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Performance Assessment of a Laboratory Scale Prototype Biofiltration System in Tropical Region

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Abstract: Biofiltration systems, as one of the best management practices, have good potentials to improve stormwater quality and hydrology of urban catchments. While biofiltration systems are well-studied in developed countries, the majority of those studies are conducted for temperate climate and there is a lack of lab-scale and field-scale studies on such systems under tropical conditions. This paper focuses on the performance of a lab-scale prototype biofiltration systems in stormwater retention efficiency as well as pollutants removal (including heavy metals and nutrients) from synthetic stormwater reproducing tropical rainfall events. A three-layer sand-based filter media with two different native plants including *Pedilanthus tithymaloides* and *Cyperus alternifolius* was selected for this study. Results showed that the system with *Cyperus* has a better stormwater retention capacity compared to the one with *Pedilanthus*. In addition, the observed infiltration rate in *Cyperus* and *Pedilanthus* were 338 mm/h and 267 mm/h, respectively. The better hydraulic performance in the system with *Cyperus* was attributed to the deeper and more extensive root penetration of this plant (as deep as 800 mm) compared to *Pedilanthus* (as deep as 250 mm). While both systems failed to perform well in removing total nitrogen, they performed significantly better in removing total phosphorus (*Cyperus* and *Pedilanthus* removed 67.3% and 62.5% of total phosphorus, respectively). The statistical analysis of results showed that the top 100 mm layer of filter media is the main contributor to total phosphorus removal. However, no major differences were observed between the two systems in phosphorus removal. Moreover, both systems were also capable of removing the available heavy metals (i.e., Fe, Cu, Mn, Ni, Pb, and Zn) as the removal efficiencies exceeded 90%, except for Fe (76%). Similar to phosphorus, it was concluded that the top layer is the major contributor to the heavy metals removal. Overall, the biofiltration system using *Cyperus* was found to be a successful system for operating under tropical conditions.

Keywords: biofiltration system; stormwater treatment; heavy metals

1. Introduction

In the 20th and 21st centuries, global-scale transformation of rural to urban areas has happened rapidly and by 2050 the percentage of the world's population lives in the urban area and will increase to 66% compared to 30% recorded in 1950 [1]. Urban development has negatively affected both the quantity and quality of water in surrounding water bodies such as groundwater, streams, and lakes [2,3]. The increase in imperviousness in urban areas due to the manmade infrastructures such as paved roadways and buildings has caused flashier hydrographs and a higher risk of flood [4]. On the other hand, human activities such as constructions and transportations have contributed to the excess pollutants such as heavy metals in surface runoff [2,5–8]. Excess phosphorus from decaying plants, birds and animals faeces, detergents, and fertilizers is another example of pollution in urban water bodies [9]. Based on a case study in an urban catchment, Todeschini [10] concluded that peak flows, volume, and pollutant loads increased significantly after conversion of 33% of nonurban pervious area to the impervious one. To overcome these issues, water sensitive urban design (WSUD), which is an approach for engineering design, can be used to minimize the stormwater pollution and maximize water resources recharge in urban areas through an integrated urban water planning approach. WSUD is developed and practiced in Australia and is conceptually similar to the Low-impact development (LID), which is common in USA and Canada.

Biofiltration systems, as one of the BMPs, have been well-practiced in developed countries such as the United States and Australia. Biofiltration systems have been found to be effective in improving the quality of stormwater runoff and managing runoff volume [2,6,11–15]. Generally, a biofiltration system consists of a few layers of soil-based media with a different range of soil particles, which has vegetation on top. Such systems can improve the quality and quantity of surface runoff by means of several processes such as absorption, soil filtration, biotransformation, and evapotranspiration [16]. Unlike developed countries such as US and Australia, very limited research is done on the application of biofiltration systems in tropical areas, where rainfall events are expected to be intense and frequent. Since rainfall patterns, soil characteristics, and the severity of stormwater pollution can vary from case to case and such variety can have an impact on a biofiltration system's efficiency [2], there is a push for in-depth studies on tropical biofiltration systems. This is important to better customize the design criteria of biofiltration system based on the local needs and understand its internal mechanisms for further improvement of its performance.

In order to study the tropical biofilters, parameters such as infiltration rate of the filter media and pollutant removal performance would be critical. According to the Australian guidelines [17], the infiltration rate for tropical biofiltration system is set to be higher than 200 mm/h in order to match the intense rainfall of tropical regions. On the other hand, Malaysian guidelines for designing the biofiltration systems recommends an infiltration rate of 100 mm/h [18]. As the infiltration rate is highly related with the filter media composition [19], there have been many research studies on the variation of filter media layer. Hermawan et al. [20] studied the performance of few fine-grained materials including halloysite nanotubes, zeolite, and fly ash as an additive for sand-based filter media in biofiltration system columns. Authors concluded that adding 2% Ultra-Hallo-Pure, which is a halloysite nanotube, performed as the best in both heavy metal removal and high infiltration rate when compared to other additives. Sileshi et al. [21] compared the infiltration performance between sand-peat mixture (1:1) and sand-only filter media with three different compaction methods. It was found that the addition of peat improved the infiltration rate due to its ability to withstand the compaction effect. However, any type of filter media will be exposed to clogging due to segregation and accumulation of fine particles such as dust [19,22]. Therefore, this issue needs to be considered in design criteria if the long-term performance of biofiltration system is targeted.

Plant roots have been found to be the natural solution to clogging issue [19,23]. As the roots grow and penetrate to the filter media, they create pores that reduce the clogging potential. In addition, plant roots play a role in pollutant removal, as plants need certain amount of nutrients to healthily survive and grow. However, it is also necessary to make sure that the plants can withstand different environmental conditions such as dry/wet periods and various pollutants' type and load. Therefore, plant selection is very critical for biofiltration design as it should be specific for each system. Read et al. [24] studied the performance of 20 Australian native plant species for biofiltration system in terms of plant nutrients uptake. Authors concluded that plants can generally contribute to the nutrient removal, while the removal of heavy metals and total suspended solid (TSS) is mainly achieved by filter media. A recent study by Payne et al. [25] discussed the plants' trait and characteristics that may benefit the biofiltration system. According to the authors, extensive root system and high total biomass are the most important factors to improve nitrogen removal efficiency. To cater for wet and dry conditions, higher root length is preferred for the plant to survive. In addition, submerged zone, or saturated zone is also suggested to be installed in the biofiltration system to support plant survivability, especially during the dry period. A study by Blecken et al. [26] showed that the submerged zone has a positive impact on pollutant removal efficiency in biofiltration systems.

Looking into the past study in tropical environment, Goh et al. [27] investigated the nutrients removal efficiency of enhanced bioretention media with additives from various waste materials (cockle shell, newspaper, printed paper, coconut husk, and tyre crumb), using mesocosm study planted with Red Hot Chinese Hibiscus (*Hibiscus rosa-sinensis*). Ong et al. [28] conducted a monitoring study on the first biofiltration system in Singapore. Twelve different plant species were used in the study including *Cyperus alternifolius*, *Typha geniculata*, *Cyathula prostata*, and *Neomarica gracilis*. Authors suggested that frequent maintenance (e.g., periodic trimming and pruning) is required to avoid overly dense vegetation due to the fast growth rate. Additionally, it was found that *geniculata sp.* requires manual watering during prolong drought conditions. Another study in tropical environment was conducted by Salih et al. [29] where three plant species including *Phragmites*, *Ipomoea aquatic*, and *Pistia* were used in biofiltration system for sewage treatment plants. Authors concluded that *Pistia* has the best potential for being used in the biofiltration compared with the other two plants due to its pollutant removal capability (up to 59% nitrate, 37% phosphorus, and 67% suspended solid removal). Overall, based on Malaysia guidelines on stormwater management [18], there are many suggested plant species for use in biofiltration systems. However, very few published works investigate the performance of each suggested plant.

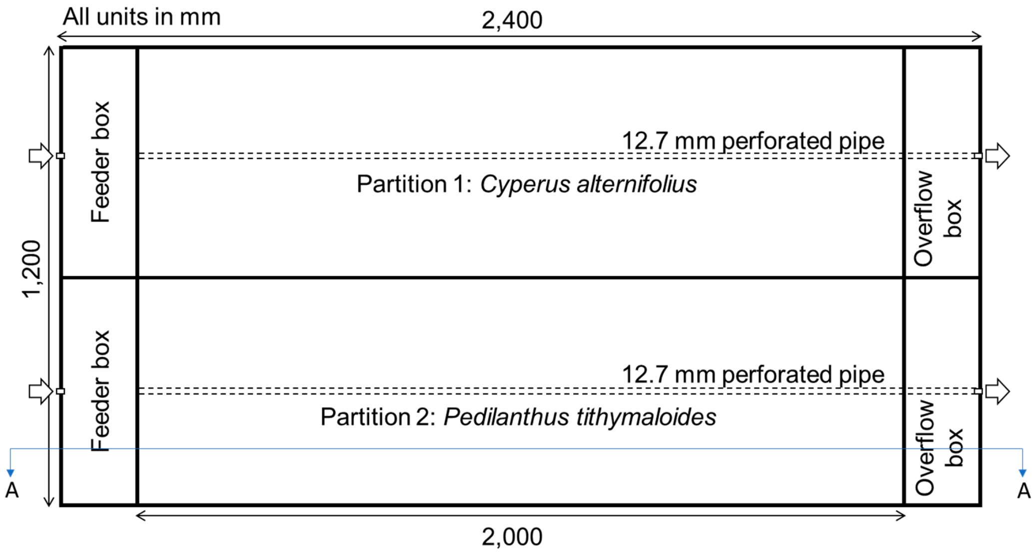
From the experimental set up point of view, most of the previous studies were conducted in soil column scale. However, column study has some limitations. For example, preferential flow of water on the column inner wall can affect the accuracy of measurements for infiltration rate and pollutant removal percentage. On the other hand, plants root growth is somehow forced to the vertical direction while in field scale studies roots may propagate significantly in horizontal direction as well. This may also affect the overall performance of the system in nutrient removal and infiltration rate. Therefore, to better understand the system's performance and functionality in a larger scale (closer to the field scale) with potential three-dimensional root growth and water flow, this study is focused on developing and evaluating a prototype biofiltration system under tropical condition. In this study, a box-shape experimental set up is used to simulate semi-field scale biofiltration system. The objectives of this study were: (1) To establish a prototype biofiltration system and simulate the tropical condition for it; and (2) to evaluate the system efficiency in terms of infiltration rate and pollutants (heavy metals and nutrients) removal.

2. Materials and Methods

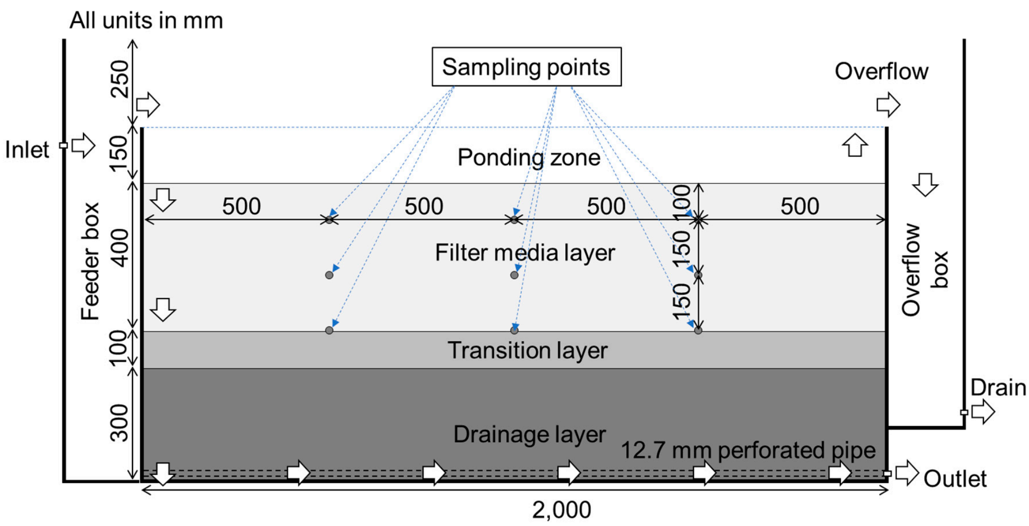
2.1. Prototype Design and Establishment

A prototype box biofiltration system was constructed at Monash University Malaysia green house facility, in early 2017. Figure 1a,b illustrate the plan view and cross-section of the setup, respectively. The prototype consisted of 6 main partitions including 2 feeder boxes, two biofilters, and two overflow boxes. The overall dimensions of the prototype system were 2.4 m (L) $\times$ 1.2 m (W) $\times$ 1.2 m (H) where the biofilter's partitions were filled with 800 mm of engineered layered soil including the filter media layer, transition layer, and drainage layer with the thickness of 400 mm, 100 mm, and 300 mm, respectively (see Figure 1b). A ponding depth of 150 mm was considered in the system, while the excess water could overflow to the overflow boxes through sharp-crested weirs.

The experimental setup consisted of 9 sampling points (3 rows $\times$ 3 columns) on each of its longitudinal walls. These sampling points were equally spaced 500 mm horizontally and 150 mm vertically apart. The sampling ports were located at the distances of 500, 1000, and 1500 mm from the inlet, and at the depths of 100, 250, and 400 mm from the top surface of the filter media. Moreover, two perforated polyvinyl chloride (PVC) pipes with diameter of 12.7 mm were installed 10 mm above the bottom of each partition (in the drainage layer) to collect the infiltrated water. It is noted that no submerged zone was considered in this system as it is not necessary in tropical conditions due to its frequent rainfall events.



(a)



(b)

Figure 1. (**a**) Plan view and (**b**) cross-section (A-A) of the box biofiltration system.

2.2. Filter Media

Three batches of washed sand aggregates with different particle sizes were provided to fill the drainage, transition, and filter layer of both biofiltration systems (the two partitions). According to Payne et al. [30], the design of engineered soil media for a biofiltration system should follow the Equations (1) and (2) to prevent segregation of filter media and clogging.

$$D_{15}\ (\text{transition}) \leq D_{85}\ (\text{filter}) \quad (1)$$

$$D_{15}\ (\text{drainage}) \le D_{85}\ (\text{transition}) \quad (2)$$

where D_{15} and D_{85} are the 15th and 85th percentile of passing particle size. To determine the distribution of particle size, sieve analysis was conducted following ASTM C136 [31]. Figure 2 illustrates the particle size distribution from sieve analysis of the filter media, transition, and drainage layers used in the box biofiltration system. It is worth mentioning that both Equations (1) and (2) were satisfied for the proposed materials to be used for each layer. In addition, it was decided to consider 200 mm/hr as the minimum targeted infiltration rate value, following the recommendation by FAWB [17].

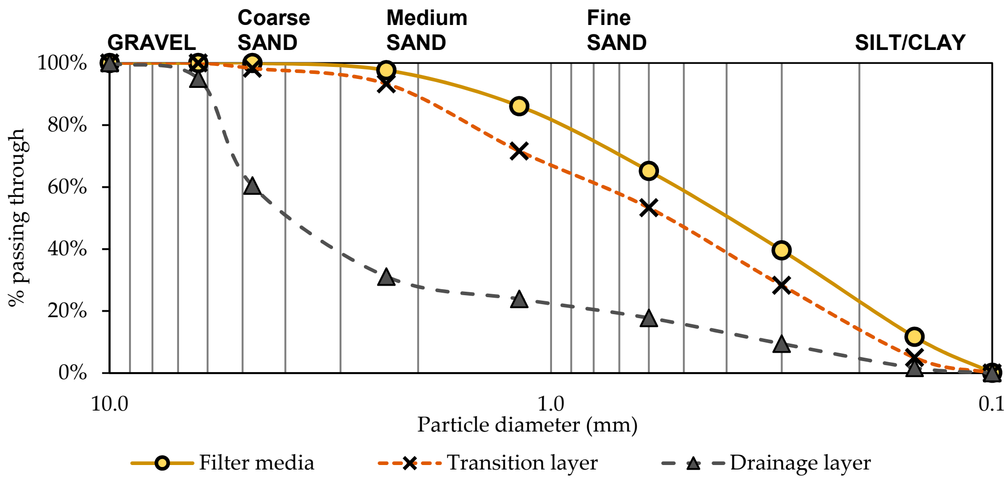


Figure 2. Particle size distribution of filter media, transition, and drainage layers in box biofilter for both plants.

2.3. Synthetic Stormwater

Synthetic stormwater was used in this study to maintain the quality of the inlet pollutant concentration by dissolving salts in 1500 L dechlorinated tap water under ambient temperature of 24–28 °C. Table 1 shows the final influent synthetic stormwater quality with the chemical salt used for each pollutant. The pollutant concentration of synthetic stormwater adopted the stormwater data from literature [18,20,32–34]. Besides total phosphorus and total nitrogen, 6 heavy metals were chosen since they can be commonly found in stormwater [2,32,34].

Table 1. Target pollutant concentrations in artificial inlet stormwater.

Parameter	Chemical Salt	Target Concentration (mg/L)
TN * (as N)	NH_4Cl KNO_3 $C_6H_5NO_2$	4.32
TP ** (as PO_4^{3-})	KH_2PO_4	1.12
Zn	$ZnCl_2$	0.25
Cu	$CuSO_4 \cdot 5H_2O$	0.15
Pb	$Pb(NO_3)_2$	0.14
Ni	$Ni(NO_3)_2 \cdot 6H_2O$	0.03
Fe	$Fe(NO_3)_3 \cdot 9H_2O$	0.86
Mn	$Mn(NO_3)_2 \cdot xH_2O$	0.23

* Total nitrogen, ** Total phosphorus.

2.4. Plant Selection

Ten different native plant species with the potential capabilities for usage in biofiltration system were monitored in the greenhouse of Monash University Malaysia for a period of 6 months during July–December 2016. Each plant was restricted in 8 replicates of pots with natural plant soil. During this period, all plants were irrigated daily with 300 mL (calculated based on a Malaysian design rainfall with 5 min duration, 3-months average recurrence interval (ARI), and intensity of 145.1 mm/hr as recommended by MSMA [18]) of synthetic stormwater (see Section 2.3) to assess their survivability [18,25] under such water quality. Based on this monitoring process the two most healthy plants namely *Cyperus alternifolius* and *Pedilanthus tithymaloides* were chosen to be used in this study. Observation also showed that the two selected plants have different root growth patterns. Therefore, it was hypothesized that such difference could possibly cause differences in their performances in a biofiltration system. *Cyperus alternifolius* has vertical root growth, while *Pedilanthus tithymaloides* has a horizontal root growth pattern. It is worth mentioning that during plants installation in the prototype, the root was introduced below the depth of 150 mm of the filter media to avoid the potential exposure of the roots to the direct sunlight. In addition, the natural plant soil attached to the roots was removed carefully by washing it with tap water before installation of plants in the experimental setup. After installation of the plants, another 6 months was given to the plants to allow their establishment, and to help them grow mature before the commencement of the experiment.

2.5. Sampling and Testing

After installation of *Cyperus alternifolius* and *Pedilanthus tithymaloides* in the experimental set up, they were watered with 600 L of synthetic stormwater every three days prior to any experiments. Watering dosage was calculated based on 3-months ARI rainfall event with duration of 5 min and intensity of 145.1 mm/hr, as suggested by MSMA [18]. The synthetic stormwater was constantly stirred to maintain a uniform concentration throughout the experiment. A DAB KPS 30/16 M centrifugal pump was used to pump the synthetic stormwater to the feeder boxes. The inlet flow rate was measured by a SBG234 flowmeter, which was installed after the pump. In each round of experiment, two water samples were collected from the influent, one at the start and one at the end of experiment to assess the consistency of influent water quality. During each experiment round, 10 samples (i.e., 9 from the side sampling points and 1 from bottom outlet) were collected from each plant box by using 18G syringe needles after the system became saturated. To ensure the accuracy of the water quality measurements, the collected water from each sampling point was split into three portions for the testing stage. The average value of the three readings was then reported as the final value for that sampling point. Considering the two samples from influent, each round of experiment had 12 samples ($12 \times 3 = 36$ readings) for each plant box. In total, six rounds of experiment were conducted with three days gap in between of each two rounds. The concentration removal efficiency (*CRE*) was calculated using Equation (3):

$$CRE = \frac{(C_{in} - C_{out})}{C_{in}} \times 100\% \quad (3)$$

where C_{in} and C_{out} represent the inlet and outlet pollutant concentrations (mg/L), respectively. The infiltration rate (*I*) was calculated using Equation (4) based on the constant-head method ASTM D2434-68 [35].

$$I = \frac{V}{A \times t} \times 1000 \quad (4)$$

where *I* is infiltration rate (mm/h), *V* is the volume of water collected from bottom outlet (m^3), *A* is the cross-sectional area of the box (m^2), and *t* is the total time taken (h). Last but not least, stormwater retention (W_r) was calculated using Equation (5):

$$W_r = \frac{V_{in} - V_{out}}{V_{in}} \times 100\% \quad (5)$$

where W_r is the water retention (%), V_{in} is the inflow volume of synthetic stormwater for each round of experiment (L), and V_{out} is the overflown volume of synthetic stormwater (L).

2.6. Analytical Methods

Total nitrogen (TN) and total phosphorous (TP) were measured using DR6000 ultraviolet-visible spectroscopy (UV-Vis) with HACH methods 10071 and 8190, respectively. Six heavy metals (i.e., Cu, Fe, Mn, Ni, Pb, and Zn) were measured using a Perkin Elmer Optima 8000 Inductively-Coupled Plasma Optical Emission Spectroscopy (ICP-OES) machine. The detection limit for UV-vis and ICP-OES was 0.01 mg/L and 0.001 mg/L, respectively.

2.7. Statistical Analysis

The obtained pollutant concentrations were grouped with respect to the depth from the surface prior to data analysis, and box plots were formed. Descriptive statistics (e.g., mean, median, and quartiles) were calculated and data points outside 1.5 times the interquartile range (IQR) of the grouped depth were classified as outliers and were removed. The normality test was assessed by conducting Shapiro-Wilk test, which is based on the relationship between the data and the corresponding normal scores, and known to provide better power than other normality test [36,37]. If the normality test failed, the data would be assumed to follow a non-Gaussian distribution, and hence the unpaired-non-parametric Kruskal-Wallis test was utilized to assess whether inlet and outlet pollutant concentrations are significantly different [38]. Statistical significance was set at a 95% confidence interval or $\alpha = 0.05$ [39]. If the median of any group was found to be statistically significant and different from the rest, the post-hoc Dunn-Bonferroni test was used to identify those groups [40]. It is worth mentioning that the statistical analyses in this study were performed in Microsoft Excel® with the add-in, XLSTAT 2014.

3. Results and Discussion

3.1. Hydraulic Performance

Figure 3a–c illustrate inlet and outlet flow rates for both *Cyperus* and *Pedilanthus* plants across three separate runs. Inlet flow rates were fixed at 9, 11, and 13 L/min for respective round of experiment. As it can be seen in Figure 3a–c, for both plant boxes, the outlet flow reached a stable level between 2–15 min after the beginning of the experiment. The stabled outlet flow rates for *Cyperus* and *Pedilanthus* plant boxes were 5.7 and 3.5 L/min, respectively. The detailed information on hydraulic performance of the two plant boxes including infiltration rate, overflown volume, and retained volume are presented in Table 2. It is worth mentioning that, the presented infiltration rate in Table 2, is the value recorded after outflow had become stable. As it can be seen, at this status, the infiltration rate in *Cyperus* was 22.6%, 27%, and 30% higher than the one in *Pedilanthus* plant box in experiment rounds 1, 2, and 3, respectively. This could be attributed to the extensive root growth of *Cyperus* compared to *Pedilanthus*. This was evident later, when the plants were pulled out at the end of experiments for further investigation on root growth and propagation patterns. It was found that *Pedilanthus* root did not penetrate below 250 mm depth of filter media, while *Cyperus* root reached the bottom of the planter box (at depth of 800 mm). In order to estimate the retention capacity of the system for the defined storm with 600L runoff volume, the overflown volume was also measured in each plant box for the 3 different experiment runs, where the inlet flow and infiltration rate were different (see Table 2). As it can be seen in Table 2, the overall retention performance of the two systems was varying between 81.3–100% for different inlet flow and infiltration rates in the three experiments. Overall, *Cyperus* planter box was found to have a better retention performance than *Pedilanthus* box due to its higher infiltration rate. However, both systems were able to meet the recommended infiltration rate by FAWB [17].

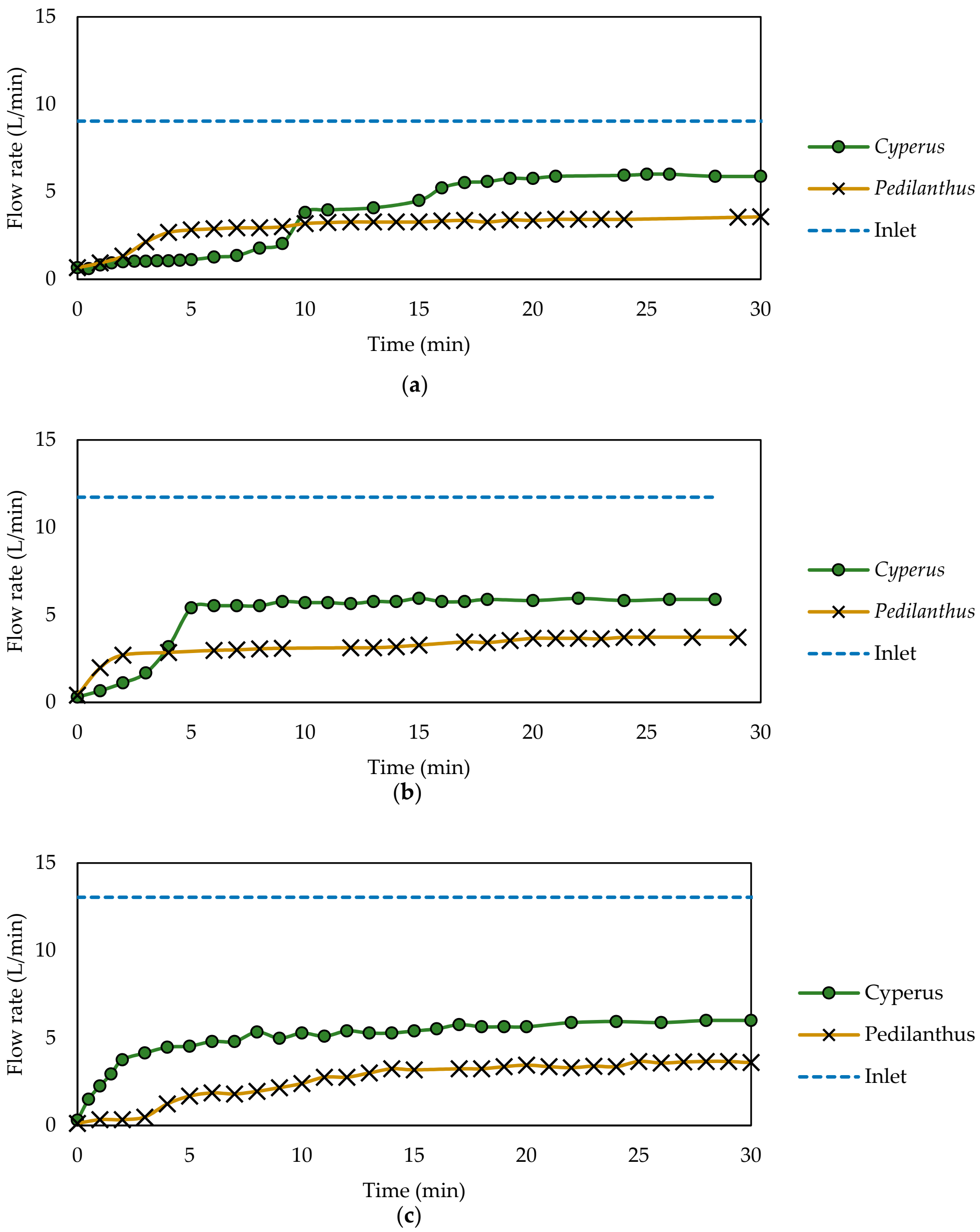


Figure 3. Inlet and outlet flow rates (L/min) for both *Cyperus* and *Pedilanthus* plants across: (**a**) Round 1, (**b**) round 2, and (**c**) round 3 of experiment.

Table 2. Hydraulic performance of box biofiltration system.

Round	Plant	Inlet Flow (L/min)	Overflown Volume (L)	Infiltration Rate (mm/h)	Stormwater Retention Performance (%)
1	*Cyperus*	9.0	0.0	289.4	100.0
	Pedilanthus	9.0	46.8	236.0	92.2
2	*Cyperus*	11.7	0.0	375.4	100.0
	Pedilanthus	11.8	59.2	295.6	90.1
3	*Cyperus*	12.7	28.4	350.2	95.3
	Pedilanthus	13.0	112.2	269.4	81.3

3.2. Pollutants Removal

3.2.1. Nutrients

Box plots of TN concentration in influent and effluent with respect to distance from the inlet and the depth from the surface are presented in Figure 4. In total, there were 12 outliers in the 144 data points, thus marking 132 data points within the range. It is noted that depth = 0 represents the surface water or inlet water (synthetic stormwater), while depth = 800 mm represents the main bottom outlet of the system. The descriptive statistics for TN after removing outliers are shown in Table 3. The overall *CRE* for TN in *Cyperus* and *Pedilanthus* at bottom outlet was 9.6% and −14.0%, respectively. It was concluded that both systems can hardly remove TN, although *Cyperus* performed marginally better than *Pedilanthus*. Low TN removal is also reported in some of the field studies on biofilters (−7% [41] to 32.2% [42]), while slightly better performance (up to 43.7%) has been recorded for soil column biofiltration studies [43]. Poor or negative TN removal performance may potentially be attributed to the leaching of dissolved organic nitrogen (DON) or nitrate-nitrogen (NO_3) from plant residue such as dead leaves and root tissue in the system [16,44]. In addition, the lack of submerged zone in the design may reduce the potential of denitrification process by bacteria [15,45]. Looking into the correlation between *CRE* and the depth of filter media (100, 250, and 400 mm), the *CRE* for *Cyperus* increased from −5.9% at 100 mm to 13.3% at 400 mm, while for *Pedilathus* it increased from −18.6% to 1.75% (see Table 3). Therefore, it was suggested that the TN removal in the deeper part of the filter media is better than the layers near the surface. This could be attributed to the contribution of plant roots to nutrients uptake, which is expected in the depth where roots are mainly propagated.

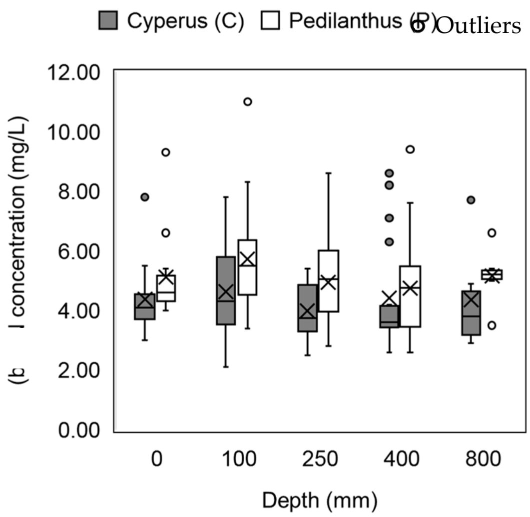


Figure 4. Box plots of TN concentration (mg/L) in water samples taken in different depths from the surface (concentration at depth = 0 represents the water quality on the top surface of filter media which is practically equal to the quality of synthetic stormwater at the inlet; concentration at depth = 800 mm represents the concentration at the main bottom outlet).

Further analyses were conducted to either prove or reject the hypothesis of correlation between TN removal and depth. Figure 5 illustrates the median TN concentrations versus depth after removing the outliers. As it can be seen, the lines of the best fit have nearly zero gradients, suggesting that TN was conserved inside the box biofilter. Furthermore, the grouped data failed the Shapiro-Wilk normality test as $p < 0.05$, thus considered as non-parametric. Further analysis through Kruskal-Wallis test indicated that there was no significant statistical difference between the inlet and outlet median TN concentrations for all groups as $p > 0.05$ (see Table 3). Therefore, TN concentration was independent from depth for *Cyperus* and *Pedilanthus* in the box biofilter, and this was likely because of little or no denitrification in the biofiltration system [46,47].

Table 3. Descriptive statistics for TN concentration at different depths with *p*-value from Kruskal-Wallis test.

	Depth (mm)	n	Min	Max	Mean	Std. dev.	*CRE* (%)
Cyperus ($p = 0.30$)	0 *	11	3.00	5.50	4.07	0.73	0.0%
	100	16	2.10	7.80	4.31	1.50	−5.9%
	250	18	2.50	5.40	3.99	0.92	2.0%
	400	14	2.60	4.20	3.53	0.40	13.3%
	800 **	5	2.90	4.90	3.68	0.81	9.6%
Pedilanthus ($p = 0.15$)	0 *	10	4.00	5.40	4.56	0.44	0.0%
	100	17	3.40	8.30	5.41	1.25	−18.6%
	250	17	2.80	6.30	4.72	1.22	−3.5%
	400	17	2.60	7.60	4.48	1.42	1.75%
	800 **	4	5.00	5.40	5.20	0.16	−14.0%

* Depth zero represents the soil surface and the corresponding TN concentration represents the inlet water quality.
** Depth of 800mm represents the bottom of the system and the corresponding TN concentration represents the bottom outlet water quality.

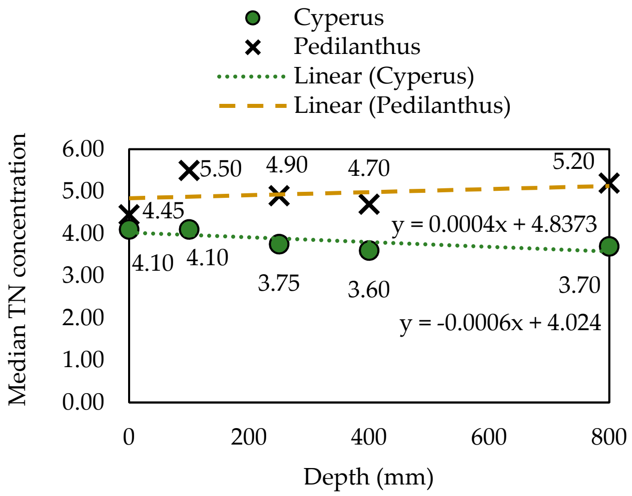


Figure 5. Median TN concentrations with respect to the depth after removing the outliers.

Figure 6 represents box plots of TP concentration in influent and effluent with respect to the depth. There are six outliers from 144 data points, thus leaving 138 data points for further discussion. The descriptive statistics for TP concentration (excluding the outliers) are summarized in Table 4. *Cyperus* and *Pedilanthus* achieved mean overall *CRE* of 67.3% and 62.5% in TP removal, respectively. As it can be seen, *Cyperus* plant box was marginally more effective in TP removal than *Pedilanthus* plant box. It is worth mentioning that TP removal was significantly better than TN removal due to the fact that soil media contributes to the TP adsorption, while it is not very effective in TN removal [48–50]. Since the performance of *Cyperus* and *Pedilanthus* in TP removal was not significantly different, it was concluded that the plant uptake should have not contribute much to the TP removal [51]. On the other hand, the TP removal showed a positive correlation with the depth of the filter media. Table 4 shows an increasing *CRE* for both *Cyperus* (42.9% to 51.9%) and *Pedilanthus* (27.5% to 50.0%) between 100–400 mm depth. This result satisfied the hypothesis that soil media play a bigger role in removing TP than the plant since the plants root was not developed at 100 mm depth of filter media layer. It was observed that the TP removal of the present study are matching with the ones reported in literature for field-scale biofiltration systems (e.g., 60% [52], 65% [9], and 17–92% [53]).

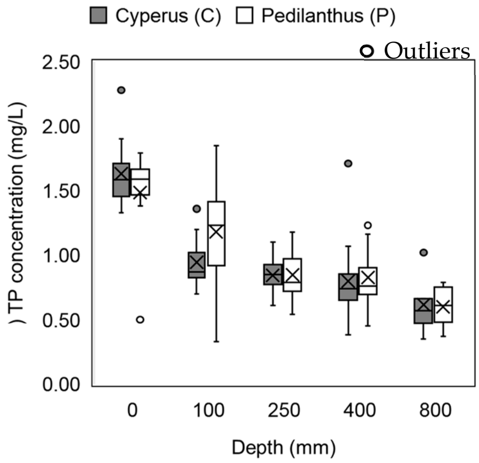


Figure 6. Box plots of TP concentration (mg/L) with respect to depth.

Table 4. Descriptive statistics for TP concentration at different depths with *p*-value from Kruskal-Wallis test.

	Depth (mm)	n	Min	Max	Mean	Std. dev.	*CRE* (%)
Cyperus ($p < 0.0001$)	0	9	1.33	1.90	1.56	0.19	0.0%
	100	13	0.70	1.20	0.89	0.13	42.9%
	250	15	0.61	1.10	0.84	0.14	46.2%
	400	13	0.38	1.07	0.75	0.17	51.9%
	800	4	0.35	0.66	0.51	0.13	67.3%
Pedilanthus ($p < 0.0001$)	0	9	1.38	1.79	1.60	0.14	0.0%
	100	14	0.33	1.85	1.16	0.38	27.5%
	250	15	0.54	1.18	0.85	0.19	46.9%
	400	14	0.45	1.16	0.80	0.19	50.0%
	800	5	0.37	0.79	0.60	0.18	62.5%

Looking into the normality of the data, the Shapiro-Wilk test failed as $p < 0.05$, thus non-parametric Kruskal-Wallis test was conducted. From Table 4, the Kruskal-Wallis test indicated that there was a significant difference between TP median concentrations ($p < 0.05$), thus post-hoc Dunn-Bonferroni test was conducted to clarify the group of each depth. Table 5 summarizes the *p*-values from the post-hoc Dunn-Bonferroni test for TP removal on each depth. Overall TP removal was statistically significant for both *Cyperus* and *Pedilanthus* as inlet TP median concentrations are significantly different from outlet TP median concentrations (Dunn-Bonferroni $p < 0.005$). Furthermore, Table 5 indicates that there was statistically significant difference (Dunn-Bonferroni $p < 0.005$) between inlet and each outlet, but not between one outlet and another. Between 0 and 100 mm depths, the test showed significant difference as *p*-values < 0.005. Comparing these *p*-values with the *p*-values between 100 mm and 250 mm outlets (see Table 5), no significant difference was observed as *p*-values > 0.005. Therefore, the statistical analysis test showed that the most significant TP removal occurs within the first 100 mm of depth from the surface. This is also evident by looking at the mean *CRE* values summarized in Table 4 for the aforementioned depths.

Figure 7 represents median TP concentrations as a function of depth using lines of best fit. Median TP concentrations decreased sharply (with a gradient of −0.0011 and −0.0010 for *Pedilanthus* and *Cyperus*, respectively) with an increase in depth. In comparison with the gradient produced from median TN concentration (see Figure 5), TP gradient was one order of magnitude higher. Thus, it was concluded that TP concentrations decrease with increasing depth more significantly than TN concentrations.

Table 5. TP-Depth *p*-values from Dunn-Bonferroni test after significant Kruskal-Wallis test. Bolded values are statistically significant at the corrected Bonferroni significance level of 0.005.

TP	Depth (mm)	0	100	250	400	800
Cyperus	0	1	**0.0025**	**0.0004**	**<0.0001**	**<0.0001**
	100	**0.0025**	1	0.6002	0.0776	0.0051
	250	**0.0004**	0.6002	1	0.1928	0.0127
	400	**<0.0001**	0.0776	0.1928	1	0.1121
	800	**<0.0001**	0.0051	0.0127	0.1121	1
Pedilanthus	0	1	0.0515	**<0.0001**	**<0.0001**	**<0.0001**
	100	0.0515	1	0.0205	0.0055	**0.0016**
	250	**<0.0001**	0.0205	1	0.6100	0.1302
	400	**<0.0001**	0.0055	0.6100	1	0.2559
	800	**<0.0001**	**0.0016**	0.1302	0.2559	1

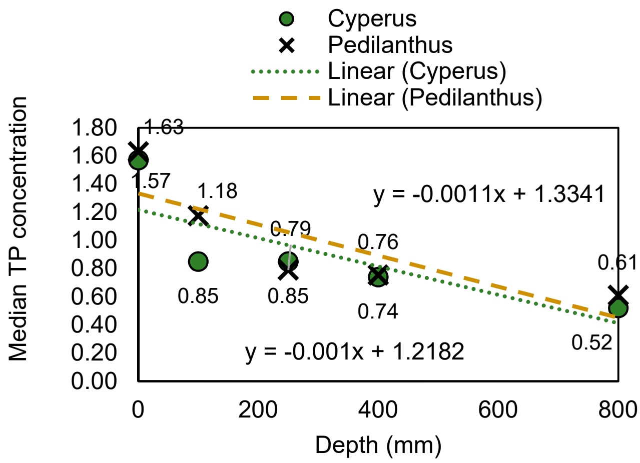


Figure 7. Median TP concentrations as a function of depth.

3.2.2. Heavy Metals

The overall mean *CRE* for both *Cyperus* and *Pedilanthus* plants are shown in Table 6. From the results obtained in this study, more than 90% removal was achieved for the targeted heavy metals (i.e., Cu, Mn, Ni, Pb, and Zn) except Fe. The high efficiency in heavy metal removal is attributed to the ion-exchange mechanism with soil media particles, whereas plants play almost no (or very limited) role in heavy metal uptake. This is consistent with several other studies on heavy metals removal in biofiltration systems [20,24,54–58]. *Pedilanthus* removed Fe by 76.5%, while the *Cyperus* removal rate of Fe was 50.6%. The overall removal rate of other heavy metals found to be similar between *Cyperus* and *Pedilanthus*. Since heavy metals removal mechanism are through ion-exchange with the soil particles (+ve charged metal ions with −ve charged soil), Fe as the only heavy metals present in the form of M(III) is more difficult to be removed by the soil media [20,59]. Higher retention time (lower infiltration rate) is generally suggested to improve the removal rate of Fe as it provides more contact between soil particles and metal ions. However, in tropical regions, low infiltration rate is not recommended due to the intense rainfall events.

Table 7 summarizes the descriptive statistics for one of the heavy metals studied (Cu) to represent the heavy metals tested in this study. Cu is chosen since its removal rate is almost similar to the ones for other metals of this study (except for Fe), while the number of outliers for Cu was also the closest to the average number of outliers found in other heavy metals. The data also failed Shapiro-Wilk normality test, and Kruskal-Wallis test indicated that there are significant differences between Cu median concentration and depth as $p < 0.05$. However, there was no significant difference between each sampling points. Therefore, most of the removal of Cu and other heavy metals happened at the top part of the soil layer, which followed the previous studies published in literature [54,60–63].

Table 6. Mean concentration removal efficiencies (*CRE*) for both *Cyperus* and *Pedilanthus* after removing outliers.

	Inlet Concentration (mg/L)		Outlet Concentration (mg/L)		Overall *CRE* (%)	
Pollutant	***Cyperus***	***Pedilanthus***	***Cyperus***	***Pedilanthus***	***Cyperus***	***Pedilanthus***
Cu	0.122	0.122	0.002	0.001	98.4%	99.5%
Fe	0.783	0.779	0.387	0.186	50.6%	76.1%
Mn	0.153	0.146	0.005	0.005	96.7%	96.6%
Ni	0.027	0.025	0.002	0.002	92.5%	91.9%
Pb	0.119	0.118	0.005	0.004	96.1%	96.6%
Zn	0.152	0.145	0.001	0.000	99.5%	100.0%

Table 7. Descriptive statistics for Cu concentration at different depths with *p*-value from Kruskal-Wallis test.

	Depth (mm)	n	Min	Max	Mean	Std. dev.	*CRE* (%)
Cyperus ($p < 0.0001$)	0	12	0.114	0.131	0.122	0.006	0.0
	100	14	0.002	0.011	0.006	0.002	95.1
	250	18	0.002	0.007	0.005	0.002	95.9
	400	18	0.001	0.007	0.004	0.002	96.7
	800	6	0.000	0.005	0.002	0.002	98.4
Pedilanthus ($p < 0.0001$)	0	12	0.114	0.138	0.122	0.007	0.0
	100	13	0.010	0.040	0.023	0.010	81.1
	250	18	0.002	0.014	0.007	0.004	94.3
	400	15	0.001	0.007	0.004	0.002	96.7
	800	6	0.000	0.001	0.001	0.001	99.2

4. Conclusions

Two lab-scale prototype biofiltration systems were developed using *Cyperus alternifolius* and *Pedilanthus tithymaloides* plants a layered sand-based filter media. The hydraulic performance as well as pollutants removal performance of these two prototypes were tested under tropical condition. The biofiltration system with *Cyperus alternifolius* had a higher infiltration rate and larger retention efficiency when compared to the one with *Pedilanthus tithymaloides*. This was attributed to the extensive root length growth of *Cyperus alternifolius* compared to the short root of *Pedilanthus tithymaloides*. It was concluded that roots contribute to the restoration of media's porosity by penetrating to the different layers of filter media. In general, both systems performed well in total phosphorus removal (67.3% removal from *Cyperus alternifolius* and 62.5% from *Pedilanthus tithymaloides*). Total phosphorus removal showed positive correlation with the depth of the filter media. This trend in phosphorus removal could be attributed to the adsorption mechanism in filter media; however, this cannot be confirmed by the limited results of this study. Moreover, according to the results, plant type was not found as a significant factor in phosphorus removal as both systems performed almost similarly. For total nitrogen, however, both systems failed to remove it well (9.6% removal in the system with *Cyperus alternifolius* and 14% leaching in the system with *Pedilanthus tithymaloides*). Low removal of total nitrogen was attributed to the lack of submerged zone as it can promote denitrification process. Additionally, there was no significant correlation between each sampling point for total nitrogen concentration; therefore, it was considered independent from the depth. For heavy metals of this study (i.e., Fe, Cu, Mn, Ni, Pb, and Zn) 90% removal rate was achieved in both systems except for Fe where the rate was lower (76%). Results confirmed that the top 100 mm depth of the filter media is the main contributor to the heavy metals removal. Overall, the biofiltration system with *Cyperus alternifolius* was found as a suitable choice in both hydraulic performance as well as pollutant removal under the tropical environment of the experiment.

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