Field application of a planted fixed bed reactor (PFR) for support media and rhizosphere investigation using undisturbed samples from full-scale constructed wetlands

A. B. Barreto, G. R. Vasconcellos, M. von Sperling, P. Kuschk, U. Kappelmeyer and J. L. Vasel

ABSTRACT

This study presents a novel method for investigations on undisturbed samples from full-scale horizontal subsurface-flow constructed wetlands (HSSFCW). The planted fixed bed reactor (PFR), developed at the Helmholtz Center for Environmental Research (UFZ), is a universal test unit for planted soil filters that reproduces the operational conditions of a constructed wetland (CW) system in laboratory scale. The present research proposes modifications on the PFR original configuration in order to allow its operation in field conditions. A mobile device to obtain undisturbed samples from real-scale HSSFCW was also developed. The experimental setting is presented with two possible operational configurations. The first allows the removal and replacement of undisturbed samples in the CW bed for laboratory investigations, guaranteeing sample integrity with a mobile device. The second allows the continuous operation of the PFR and undisturbed samples as a fraction of the support media, reproducing the same environmental conditions outside the real-scale system. Investigations on the hydrodynamics of the adapted PFR were carried out with saline tracer tests, validating the proposed adaptation. Six adapted PFR units were installed next to full-scale HSSFCW beds and fed with interstitial liquid pumped from two regions of planted and unplanted support media. Fourteen points were monitored along the system, covering carbon fractions, nitrogen and sulfate. The results indicate the method as a promising tool for investigations on CW support media, rhizosphere and open space for studies on CW modeling, respirometry, kinetic parameters, microbial communities, redox potential and plant influence on HSSFCW.

Key words | horizontal subsurface-flow constructed wetlands, planted fixed bed reactor, rhizosphere, redox potential

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INTRODUCTION

Constructed wetlands (CWs) are wastewater treatment systems that enhance purification processes occurring in natural ecosystems, such as wetlands and swamps, where a great number of chemical, physical and biological processes take place in parallel and mutually influence each other. Owing to the high complexity of these processes, CWs have been traditionally considered as a 'black box' where design parameters of interest are related to the inlet and outlet (Stottmeister *et al.* 2003; Kumar & Zhao 2011; Samsó & García 2014). Owing to the complexity of CWs, most available design guidelines are based on empirical rules of thumb, such as those using surface area requirements or simple first-order decay models. However, the elucidation of the processes that govern contaminant degradation and the compartment where they occur is essential to maximize CW removal efficiency and design parameters (Stottmeister *et al.* 2003; Kumar & Zhao 2011; Samsó & García 2014).

Models such as CW2D (Constructed Wetlands 2D; Langergraber & Šimůnek 2005), CWM1 (Constructed Wetland Model No. 1; Langergraber *et al.* 2009) and BIO_PORE (Samsó & García 2013a) are biokinetic models that describe the main processes occurring in CWs and have been used as

doi: 10.2166/wst.2015.238

tools for technology development. As these models show, inside a CW a diversity of microenvironments can establish, presenting variable conditions such as oxygen concentration, redox potential, ionic strength, pH, nutrient availability or contaminant concentration. Depending on these gradients in specific regions within a CW, different microbial communities will develop and therefore different metabolic pathways will be responsible for the removal of pollutants from the liquid (Faulwetter *et al.* 2009; Weber 2013; Samsó & García 2013b).

Many studies have shown that microbial density, diversity and activity are enhanced in the rhizosphere region, suggesting that plants favor the establishment and diversification of microbial communities responsible for contaminant degradation (Münch *et al.* 2005; Gagnon *et al.* 2007; Faulwetter *et al.* 2009, 2013; Langergraber & Simunek 2012). Nevertheless, despite studies showing higher oxygen concentration and redox potential in planted CW and their variation with the photoperiod (Wiessner *et al.* 2005), the main conclusion is that the oxygen transfer rates by roots are not high enough to satisfy the oxygen demand of wastewater in beds with conventional loading rates. As a result, the design guidelines tend to neglect the plant contribution (Nivala *et al.* 2013b).

The planted fixed bed reactor (PFR), developed in the Helmholtz Center for Environmental Research (UFZ), works as a laboratory-scale universal test unit for planted soil filters (Kappelmeyer *et al.* 2002). This reactor allows investigations on the processes occurring inside the rhizosphere with 'ideal' flow conditions in porous media, without hydraulic gradients, ensuring complete mixing in its interior. These characteristics reproduce the subsurface environment around the root zone, where the complex interactions between roots, micro-organisms, support media and pore water occur. Among many of its applications stand out investigations on redox and dissolved oxygen dynamics, metabolic parameters and microbial investigations. The PFR was developed for operation in laboratory-scale under controlled conditions, and there is no record of its use under field conditions to date.

Given these considerations, two main needs for rhizosphere investigations in real-scale CWs can be highlighted: (i) a device that reproduces the bed environment outside the system and its controlled operation in field conditions; and (ii) obtaining undisturbed samples from the original bed. This paper describes the methodology developed to obtain undisturbed samples from CW beds, demonstrates the design and implementation of the PFR in field conditions and discusses its potential applications.

MATERIAL AND METHODS

Study area and experimental unit configuration

The study was conducted in the Center for Research and Training in Sanitation (CePTS), from the Federal University of Minas Gerais (UFMG) and the Water and Sanitation Company of Minas Gerais (COPASA), in Belo Horizonte, Brazil (coordinates 19°53′42″ S and 43°52′42″ W). The CePTS facility is located inside Arrudas wastewater treatment plant, which receives municipal effluent. The region has a Cfa or Cwa humid subtropical climate according to the Köppen classification, with a mean annual rainfall of 1,450 mm and mean annual temperature of 21 °C.

The system in focus consists of horizontal subsurface-flow constructed wetlands (HSSFCW) receiving municipal wastewater after an upflow anaerobic sludge blanket (UASB) reactor. Raw sewage undergoes preliminary treatment (screening with coarse and fine bars and grit removal) before being forwarded to CePTS units. Wastewater, after preliminary treatment, presents mean pH of 6.6 and mean concentrations of total organic carbon (TOC), total nitrogen (TN), S-SO_4^{2-} and P-PO_4^{2-} of 91.4 mg/L, 46.1 mg/L, 6.2 mg/L and 2.0 mg/L, respectively. The UASB reactor, with 22 m³ volume, operates with 80 m³/d flow, resulting in a mean hydraulic retention time (HRT) of 6.6 hours. Part of its effluent is sent to two HSSFCW units operating in parallel, one planted with *Typha latifolia* and another unplanted. Each bed, designed for population equivalent of 50 inhabitants, is 25 m long, 3 m wide, 0.4 m high, and with water level of 0.30 m. Both units have steel slag as support media, with porosity of 40%, d_{10} of 19 mm and uniformity coefficient d_{60}/d_{10} of 1.2. The HSSFCW has operated since June 2007, and each unit receives 7.5 m³/d flow, with real HRT of 1.51 d and 1.42 d for the planted and unplanted units, respectively (von Sperling & de Paoli 2013). Each unit has been designed for a population equivalent of 50 inhabitants and has been the subject of several studies. Da Costa *et al.* (2013), de Paoli & von Sperling (2013) and von Sperling & de Paoli (2013) present various details about the system performance and operation.

PFR adaptation and mobile device for undisturbed samples

The original PFR design (Kappelmeyer *et al.* 2002) was adapted and installed in field conditions, allowing an accurate control and manipulation of operational parameters (Figure 1, center and right). The reactor was built of PVC,

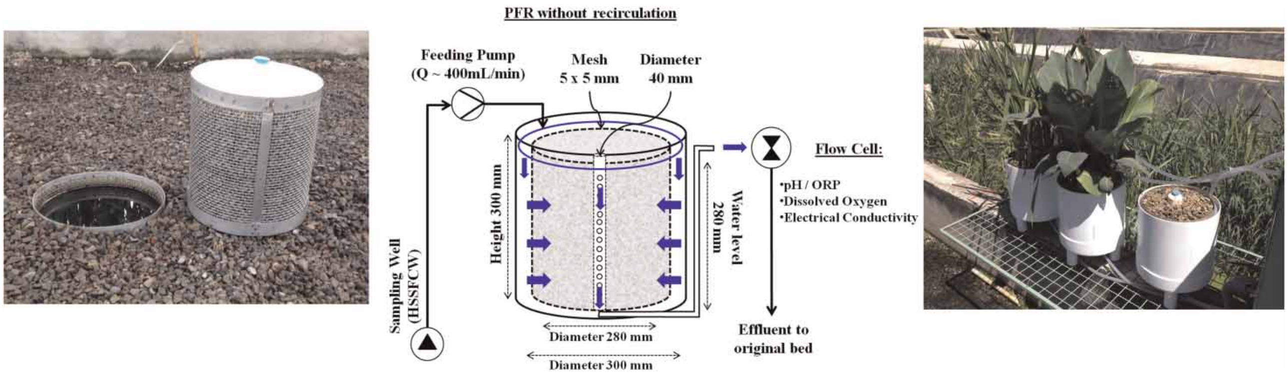

Figure 1 | Mobile baskets for undisturbed sample collection (left), schematics of the modified PFR with main dimensions (center), and the modified planted and unplanted PFRs (right).

having as dimensions 300 mm height and 300 mm diameter. The reactor is fed by a peristaltic pump and the liquid distribution occurs through a hose surrounding the reactor at the top. The liquid outlet is through a hole in the bottom of the reactor, which is connected to the central tube (40 mm diameter) fitted inside the internal basket, containing the undisturbed sample from the original bed, or clean support media. The water level is controlled by an overflow made of an adjustable tube on the reactor's outside. This configuration also permits the installation of a recirculation line that increases the mixing in the reactor. This hydraulic configuration aims at reproducing a continuous stirred tank reactor (CSTR), with radial flow and high mixture, according to the original design of Kappelmeyer *et al.* (2002).

Next, a method was developed to obtain undisturbed samples from the original CW bed. The device consists of mobile baskets (300 mm height and 280 mm diameter) built with stainless steel mesh (5 × 5 mm) with a capacity of 18 L. This method allows the removal and repositioning of the samples, guaranteeing the preservation of the attached biomass under the original environmental conditions in support media (Figure 1, left). One possible approach is to take the core sample, install it inside the PFR, conduct investigations, and return it back to the original bed. This procedure can be performed periodically, allowing a long-term monitoring of support media and rhizosphere evolution. Another possibility is to install the core samples inside the PFR and feed them with interstitial liquid pumped from the original bed. By this approach it is possible to operate the PFR under field conditions guaranteeing that the samples inside the reactor work as fractions of the original system (Figure 1, right).

Both approaches proposed were tested. The first one involved the use of tracer tests to characterize the hydrodynamics of the modified PFR (baskets removed from and replaced in the original bed). The second one was to demonstrate that this methodology creates a reliable fragment of the real-scale system (baskets acclimatized for 130 days in the original bed and installed in PFR). Six PFRs were built and used, three representing a transect in the CW middle zone (13 m from the inlet end) and three representing a transect in the outlet zone (23 m from the inlet end). Two PFRs were planted with *Typha latifolia*, two with *Canna* x *generalis* and two were unplanted (Figures 1 (right) and 2).

To pump the interstitial liquid from the original bed, four sampling wells were installed. Each CW had two sampling wells located in the middle zone (13 m from the inlet end) and outlet zone (23 from inlet end), in the same position where the mobile baskets were installed. The wells were made of PVC pipes (2 cm diameter, 2 m long), with holes of 0.2 cm diameter and 10 cm spacing between holes. Each well was installed perpendicular to the flow direction at 25 cm depth (middle of water level). The interstitial liquid was pumped to the PFR by a peristaltic pump. To define the flow rate of the PFR, a proportional flow to the cross-sectional area of the original bed was adopted, maintaining the same hydraulic surface loading rate in the cross-section. The flow rate of the HSSFCW (cross-section with 0.971 m^2) was 7.5 m^3/d, resulting in a cross-section surface loading rate of 7.72 $m^3/(m^2\ d)$. Thus, the flow rate of the PFR (cross-section with 0.11 m^2) was defined as 0.894 m^3/d or 590 mL/min. The operational flow rate varied from 350 to 450 mL/min, due to operational limitations of the feeding pump. This configuration used sampling methods proposed by Nivala *et al.* (2013a).

Hydrodynamic characterization of the adapted PFR

The original Helmholtz Center design of the PFR operated with a flow rate of 15–40 mL/min. Given the PFR dimensions, complete mixing inside the reactor is guaranteed by a recirculation line of the effluent to the influent, with a flow rate ranging from 45 to 1,200 mL/min (Kappelmeyer *et al.* 2002). Under the present study conditions, complete mixing in the reactor was guaranteed only by the influent flow rate (~450 mL/min), without the need for a recirculation line, simplifying the system for field operation. To validate this adjustment, hydrodynamic tests (peak tracer test) were carried out, with and without recirculation. The tests used a saline tracer (NaCl) and clean water. Saline solution was prepared with 60 g of NaCl diluted in 450 mL of clean water. In total, 12 trials were performed using clean support media and undisturbed samples from transects (middle and outlet zone) in planted and unplanted HSSFCW. Only tests with and without recirculation, carried out with clean steel slag, will be presented here, which are assumed as typical tests.

Salinity estimation was made by correlation with electric conductivity, which was measured by a multiparameter YSI 600XLM probe (YSI, USA), collecting data every 10 seconds. Background salinity was discounted. The tests lasted around 2 hours each. The mass of salt applied was 60 g and the flow rate was 450 mL/min. The total system volume (reactor and hoses) was 10.86 L. The hydrodynamic behavior was evaluated using the 'stimulus-response' and pulse models (Levenspiel 2000; Metcalf & Eddy, Inc 2003). The number of tanks in series was calculated by the gamma function, as described by Kadlec & Wallace (2009).

System monitoring

To demonstrate that the experimental setting reproduces the environmental conditions on real-scale HSSFCW support media, water quality parameters were monitored in the interstitial liquid collected in 14 sampling points (Figure 2), as follows: total carbon (TC), TOC, inorganic carbon (IC), TN, ammonium (NH_4^+) and sulfate (SO_4^{2-}). The carbon and nitrogen analyses were performed in a TOC analyzer, Shimadzu, TOC-VCPN model. The ions were analyzed in an ion chromatograph, Metrohm 850 Professional IC AnCat MCS. These parameters are indicators of the main microbial processes occurring in HSSFCW and are closely related to the operational and metabolic conditions in the support media. Seven monitoring campaigns with simple weekly samples were performed.

Redox potential (E_H) in the interstitial liquid was also monitored using three YSI 600XLM multiparameter probes, coupled to a flow cell, equipped with internal data logger. E_H was measured using Ag/AgCl electrodes and the values were corrected relative to the normal hydrogen electrode by adding +200 mV. The probes stored readings in intervals of 15 minutes for a period of around 4 days in each campaign. In addition to E_H, the probes monitored pH, dissolved oxygen, temperature and electrical conductivity (data not reported here). Equipment calibration with standard solutions was always conducted before deployment.

RESULTS AND DISCUSSION

Adapted PFR hydrodynamic behavior

Tracer tests resulted in an average real HRT ranging from 15 to 20 minutes. The number of tanks in series analysis, using gamma function adjustment (Kadlec & Wallace 2009), showed that the reactor behaves as one tank, when there is recirculation (3:1), and as two tanks in series, when there is no recirculation (Figure 3). The pattern of curves obtained is typical of complete mixing reactors, with the salinity peak being observed closely after the pulse injection. The curves fitted well to the gamma function having *N* (number of reactors in series) values of 1 (with recirculation)

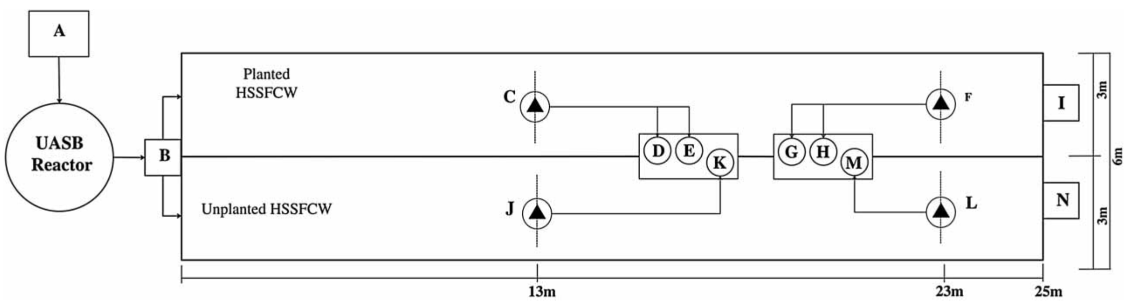

Figure 2 | Schematics of the monitoring points distribution. (A) raw sewage; (B) UASB outlet/CW Inlet; (C) sampling well, planted CW 13 m; (D) PFR 13 m with *Typha latifolia*; (E) PFR 13 m with *Canna* x *generalis*; (F) sampling well, planted CW 23 m; (G) PFR 23 m with *Typha latifolia*; (H) PFR 23 m with *Canna* x *generalis*; (I) planted CW outlet; (J) sampling well, unplanted CW 13 m; (K) PFR 13 m unplanted; (L) sampling well, unplanted CW 23 m; (M) PFR 23 m unplanted; (N) unplanted CW outlet.

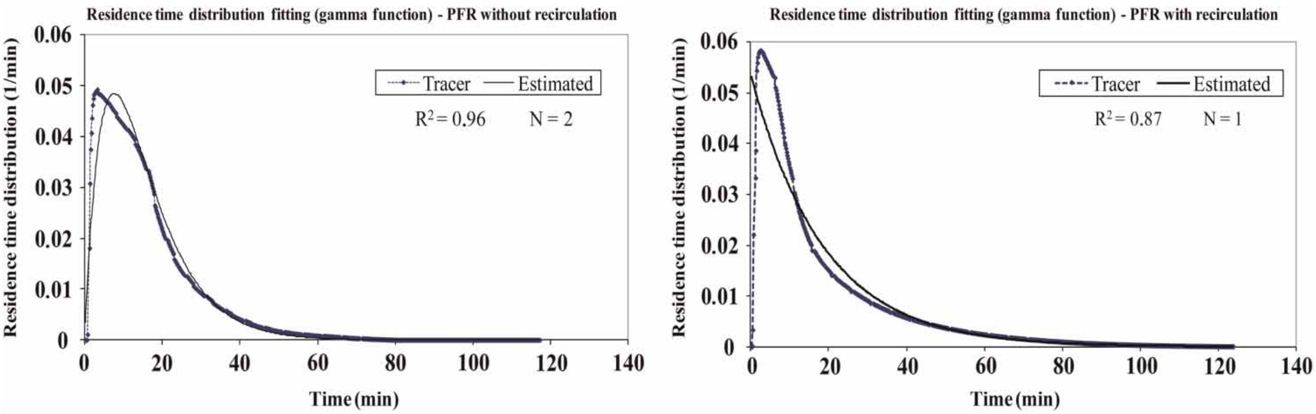

Figure 3 | Hydraulic residence times distribution fitting through gamma function in the tests in the PFR with and without recirculation.

and 2 (without recirculation). These data are confirmed by the volumetric efficiency, which shows a better efficiency in the tests with recirculation. Although results show a better mixing in the reactor with recirculation, one could say that in both tests the reactor was well mixed when operated under a flow rate of 450 mL/min, closely related to the CSTR model, as described by Kappelmeyer *et al.* (2002). It is worth noting that gamma function uses whole numbers, and possibly the hydrodynamic behavior in both tests would be better represented by an intermediate number between 1 and 2. Thus, in order to simplify the experimental setting and system operation in field conditions (no recirculation pump), it was decided to remove the recirculation line (Table 1).

Table 1 | Summary of two tracer tests in the PFR, with and without recirculation (3:1)

Test parameters	Without recirculation	With recirculation
Influent flow rate (mL/min)	450	450
Recirculation flow rate (mL/min)	–	1.500
Recirculation ratio	–	3:1
System volume (L)	10.860	10.860
Support medium porosity (%) – steel slag	0.50	0.50
Theoretical HRT (min)	24.1	24.1
Real HRT (min)	15.0	18.6
Number of reactors in series N	2	1
Coefficient of determination R^2 – Gamma function for N	0.96	0.88
Volumetric efficiency (real HRT/ theoretical HRT)	0.62	0.77
Dead zones fraction	0.38	0.23

Monitoring of real-scale support media and undisturbed samples in the PFR

Monitoring results in the HSSFCW and PFRs are shown in planted and unplanted sequence. Data from carbon fractions, TN and ions are reported by median concentrations during seven monitoring campaigns (Figure 4). Redox potential data are reported on a boxplot graphic covering 28 monitoring campaigns (15 August 2014 to 30 November 2014) with an average duration of 4 days at each monitoring point. Data were collected every 15 minutes (Figure 5). During this period there were varying weather conditions with wide air temperature amplitude (12.1–34.9 °C) and rainfall (data not reported), justifying the amplitude of values obtained.

PFR inlet and outlet parameters analysis (sampling well 13 m/23 m and PFR 13 m/23 m/planted and unplanted) revealed that the experimental setting closely reproduced the environmental conditions in the original support media. The interstitial liquid parameters did not show significant changes when leaving the original bed and passing through the PFRs, exhibiting a linear tendency when flowing through the planted and unplanted systems (Figures 4 and 5). Therefore, it is concluded that the undisturbed samples inside the PFRs behaved as representative fragments of the real-scale system, even outside the bed.

Redox potential results from the support media and PFRs (Figure 5) showed a reducing environment (E_H ranging from −100 to −300 mV), justified by the high oxygen demand exerted by the organic loading rate (mean of 5.12 g biochemical oxygen demand/(m^2 d)), by the solids loading rate (mean of 6.6 g total suspended solids/(m^2 d)) (data not reported) and by the operational history of the system, that brought it to an advanced clogging stage after 7 years of operation.

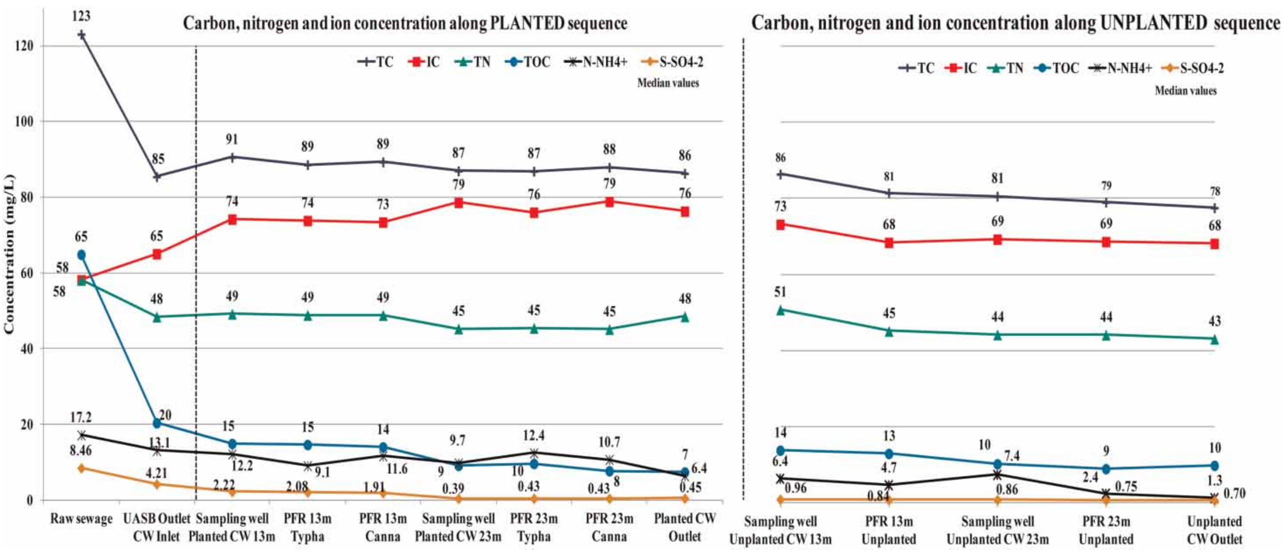

Figure 4 | Carbon, nitrogen and ions concentration along the planted and unplanted sequences. Note that HSSFCW systems receive the same influent (effluent from UASB reactor) and run in parallel.

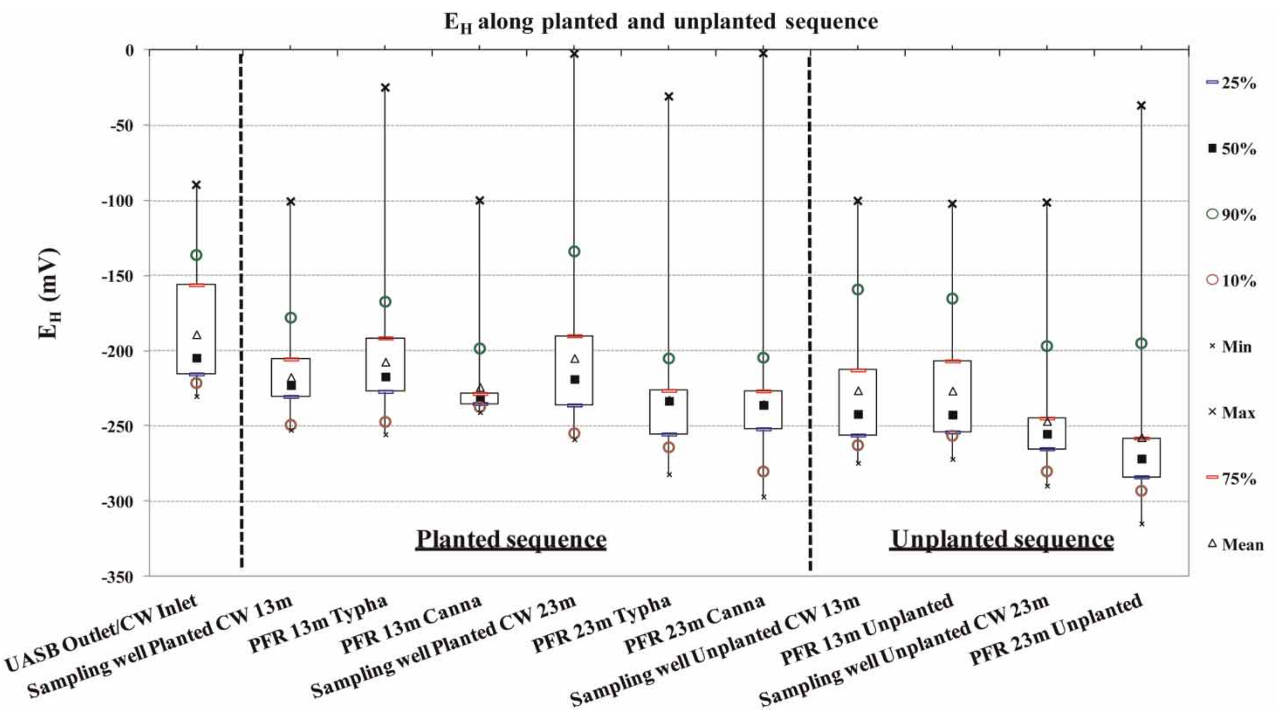

Figure 5 | E_H conditions along planted and unplanted sequence.

These results are consistent with previous investigations in the same system (da Costa *et al.* 2013), with microbial community distribution simulations conducted by Samsó & García (2013b) and with the cartridge theory from Samsó & García (2014). As the support media, in this research, was at an advanced clogging stage, there was a high amount of solids trapped on the bed pores, which exerted oxygen consumption and favored anaerobic microbial groups. As is known, the prevailing respiration process and the associated terminal electron acceptor compound, which depends on the redox conditions prevailing in the wetland environment (low redox/reduced environment), will promote anaerobic processes, such as sulfate reduction and methanogenesis (Faulwetter et al. 2009). The redox range in which the system operates, the low removal efficiency of total nitrogen and ammonium and high reduction of sulfate corroborate these arguments (Figures 4 and 5).

It can be observed that redox potential values in the sampling wells (representing the real-scale system) and in the PFRs (representing a fragment of the system) are distributed along the same range. These results, together with the ions and carbon and nitrogen fractions, show that the adapted PFR, operating as fragments of the real-scale system, is a valid method for research on the support media in CW.

Wiessner et al. (2005) observed daily variations in redox potential and dissolved oxygen driven by light intensity in a PFR, but operating under organic loading rates much lower than the ones from the present study. At high loading rates, and in support media samples under advanced clogging stage, it was not possible to observe such influences of the plants on the system. As the oxygen dynamics is very fast, this element is rapidly consumed by the high oxygen demand exerted by the biomass and by the loading rates applied to the support media (Nivala et al. 2013b), preventing aerobic or anoxic conditions to prevail.

CONCLUSIONS

The support media investigation method using undisturbed samples and field operation of the PFR has revealed itself as a viable tool for studies on CWs. Care should be taken with PFR exposure to weather conditions, due to temperature influence on the surface of the reactors. The use of this methodology for respirometry tests in undisturbed samples meets the recommendations of Andreottola et al. (2007), Ortigara et al. (2010) and Nivala et al. (2013b) and contributes to a method for investigation of oxygen consumption, kinetic parameters and microbial communities in CW support media samples, which would be useful for studies on modeling of wetland systems.

FINAL REMARKS

The authors highlight this methodology as a meeting point between laboratory-scale and real-scale research. Its potential consists of the possibility of performing field-scale investigations on system metabolism in a controlled reactor that reproduces the operational conditions of the support media. Various metabolic indicators can be investigated through this method, such as gas consumption and emission rates, contaminant turnover, and electron acceptor and donor availability. This method still allows monitoring of the temporal evolution of these parameters and obtaining of data for mathematical model calibration/validation. It also can be used to study the effects of different plant species on CW behavior.

ACKNOWLEDGEMENTS

The authors thank the support given by the following institutions: Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq), Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES), Fundação de Amparo à Pesquisa do Estado de Minas Gerais (FAPEMIG), Companhia de Saneamento de Minas Gerais (COPASA), Universidade Federal de Minas Gerais (UFMG), University of Liège (ULg) and Helmholtz Center for Environmental Research (UFZ).

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First received 28 January 2015; accepted in revised form 30 April 2015. Available online 20 May 2015