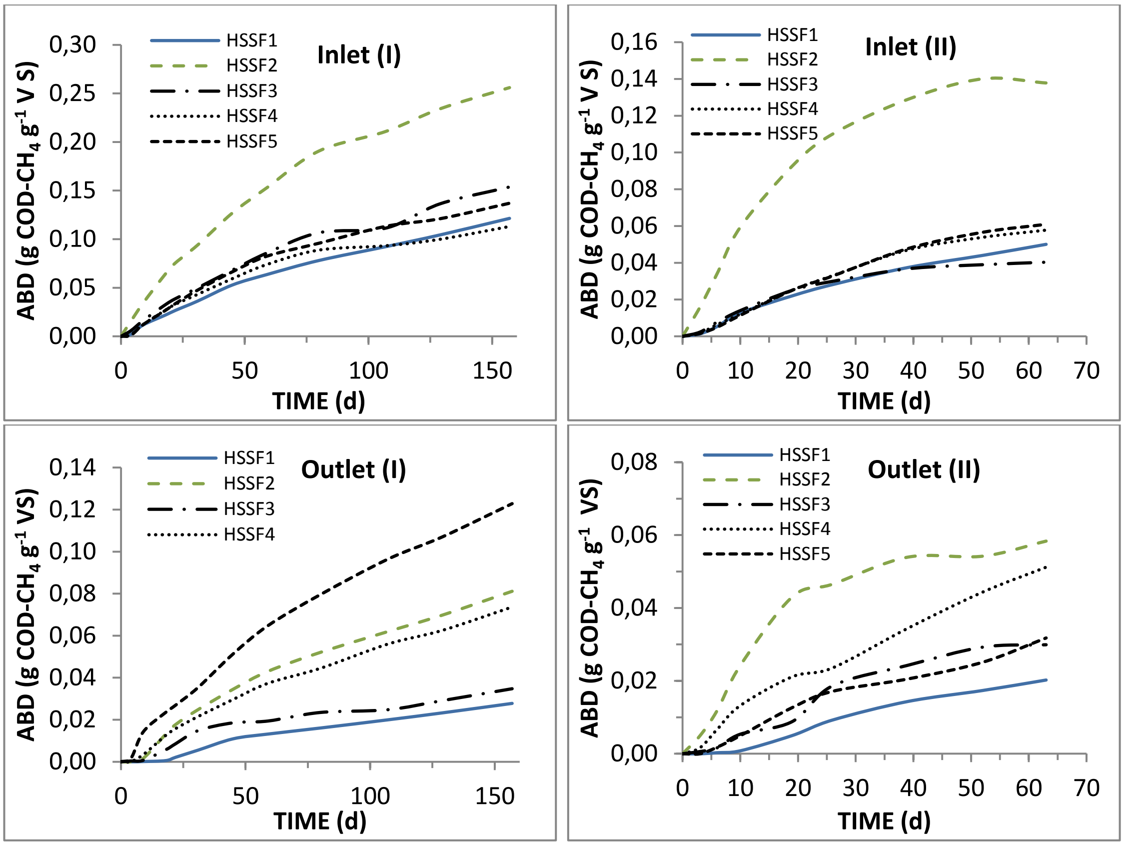

Fig. 2. Time profiles of anaerobic biodegradability (ABD curves) of solids from near the inlet and near the outlet of HSSF units at campaigns I and II.

From Fig. 1 and 2 we calculated the biodegradation rates during the initial high rate period (14 d for BOD assays and 21-60 d for ABD assays as indicate above) by linear regression. The obtained rates were transformed to percentage COD removed during the initial high rate period (R1) throughout the following equations:

$$\text{BOD-R1 (\%COD)} = (\text{BOD}_{R1} \cdot t_{R1} / \text{COD}) \cdot 100$$
$$\text{ABD-R1 (\%COD)} = (\text{ABD}_{R1} \cdot t_{R1} / \text{COD}) \cdot 100$$

where BOD-R1 and ABD-R1 are the aerobic and anaerobic biodegradability of accumulated solids in terms of COD percentage, BOD_{R1} (g O_2 g^{-1} VS d^{-1}) and ABD_{R1} (g COD-CH_4 g^{-1} VS d^{-1}) are the slope of biodegradations curves during the time t_{R1} (d) of initial high rate period, and COD (g O_2 g^{-1} VS) is the COD content of VS given in Table 2.

The obtained results are presented in Fig. 3. The total biodegradability in aerobic and anaerobic conditions is directly obtained from BOD_L and ABD data in Table 2. As we can see in Fig. 3, approximately 35% of accumulated solids COD can be removed in aerobic conditions during a period of 44 d of batch digestion (Fig. 3A) while this percentage resulted in approximately 20% in a period of 14 d, which constituted the readily biodegradability of accumulated solids. On the other hand, the biodegradation rates in anaerobic conditions resulted much lower, of approximately 4% (overall period, Fig. 3A) and 3% (initial high rate period, Fig. 3B), instead of the very long periods considered.

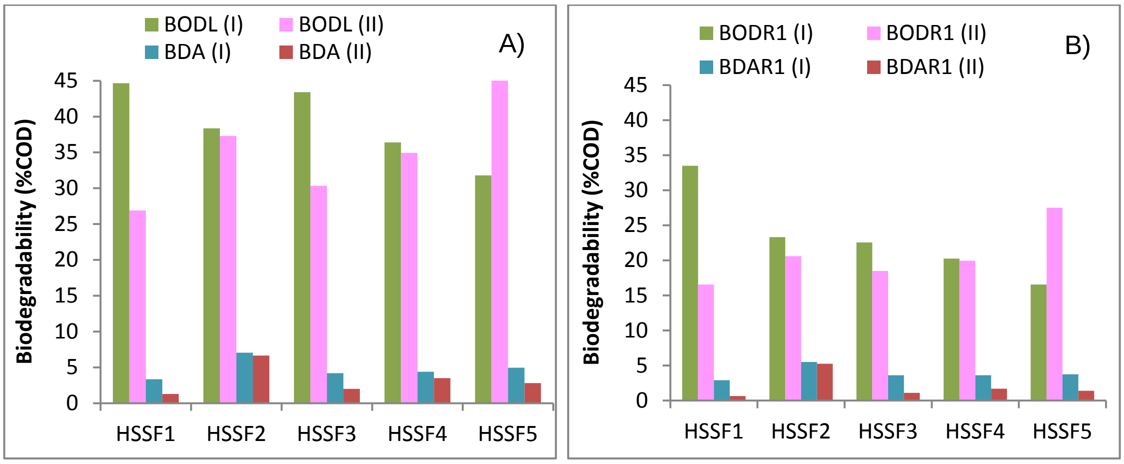

Fig. 3. Aerobic (BOD) and anaerobic (ABD) biodegradability of accumulated solids COD during campaigns I and II (A: total biodegradability obtained from BOD_L and final ABD values, B: readily biodegradability obtained from the initial R1 high rate period).

Surface biodegradation rates of accumulated solids can be obtained. From biodegradation rates during the initial high rate periods (BOD_{R1} and ABD_{R1} from Fig. 2 and 3, respectively), obtained surface biodegradation rates were 4.4 (I) and 12.0 (II) g COD m^{-2} d^{-1}, in aerobic conditions, and 0.2 (I) and 0.8 (II) g COD m^{-2} d^{-1} in anaerobic conditions. Thus, aerobic biodegradation rates were 15-19 times higher than anaerobic biodegradation rates. Expressed in terms of VS, these biodegradation rates corresponded to 2.8-6.9 and 0.1-0.2 g VS m^{-2} d^{-1} in aerobic and anaerobic conditions, respectively.

These surface biodegradation rates may be considered in comparison to applied SLR (Table 1). Anaerobic biodegradation rates of accumulated solids achieved only 4-11% of applied COD SLR (or 6-25% of TSS SLR). As anaerobic conditions dominated in HSSF CWs, an increase in SLR would lead to a higher solids accumulation. On the contrary, aerobic degradation rates of accumulated solids achieved 88-167% of applied COD SLR (or above 165% of TSS SLR), indicating that promoting aerobic conditions would allow a higher SLR without an increase in solids accumulation.

Hydraulic conductivity

Results for HC are shown in Fig. 4. Significantly higher values were obtained for near the inlet zone than for near the outlet zone (p 0.016) while differences between campaigns I and II (p 0.756) or between planted units (p>0.26) were not found. Similar HC values at campaign I (mean 158 m d^{-1}) and II (157 m d^{-1}) contrast with the significant increase in solids accumulation from campaign I to II (Table 2). However, as indicated in section 3.2, even at campaign II, total accumulated solids were in the low range of reported values and far enough from thus causing severe clogging (de la Varga et al., 2013).

Higher values near the inlet than near the outlet contrast with the opposite (although not significant) trend for TS accumulation and the general rule indicating that high solids densities leads to lower hydraulic conductivities (Caselles-Osorio et al., 2007; Pedescoll et al., 2011). However, this behaviour could be related to the different characteristics of the accumulated solids in the inlet and outlet zones and the reduced variation in TS densities. As previously reported, plant density and growth was higher near the outlet than near the inlet (Carballeira et al., 2016). Thus, while influent suspended solids probably accumulated in a major extension near the inlet, fine rhizomes gave a higher contribution to total accumulated solids near the outlet.

On the other hand, considering both campaigns, HSSF1 showed significantly higher values than HSSF4 (p 0.060) and HSSF5 (p 0.053) but not than HSSF2 (p 0.170). Comparisons for HSSF3 cannot be made because of the lack of data. Accordingly to this, mean HC were 195±37 (HSSF1 Inlet), 166±8 (HSSF1 Outlet), 167±23 (HSSF planted units, Inlet), and 134±10 (HSSF planted units, Outlet). The lower HC found in planted units (16%, p 0.056) compared to unplanted unit agree with the result of Pedescoll et al. (2011, 2013) that indicate that below-ground plant biomass significantly contributes to hydraulic conductivity reduction. However, we hypothesized that HC reduction due to below-ground biomass probably occurred during plant cover establishment over the two or three first growing seasons. Afterwards, the below-ground biomass must reach a stead state situation, additional HC reductions due to this factor being not probable. Thus, even if below-ground plant biomass contributes to initial HC reduction, long-term severe clogging could be mainly due to inert and refractory solids accumulation.

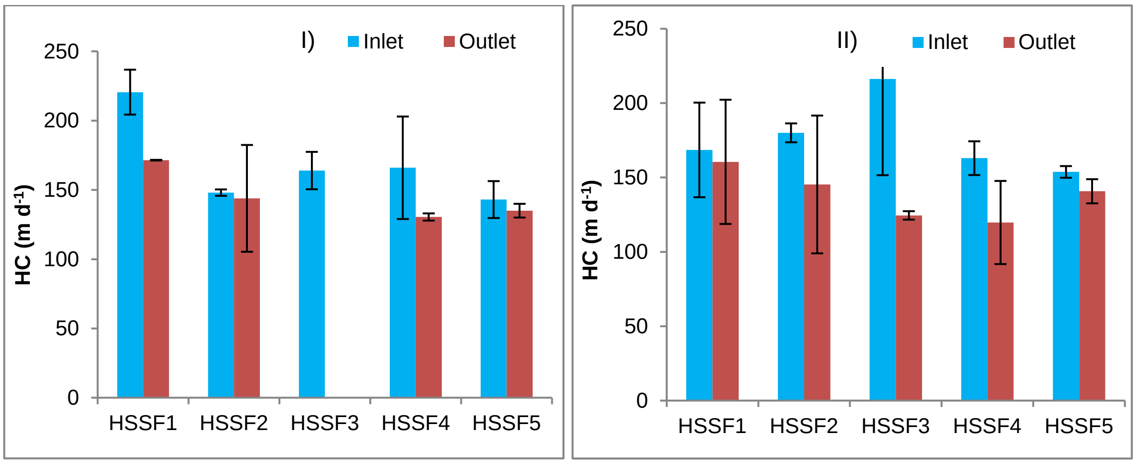

Fig. 4. Hydraulic conductivity of gravel medium at near the inlet and near the outlet zones during campaign I (left) and campaign II (right). Each bar corresponds to the mean value obtained for the same transversal position (n=4) while error bars correspond to standard deviation.

CONCLUSIÓNS

The results of this study indicate that differences in solids accumulation and solids characteristics among HSSF CW units planted with different macrophyte species (*Juncus effusus*, *Iris pseudacorus*, *Thypha latifolia L.* and *Phragmites australis*) or unplanted were limited. Significant differences between units for TS density, VS density, and COD/VS ratio were not found. Anaerobic biodegradability was significantly higher in the unit planted with *Juncus effusus*. On the contrary, significant differences between near inlet and outlet zones were found for most characteristics of accumulated solids as well as between campaigns corresponding to different operation times and loading rates.

TS accumulation significantly increased from 2.2 kg TS m^{-2} during campaign I (1.4 yr, 2.5 g BOD_5 m^{-2} d^{-1}) to 4.3 kg TS m^{-2} during campaign II (2.4 yr, 4.7 g BOD_5 m^{-2} d^{-1}). Derived solids accumulation rates were 1.5 kg TS m^{-2} yr^{-1} from the beginning of the operation to campaign I and 2.5 kg TS m^{-2} yr^{-1} between campaigns I and II, reflecting the influence of SLR on solids accumulation. However, these TS accumulation rates are in the low range of reported values in literature, being in accordance with the low to medium SLR applied. Approximately 35% of accumulated solids COD were biodegradable in aerobic conditions, but only 4% in anaerobic conditions. However, the biodegradation potential during the initial high rate period decreased to approximately 20% and 3%, respectively. Maximum surface aerobic biodegradation rates were at least one order of magnitude higher than anaerobic biodegradation rates. Thus, promoting aerobic conditions in HSSF CWs prevents clogging and allows a higher loading rate without an increase in solids accumulation. A high hydraulic conductivity was registered in this study that was coherent with the relative low densities of accumulated solids and drainable porosity losses found. Nevertheless, the HC was approximately 16% lower in planted units than in the unplanted unit.

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