Assessing the effectiveness of pollutant removal by macrophytes in a floating wetland for wastewater treatment

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Abstract The study aimed to evaluate the removal of pollutants by floating treatment wetlands (FTWs) using an edible floating plant, and emergent macrophytes. All experiments were performed under ambient conditions. Physico-chemical parameters were measured, along with microbiological analysis of biofilm within the roots, water column, and sludge and gravel zone. Nitrification and denitrification rates were high in the water zone of *Azolla filiculoides*, *Lemna minor*, *Lactuca sativa*, *P. stratiotes*, and *Phragmites australis*. Phosphate removal efficiencies were 23, 10, and 15% for the free-floating hydrophytes, emergent macrophytes, and control and edible plants, respectively. The microbial community was relatively more active in the root zone compared to other zones. *Pistia stratiotes* was found to be the efficient in ammonium (70%) and total nitrogen (59%) removal. *Pistia stratiotes* also showed the highest microbial activity of 1306 mg day⁻¹, which was 62% of the total volume. Microbial activity was found in the water zone of all FTWs except for *P. australis*. The use of *P. stratiotes* and the edible plant *L. sativa* could be a potential option to treat domestic wastewater due to relatively high nutrient and organic matter removal efficiency.

Keywords Denitrification · Floating treatment wetlands (FTWs) · Macrophytes · Microbial activity · Nitrification

Introduction

Floating treatment wetlands (FTWs) are efficient systems in pollutant removal, with rooted emergent plants growing on the surface of the water instead of being rooted in the sediment (Headley et al. 2006; Kanyiginya et al. 2010; Oladoja 2016). FTWs are hybrids between ponds and wetlands, and they behave hydraulically similar to storm water detention ponds, while imparting similar treatment processes as a wetland. FTWs require low maintenance (Headley et al. 2006; Dong et al. 2013). They represent a means of potentially improving the treatment performance of the conventional pond systems by integrating the beneficial aspects of emergent macrophytes without being constrained by the requirements of shallow water depth (Dalu et al. 2012; Wanda et al. 2015). Similarly, they also offer some advantages over the conventional sediment-rooted wetlands such as their ability to cope up with variable water depths that are typical of any event-driven storm water system (Kerr-Upal et al. 2000; Manyumba et al. 2009; Srivastava et al. 2014, 2016).

Plant roots play a key role in FTWs, by providing a living, yet high specific surface area for the development of biofilms that contain diverse micro-organism communities responsible for filtering and entrapping fine suspended particles (Smith and Kalin 2000; Merkhali et al. 2015). The thick networks of roots and associated biofilms have proven to be effective in physically trapping particulates within the water column, which subsequently slough off the roots as heavier particles settle. FTWs have been

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studied for many decades all over the world for their application in water quality improvement and storm water control (e.g., Revitt et al. 1997; Kerr-Upal et al. 2000), combined stormwater and sewer overflow (e.g., Van Acker et al. 2005), sewage treatment (e.g. Ghobrial and Siam 1998; Ash and Truong 2003; Todd et al. 2003), acid mine drainage treatment (e.g. Smith and Kalin 2000), piggery effluent treatment (e.g., Ash and Truong 2003; Hubbard et al. 2004), poultry processing wastewater treatment (e.g., Todd et al. 2003), and water supply (e.g., Garbett 2005; Dong et al. 2013). FTWs have proved to be effective in removing suspended solids, nutrients, and heavy metals like copper, zinc, and cadmium (Headley et al. 2006).

The aim of this study was to monitor and assess the performance of an emergent (*Phragmites australis*), floating (*Azolla filiculoides*, *Pistia stratiotes*, *Lemna minor*), and edible (*Lactuca sativa*) plants in a floating wetland system for domestic wastewater treatment. We aimed to determine the wastewater purification potential of the root, the free water column, and gravel and sludge zone in five selected plant systems in the FTW, evaluate the possibility of using the edible plant (*L. sativa*) for wastewater treatment, and determine the microbial activity in multiple compartments of FTWs for different plants, and compare potential nitrification and denitrification activities in the

root, water, gravel, and sludge-associated biofilms for different plant species.

Materials and methods

Experimental setup and operation

The experimental work was carried out continuously at the IHE Delft Institute for Water Education greenhouse during December 2012 to March 2013 (3.5 months; Fig. 1). We ensured that all conditions (i.e., temperature and humidity) were maintained constant including the wastewater for the experiments. The macrophytes were selected as they are common or dominant wetland macrophyte species. Emergent macrophytes *P. australis*; floating macrophytes *A. filiculoides*, *P. stratiotes*, *L. minor*; and edible plant *L. sativa* were used as the experimental plants, with at least three replicates ($n = 3$) per each macrophyte type. *L. sativa* was selected to test the efficiency of treating wastewater. Research has shown that *L. sativa* can be grown in hydroponic systems. The influent tanks were kept 60 cm higher than FTWs mesocosms and the effluent was kept 55 cm lower than the wetlands, and all the tubing covered with aluminium foil to prevent algal growth. The different influent storage buckets were filled with the daily

Fig. 1 Design of the floating treatment wetland experimental setup. Abbreviations: CWG, control; PA, *Phragmites australis*; PA dup., *Phragmites australis* system; PS *Pistia stratiotes*; PS dup., *Pistia stratiotes* system; LM, *Lemna minor*; AF, *Azolla filiculoides*; LS, *Lactuca sativa*

wastewater requirement, corresponding to 1.58 L day^{-1} . Influent and wetland buckets were 25 L (40 cm diameter and 30 cm height), while the effluent buckets were 15 L (21 cm diameter and 19 cm height).

The floating wetland mesocosms buckets bottom were covered with gravel (8–16 cm), to avoid removal of nutrients and clogging problems by fast growing algae. All buckets were randomly arranged. White floaters were used in FTWs with *P. australis* and *L. sativa* for plant root suspension. In addition, temple floaters were also added in all FTWs to act as substrata. Five individual plants of *P. australis*, seven *P. stratiotes*, four *L. sativa*, and 22 g wet weight of *A. filiculoides* and *L. minor* were placed in model wetlands, respectively. During the experiment, 15 g of *A. filiculoides* and *L. minor* were removed from the wetlands, because they were growing rapidly due to nutrient enrichment.

The FTWs were run with primary effluent collected from the Harnaspolder Municipal Wastewater Treatment Facility (WTF), Delft, after the removal of suspended solids and heavy organic solids by screening, skimming, and sedimentation. Primary effluent wastewater was passed through the emergent macrophytes *P. australis*; floating hydrophytes *A. filiculoides*, *P. stratiotes*, *L. minor*; edible plant *L. sativa* and the control. All the treated wastewater from the wetland plants was collected separately in different effluent buckets. The different influent storage buckets were filled with wastewater daily, corresponding to a 1.58 L day^{-1} flow rate of wastewater in the system.

Physico-chemical measurements

The physico-chemical parameters pH, temperature, and electrical conductivity were measured in situ bi-weekly using a multi-probe digital meter (Weilheim & Co. KG, Germany) from each FTW. Dissolved oxygen concentration was measured using a WTW Oxi 340 portable oximeter (Weilheim & Co. KG, Germany). Biological oxygen demand (BOD), chemical oxygen demand (COD), and dissolved organic carbon (DOC) were analysed using the respective standard methods (Eaton et al. 2005). The total organic carbon (TOC) and dissolved organic carbon (DOC) were analysed using a TOC-LVCPN total organic analyser (Shimadzu, Japan).

Water samples were taken from each influent tank and each effluent tank bi-weekly; samples were analysed immediately after collection. Ammonium (NH_4^+ -N) was analysed using the dichloroisocyanurate method (Eaton et al. 2005). Nitrate (NO_3^- -N) and orthophosphate (PO_4^{3-} -P) were determined by ion chromatography using a Dionex ICS-1000 with DV sampler ion chromatography (Eaton et al. 2005). Total nitrogen (TN) was analysed using

TOC-VCPN or TOC-LVCPN total organic analyser (Shimadzu, Japan).

Microbiological analysis

Microbial analysis was started after observable biofilm in the gravel and root (i.e., ~ 3 days after setup). Sampling was conducted bi-weekly. Microbial activity for the root zone, water column, and gravel and sludge was assessed using the fluorescein diacetate assay (FDA) method (Adam and Duncan 2001; Sirage 2012). Biofilm analysis was conducted when sufficient biofilm growth was observed in the roots, water column, and gravel and sludge. The roots were cut carefully with sharp scissors, so that the loosely attached biofilm was not disturbed. 100 mL of water was taken for the analysis from the water column. In the gravel, the biofilms analysis was conducted by randomly selecting gravel and sludge. Extraction of biofilm was done with a handmade sludge extractor. The gravel sample was collected from the bottom of each wetland. Biofilm was determined in terms of total microbial activity. The total microbial activities in four compartments of the FTWs were estimated using the fluorescein diacetate hydrolysis test (Adam and Duncan 2001). Colourless fluorescein diacetate was hydrolysed by both free and membrane bound enzymes, releasing a coloured end product fluorescein which was then measured using spectrophotometer at 490 nm (Adam and Duncan 2001).

Nitrification and denitrification tests

From the roots, gravel and sludge column, and 10 mL of water sample collected from all FTWs, a potential nitrification and denitrification test was carried out to confirm the presence and activity level of nitrifying and denitrifying microbial communities. The test for potential nitrification and denitrification activity in FTWs was carried out according to Persson et al. (2000), Kyambadde et al. (2004a, b) and Ekama and Wentzel (2008). About 10 g of biofilm from roots, water, gravel, and sludge samples from each FTW was incubated aerobically at 30°C in 180 mL containers containing phosphate buffer at pH 8.0 and ammonium sulphate solution on a rotary shaker. In all cases, sodium chlorate was added to inhibit the conversion of nitrite to nitrate. For the denitrification tests, the collected samples were placed in 180 mL plastic bottles; before 60 mL of phosphate buffer, 2 mL each sodium nitrate and glucose solution were added in each sample. The samples were then bubbled with nitrogen gas to create anaerobic conditions. For all procedures, about 10 mL of sample was collected from each of the sampling flasks and the initial NO_3^- -N was measured using anion-exchange chromatography.

Data analysis

Analysis of variance (ANOVA) with Tukey's post hoc analysis was used to test for the differences in physico-chemical variables and microbial activity. Initially, to test for homogeneity of variance, the Bartlett's and Levene's tests were used. The data were found to be normal based on the Shapiro–Wilk test. All statistical analyses were carried out using SPSS version 16 (SPSS Inc. 2016). Pollutant removal efficiency was estimated based on studies by Mallin et al. (2002, 2012), with the inflow and outflow effluent readings sampled bi-weekly over the duration of the study.

Results and discussion

Physico-chemical variables

Physico-chemical parameters measured in the FTWs showed that the average pH in the effluent was 8.3 for all the wetlands, whereas the average influent pH was 7.5 (Table 1). Significant differences ($F = 12.65$, $P = 0.020$) were observed for pH between different wetlands, with varying pH values being recorded for all FTWs and control. This was likely due to the high concentration or low concentration of carbon dioxide (CO_2) as a result of microalgal activity uptake during photosynthesis.

Electrical conductivity (EC) for the primary influent and effluent was 1288 and 1175.5 $\mu\text{S cm}^{-1}$, respectively. Ion removal was found to be significantly different ($F = 8782.93$, $P < 0.001$) among the FTWs. Emergent macrophytes showed high EC in comparison to free-floating hydrophytes and *L. sativa*. Vegetation resulted in an increase in ion removal, and interestingly, there was high

ion removal by the control, which may be due to microbial activity inside the FTW.

Dissolved oxygen (DO) concentration varied between 0.6 and 0.9 mg L^{-1} in the primary wastewater, whereas in the control, the DO concentration varied between 0 and 0.2 mg L^{-1} . Dissolved oxygen concentrations among all the FTWs were found to be significantly different ($F = 27.76$, $P < 0.001$). Dissolved oxygen concentration was low due to oxygen increased microbial activity in all the FTWs. Kyambadde et al. (2004a, b, 2005) found that the DO concentrations in pilot-scale FTWs loaded with sewage and with complete vegetation cover were suppressed and remained significantly lower than in adjacent open water basins. Research carried out by Brix (1993) concluded that low DO concentrations were also typical in wetlands dominated by free-floating macrophytes.

Nutrient removal

Removal efficiency of ammonium for all wetlands ranged between 40 and 70% (Fig. 2a–d). The results show that the floating macrophytes were more efficient in removing ammonium than emergent macrophytes (Fig. 2a). Significant differences ($F = 100.32$, $P < 0.001$) in the removal efficiency of ammonium were observed between the different FTWs. Nitrate concentration was lowest in the primary influent during the treatment process in FTWs and nitrate concentration was found to be 3 mg L^{-1} in *A. filiculoides* FTW (Fig. 2b). Significant variation ($F = 11.85$, $P < 0.005$) among the FTWs for nitrate concentration was observed.

Total nitrogen concentration in primary influent ranged between 27 and 47 mg L^{-1} . Most of the FTWs had more than 40% total nitrogen removal efficiency, except for emergent macrophytes (Fig. 2c). The removal efficiency was found to be highest for all floating hydrophytes, with

Table 1 Mean and standard deviation ($n = 3$) of physico-chemical parameters of influent and effluent concentrations in the floating wetlands

	pH	Temperature ($^{\circ}\text{C}$)	Conductivity ($\mu\text{S cm}^{-1}$)	Dissolved oxygen (mg L^{-1})
Influent	7.5 $\pm$ 0.1	14.3 $\pm$ 2.2	1288.0 $\pm$ 165.9	0.70 $\pm$ 0.1
Floating wetlands				
<i>Phragmites australis</i>	8.1 $\pm$ 0.2 ^a	14.9 $\pm$ 1.3 ^a	1235.3 $\pm$ 102.7 ^a	0.15 $\pm$ 0.1 ^a
<i>Phragmites australis</i> system	8.1 $\pm$ 0.2 ^a	13.8 $\pm$ 1.3 ^a	1231.5 $\pm$ 98.9 ^a	0.13 $\pm$ 0.1 ^a
<i>Pistia stratiotes</i>	8.2 $\pm$ 0.3 ^a	13.1 $\pm$ 1.2 ^a	1139.5 $\pm$ 84.1 ^b	0.23 $\pm$ 0.1 ^b
<i>Pistia stratiotes</i> system	8.3 $\pm$ 0.1 ^b	12.1 $\pm$ 1.7 ^b	1154.3 $\pm$ 82.9 ^b	0.18 $\pm$ 0.1 ^c
<i>Lemna minor</i>	8.3 $\pm$ 0.1 ^b	13.7 $\pm$ 0.8 ^a	1194.8 $\pm$ 96.4 ^a	0.25 $\pm$ 0.1 ^b
<i>Azolla filiculoides</i>	8.2 $\pm$ 0.1 ^a	12.4 $\pm$ 1.7 ^b	1180.0 $\pm$ 91.7 ^a	0.25 $\pm$ 0.1 ^b
<i>Lactuca sativa</i>	8.2 $\pm$ 0.1 ^a	14.9 $\pm$ 1.2 ^a	1166.3 $\pm$ 108.5 ^b	0.20 $\pm$ 0.0 ^c
Control	8.4 $\pm$ 0.1 ^b	13.5 $\pm$ 1.5 ^a	1151.75 $\pm$ 82.5 ^a	0.20 $\pm$ 0.0 ^c

Letters indicate significant differences at $P < 0.050$ based on Tukey's post hoc analysis

Fig. 2 Nutrient removal efficiency for the different FTWs: **a** ammonium, **b** nitrate, **c** total nitrogen, and **d** orthophosphate. PA, *Phragmites australis*; PA dup., *Phragmites australis* system; PS, *Pistia stratiotes*; PS dup., *Pistia stratiotes* system; LM, *Lemna minor*;

P. stratiotes having the highest removal of total nitrogen. The phosphate removal efficiencies were 38, 23, and 15% for the free-floating hydrophytes, emergent macrophytes, and *L. sativa*, respectively. Significant differences ($F = 28.78$, $P < 0.001$) were observed for phosphate concentrations among the different treatments. Sekiranda and Kiwanuka (1997) and Kansime et al. (2005) found that gravel rooted systems achieved significantly greater reduction in phosphates concentrations. A study by Van De Moortel et al. (2010) using two constructed floating wetlands (CFWs), including floating macrophyte mats and controls, highlighted that the average removal efficiencies for ammonium, total nitrogen, phosphates, and COD were 35, 42, 22, and 53%, respectively, similar to our study findings.

BOD and COD reduction

Biological oxygen demand (BOD) in influent varied between 86 and 90 mg L⁻¹, with the removal efficiency ranging between 87.2 and 94%, resulting in a significant reduction of lowering of BOD levels. Significant differences ($F = 4739.1$, $P < 0.001$) among the FTWs for BOD were observed. Chemical oxygen demand (COD) concentrations in the influent ranged between 154 and 333 mg L⁻¹, with removal efficiency varying between 50 and 57%. Significant differences ($F = 1782.2$, $P < 0.050$) among all FTWs for COD removal were observed. Van Acker et al. (2005) in Belgium reported a reduction of 33–68% of COD

in sewer water treated using FTWs. The removal efficiency for total organic carbon (TOC) and dissolved organic carbon (DOC) was almost similar, with the average removal efficiency for TOC and DOC ranging from 48 to 53% and 48 to 57%, respectively (Table 2).

In our study, the BOD/COD ratio was found to range between 0.80 and 0.04 in the final FTW readings, with *P. stratiotes* being the most efficient of all the FTWs. Comparable to our findings, DeWalle and Chian (1976) and Samudro and Mangkoedihardjo (2010) reported similar decreases in BOD/COD ratios from 0.70 to 0.04. Thus, the degree of wastewater organic matter stabilisation has a significant effect on wastewater leachate characteristics, resulting in low BOD/COD ratio and fairly high ammonium concentration (El-Fadel et al. 2003). Initial BOD/COD ratios were > 0.4 before decreasing to < 0.1 in some cases, suggesting that the macrophytes were more efficient.

The chemical oxygen demand (COD) and biological oxygen demand (BOD) had similar values (i.e., ranges, respectively) for all FTWs (Table 2). This suggests that the presence and activities of microorganisms that remove organic matter, i.e., the ordinary heterotrophic organisms, were equal in all FTWs. Organics were also removed by physical settling, similar to Ekama and Wentzel (2008), and most of the organic matter was enmeshed within the sludge and settled within the gravel zone. This could be the reason for high microbial activity observed within the gravel zones of all FTWs and controls. The nitrification and denitrification rates were relatively low and did not

Table 2 Comparison of mean BOD, COD, TOC, and DOC removal (%; $n = 3$) by different FTWs including the control

FTW	Removal rate (%)			
	BOD	COD	TOC	DOC
<i>Phragmites australis</i>	89.0 ± 6.0 ^a	52.0 ± 9.8 ^a	51.4 ± 8.1 ^b	57.5 ± 3.2 ^a
<i>Phragmites australis</i> system	89.0 ± 4.0 ^a	50.8 ± 9.6 ^a	45.8 ± 6.0 ^a	50.7 ± 3.9 ^b
<i>Pistia stratiotes</i>	91.0 ± 3.0 ^a	52.8 ± 12.1 ^b	50.9 ± 3.8 ^b	53.0 ± 3.5 ^c
<i>Pistia stratiotes</i> system	89.3 ± 4.5 ^a	50.9 ± 11.7 ^a	50.1 ± 3.6 ^b	52.5 ± 3.6 ^c
<i>Lemna minor</i>	94.5 ± 0.8 ^b	51.7 ± 8.6 ^a	47.0 ± 6.9 ^a	50.5 ± 4.2 ^b
<i>Azolla filiculoides</i>	94.5 ± 1.2 ^b	53.2 ± 4.5 ^b	45.5 ± 2.2 ^a	48.9 ± 2.7 ^b
<i>Lactuca sativa</i>	89.1 ± 4.3 ^a	52.1 ± 9.2 ^a	47.6 ± 4.0 ^a	52.7 ± 3.3 ^c
Control	87.5 ± 0.4 ^a	56.4 ± 9.4 ^b	49.1 ± 2.6 ^b	50.7 ± 2.0 ^b

Letters indicate significant differences at $P < 0.05$ based on Tukey's post hoc analysis

BOD biological oxygen demand, COD chemical oxygen demand, TOC total organic carbon, DOC dissolved organic carbon

reflect the actual behaviour of the FTWs. Similarly, removal values were within the range found by Van Acker et al. (2005), who reported COD concentration reduction of 33–68% for a FTW receiving combined sewer overflow in Belgium, while Revitt et al. (1997) reported a COD reduction of 31% (mean influent concentration of 47 g m⁻³) from airport runoff containing de-icing compounds.

Potential nitrification and denitrification

Potential nitrification and denitrification rate per mL of root, water, and sludge and gravel zone were extrapolated using the whole volume of root, water, and sludge and gravel surface of each wetland system. Based on the nitrification and denitrification rate, the potential nitrification rate could occur in all the FTWs zones (Table 3). Nitrification and denitrification rates were high in the water zone of *L. minor*, *A. filiculoides*, and *P. australis*. *L. minor*

and *A. filiculoides* are plants with small roots and the rapid growth of these plants was controlled by frequent harvesting. The nitrification and denitrification rates were found to be similar to studies by Kyambadde et al. (2004a, b), Ozturk et al. (2005), Ozengin and Elmaci (2007), Patel and Kanungo (2010) and Dong et al. (2013) that worked on the FTWs. This resulted in these FTWs having the upper part of water zone being aerobically conditioned, while the lower part was anaerobic. This phenomenon was not observed in *P. australis* as it is a highly dense plant with floaters which likely reduce contact of atmospheric air with water. The removal of total nitrogen in FTWs highlighted that when the nitrification and denitrification processes were occurring separately in two different zones; as in the case of *P. stratiotes* and *L. sativa*, the total nitrogen removal was better when compared to FTWs, with nitrification and denitrification processes happening in the same zones (i.e., *L. minor*, *P. australis*, and *A. filiculoides*). Orthophosphate removal was

Table 3 Extrapolated potential denitrification and nitrification rate in root, water, sludge, and gravel in *Lactuca sativa* wetland

Components in FTWs	FTWs' zones volume (mL)	FTWs' zones volume (%)	Specific denitrification activity (μg mL ⁻¹ h ⁻¹)	Denitrification activity (μg h ⁻¹ 25 L ⁻¹)	Efficiency rate (%)
Denitrification					
Gravel	1000	23.2	11.7	1138.1	19.8
Sludge	283.6	6.6	1.4	110.6	1.9
Water	3000	69.7	0.4	4254.4	74
Roots	21	0.5	1.1	244.7	4.2
Nitrification					
Gravel	1000	23.2	4.7	3.3	2.5
Sludge	283.6	6.6	0.001	5.8	4.3
Water	3000	69.7	0.002	24.2	18.3
Roots	21	0.5	0.003	99.1	74.9

Table 4 Microbial activities, nitrification, and denitrification rates ($n = 3$) for FTWs at different zones

FTW/zone	Microbial activity		Nitrification rate		Denitrification rate	
	Concentration (mg N day ⁻¹ 25 L ⁻¹)	Rate (%)	Concentration (mg N day ⁻¹ 25 L ⁻¹)	Rate (%)	Concentration (mg N day ⁻¹ 25 L ⁻¹)	Rate (%)
<i>Phragmites australis</i>						
Roots	714.6 ± 56.4	53.7	0.2 ± 0.2	17.8	81.0 ± 6.7	22.1
Water	218.1 ± 12.8	16.4	0.9 ± 0.3	69.1	244.3 ± 15.9	66.6
Sludge	197.2 ± 45.0	14.8	0.1 ± 0.01	3.7	5.4 ± 1.0	1.5
Gravel	201.9 ± 16.0	15.2	0.1 ± 0.04	9.4	35.9 ± 3.5	9.8
<i>Pistia stratiotes</i>						
Roots	381.4 ± 60.3	18.2	5.2 ± 0.9	66.4	4.7 ± 0.3	3.5
Water	1306.0 ± 50.0	62.3	0.0 ± 0.0	0.0	100.4 ± 7.0	74.2
Sludge	87.8 ± 8.7	4.2	2.5 ± 0.5	31.9	2.5 ± 0.1	1.8
Gravel	321.2 ± 28.5	15.3	0.1 ± 0.04	1.7	27.8 ± 2.0	20.6
<i>Azolla filiculoides</i>						
Roots	148.9 ± 13.6	21.2	3.6 ± 0.7	15.6	12.7 ± 1.4	8.5
Water	260.6 ± 15.0	37.1	19.0 ± 2.1	82.8	103.8 ± 6.9	70.1
sludge	20.2 ± 14.3	2.9	0.3 ± 0.1	1.2	3.5 ± 0.2	2.3
Gravel	272.7 ± 32.0	38.8	0.1 ± 0.0	0.4	28.2 ± 5.1	19.0
<i>Lemna minor</i>						
Roots	175.4 ± 12.4	18.5	10.1 ± 1.4	21.6	9.0 ± 1.0	6.7
Water	598.7 ± 60.7	63.3	35.3 ± 3.2	75.4	101.4 ± 4.8	75.0
Sludge	46.1 ± 3.8	4.9	1.2 ± 0.07	2.5	3.6 ± 0.4	2.7
Gravel	126.2 ± 8.9	13.3	0.3 ± 0.0	0.5	21.2 ± 1.1	15.7
<i>Lactuca sativa</i>						
Roots	377.1 ± 22.8	31.9	2.4 ± 0.2	74.9	5.9 ± 0.8	4.3
Water	403.4 ± 27.7	34.2	0.6 ± 0.04	18.3	102.1 ± 5.2	74
Sludge	78.4 ± 10.0	6.6	0.1 ± 0.01	4.3	2.7 ± 0.1	1.9
Gravel	322.3 ± 18.9	27.3	0.1 ± 0.0	2.5	27.3 ± 3.0	19.8
Control						
Water	536.4 ± 20.0	61.3	1.5 ± 0.03	80.1	28.0 ± 1.8	45.5
Sludge	93.6 ± 12.9	10.7	0.3 ± 0.01	13.7	30.7 ± 2.7	50.0
Gravel	245.0 ± 30.1	28	0.1 ± 0.1	6.2	2.7 ± 0.6	4.5

satisfactory, similar to a study by Sekiranda and Kiwanuka (1997) who found phosphate removal efficiency of 75%.

Microbial activities

Pistia stratiotes showed the highest microbial activity of 1306 mg day⁻¹ 25 L⁻¹, which is about 62% of the total volume, with plant roots and gravel zones, having 18.2% and 15.3% microbial activity rates, respectively (Table 3). Similarly, a study conducted by Sirage (2012) showed high microbial activity in the water zone for *P. stratiotes*. However, nitrification was not observed to occur in this root zone. The denitrification rate was high at 74.2%, indicating that most of the microorganisms present in the water zone of *P. stratiotes* were denitrifying bacteria. The nitrification rate in the sludge zone of *P. stratiotes* was high

at 31.9%, confirming that the nitrifying bacteria were mostly present in sludge. In addition, the root zone of *P. stratiotes* had good nitrification rates of 66.4% (Table 4). In the case of *L. sativa*, high nitrification rates were found in the root zone (74.9%), whereas the high denitrification rates were found in the water zone (74.0%). The microbial activities were almost equally distributed among the roots, water, and gravel zones in *L. sativa* (Table 4).

Conclusions

Among the various FTWs tested for nutrient and organics removal, *P. stratiotes* was found to be best in ammonium (70%) and total nitrogen (59%) removal efficiency. Phosphate removal efficiency was highest for *P. stratiotes*

(29.5%) and *L. sativa* (24.1%) wetland treatment. The specific microbial activity was higher in the root zone of all the FTWs, followed by sludge, gravel, and water. In case of *P. stratiotes*, the specific microbial activity was highest in the root zone followed by water zone, and gravel and sludge zone. Since the volume of water zone in the bucket was more than other zones (root, sludge, and gravel), the microbial activity was found to be high in the water zone of all FTWs expect for *P. australis* and *P. australis* system. The use of *P. stratiotes* and the edible plant *L. sativa* could be a potential option to treat domestic wastewater as it showed relatively high nutrient and organic removal efficiencies.

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