



## Research article

## Implementation of a full-scale constructed wetland to treat greywater from tourism in Suluban Uluwatu Beach, Bali, Indonesia

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## ABSTRACT

This original research examines a full-scale subsurface Constructed Wetland (CW) system in Indonesia, where most CW research has been limited to laboratory scale experiments. The CW system was located in Bali and built in 2015 in a single series formation. This study aims to demonstrate the performance of the system in treating greywater and examine the nutrient content plants' above-ground biomass. The CW was arranged in linear sequence composed of one unplanted (CW1) and five planted treatments of *Iris pseudacorus* (CW2), *Caladium bicolor* (CW3), *Rhoe discolor* (CW4), *Sansevieria trifasciata* (CW5) and *Heliconia psittacorum* (CW6). There has been little research on *Caladium bicolor*, *Rhoe discolor* and *Sansevieria trifasciata* in a full-scale CW application. Our results showed fluctuating efficiency (%) in the reduction of Biological Oxygen Demand (BOD), Chemical Oxygen Demand (COD), Total Suspended Solid (TSS), Oil and Grease (O&G), Nitrate and Phosphate. The highest removal efficiency for CW1, CW2, CW3, CW4, CW5, CW6 were O&G (63.63%), BOD (90.66%), Nitrate (83.55%), BOD (80%), BOD (82.88%) and Phosphate (89.93%) respectively. After the experimental period, *S. trifasciata* and *H. psittacorum* experienced a decrease in Total N concentration, while *H. psittacorum* experienced a decrease in phosphate in above-ground biomass. Species of *R. discolor*, *C. bicolor* and *I. pseudacorus* showed good performance in terms of their growth and development. Although high removal efficiency was observed at certain times, this study showed the negative removal efficiencies at times among parameters as a consequence of the low Hydraulic Retention Time (HRT) and high Hydraulic Loading Rate (HLR).

## 1. Introduction

As one of the most famous and exotic locations in the world, Bali encounters rapid development of the tourism industry. The tourism industry uses 65% of local water consumption (Cole, 2012). It has been suggested that the disproportionate utilization of water between the needs for tourism and other sectors diminishes its environmental quality (Sunarta et al., 2014). Moreover, a severe water crisis predicted by 2025 if water management is not prioritized (Cole, 2012). Suluban Uluwatu Beach is one of the attractive coasts hidden by steep cliffs made of natural limestone formations. Regardless of its apparent beauty, environmental problems related to wastewater are already significant. Before 2015, wastewater produced by cafes was not treated properly since it was directly discharged to the ravine and infiltrated the limestone or flowed to the sea.

To overcome the problem, a local organization named Project Clean Uluwatu (PCU) collaborated with IDEP, an Indonesian foundation with

environmental concern, to build a series of horizontal subsurface flow constructed wetlands system (HSSFCW) in 2015. This system is known as a WWG (Wastewater Garden) by the local people. Constructed wetland (CW) denotes an effective and low-cost system which can reduce pollutants through physical, chemical and biological processes (Machado et al., 2017). As one of the types of CW, horizontal subsurface constructed wetlands, have been applied in a wide range of wastewater treatments, especially for domestic wastewater (Vymazal, 2009; Hua et al., 2017). Ornamental plants have also been used in some CWs due to their aesthetics and good removal efficiency (Konnerup et al., 2009; Calheiros et al., 2015; Méndez-Mendoza et al., 2015). Plants provide the need for degrading microbes such as carbon, and habitat for microbes (zones of roots, stems and leaves) that are submerged in wastewater (Brix, 2003).

CWs have been studied at a number of different scales. Higher removal efficiency has been shown in laboratory-scale CW compared to full-scale CW since plants often show better removal efficiency at a smaller scale and in controlled environments (Méndez-Mendoza et al.,

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2015). Full-scale CWs have the advantage of providing real world results in comparison to implementation at micro or meso scales (Brisson and Chazarenc, 2009) and several studies have demonstrated full-scale CWs can achieve more than 60% removal efficiency (Sim et al., 2008; Vys-tavna et al., 2017; Ali et al., 2018). However, full-scale CW experiments infrequently use replication due to its cost (Brisson and Chazarenc, 2009; Stefanakis et al., 2019).

Due to technical reasons, the HSSFCW in Suluban Uluwatu was not managed for a long time leading to the growth of unwanted wild plants in the system and decreased aesthetics. In many settings, aesthetics should be an important aspect of wastewater treatment (Calheiros et al., 2015). This study recreates HSSFCW at Suluban Uluwatu by planting five ornamental plants (*Iris pseudacorus*, *Caladium bicolor*, *Rhoeo discolor*, *Sansevieria trifasciata* and *Heliconia psittacorum*). *Iris pseudacorus* has been commonly used in many CW's studies (Wu et al., 2011; Barco and Borin, 2017; Burgos et al., 2017; Šereš et al., 2017) as has *H. psittacorum* (Konnerup et al., 2009; Méndez-Mendoza et al., 2015). Only one laboratory scale study was found for *Caladium bicolor* (Cuong and Loan, 2017). No studies were found for *Rhoeo discolor* or *Sansevieria trifasciata*.

This study examines the implementation of the HSSFCW series and does not compare individual planted treatments or the effect of their order because of the constraints of the existing construction. The study had three main objectives: (1) to evaluate the performance of the HSSFCW on wastewater produced by cafes and toilets in Suluban Uluwatu Beach; (2) to examine the initial (the time when plants were firstly acclimatized) and final (the last water sampling conducted) concentration of Total N and phosphate in above-ground biomass; and (3) to inspect the effect on the plants (growth and development).

## 2. Materials and methods

### 2.1. Duration of study and site description

The study was conducted from May to August 2018 at dry season. It was carried out at the entrance to Suluban Uluwatu Beach (8°48'57.5"S 115°05'20.1"E) at the altitude of 197 MASL. The reactor was built in a

single-series formation on a slope of around  $\pm 30^\circ$  (Figure 1). Day time temperature ranged from 27 to 46.6 °C. The air humidity ranged from 68 to 70% in the morning, 30–35% during the day and 66–69.2% in the evening. Light intensity in the morning and evening ranged from 2.920 to 12.000 lux, while midday was more than 20.000 lux.

### 2.2. System design and operational condition

Before reaching the HSSFCW system, greywater was collected in a storage tank with a capacity 12,990 L then pumped to the HSSFCW through a pipe with a length of  $\pm 150$  m (Figure 2). In the HSSFCW wastewater flowed through a series of treatments by gravity (Figure 1). In this study, existing materials were utilized such as the concrete tanks and gravels of  $\varnothing$  4–6 cm as media. Concrete tanks for each treatment had different dimensions (see Table 1) with a minimum of  $(2.4 \times 1.1 \times 0.6)$  m<sup>3</sup> to a maximum of  $(5.6 \times 1.1 \times 0.6)$  m<sup>3</sup>. The entire system consisted of 12 tanks from top to bottom, however, this study only used the first 6 tanks. The sequence of treatment was unplanted (CW1), *Iris pseudacorus* (CW2), *Caladium bicolor* (CW3), *Rhoeo discolor* (CW4), *Sansevieria trifasciata* (CW5) and lastly *Heliconia psittacorum* (CW6). All vegetation and litters were removed from the media, then the ornamentals were planted and allowed to acclimatize for two months before testing. The wastewater flowed for 2.5 h in a day with HRT for each treatment (tank) was 30 min and the discharge rate ranged from 0,242 L/s to 0,561 L/s.

### 2.3. Wastewater sampling and analyses

The wastewater parameters measured were BOD, COD, Nitrate, Phosphate, Oil and Grease, TSS (Total Suspended Solid), pH and temperature. Inlet characteristic can be seen in Table 2. Six samples were taken weekly from the outlet hole of each tanks, except for CW6 (*H.psittacorum*) which had five samples obtained weekly due to technical reason. BOD and COD were analyzed by titration, Nitrate was measured by Brucine, Phosphate by Ammonium Molybdate, Oil and Grease using gravimetry, and TSS using spectrophotometry with HACH DR/2010. pH



**Figure 1.** (a) The system outlook (b) CW1: unplanted (c) CW2: *I.pseudacorus* (d) CW3: *Caladium bicolor* (e) CW4: *Rhoeo discolor* (f) CW5: *Sansevieria trifasciata* (g) CW 6: *Heliconia psittacorum*.

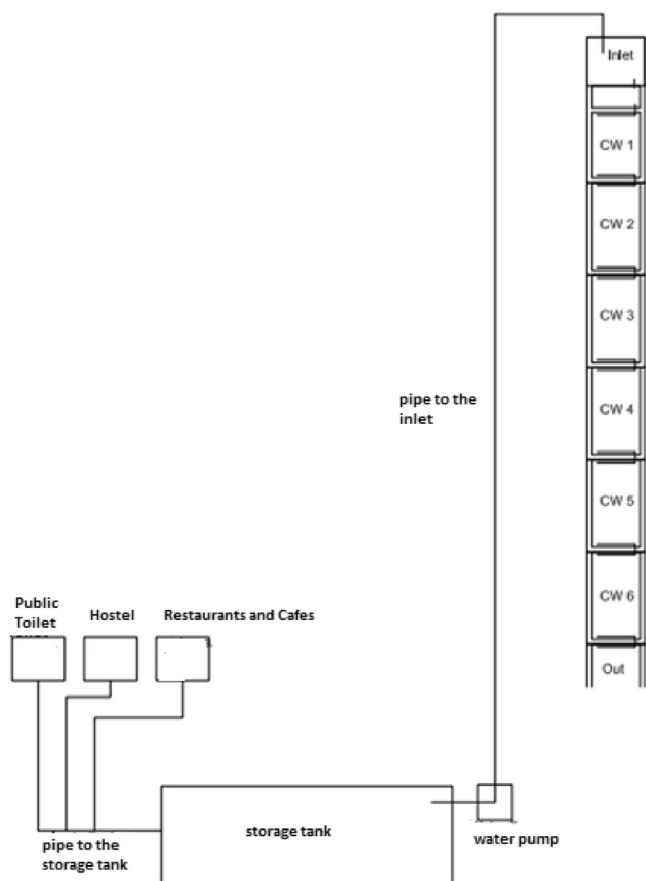


Figure 2. Schematic design of the system.

and temperature were measured directly on-site using a digital pH meter ATC and glassware thermometer.

#### 2.4. Plant nutrient sampling and analyses

Above ground biomass was obtained at the time of planting and after 14 weeks, by sampling leaves and stems picking from all the plants. Total N and phosphate in above-ground biomass were measured with the methods SNI-4721-1998 and Isric 6th 2002 respectively. For Total N, the sample was digested by 1 gr selenium mixture and 3,5 ml  $H_2SO_4$  for 4 h then distilled to a receiving flask containing 10 ml boric acid and Conway indicator. After distillation finished, it was titrated with  $H_2SO_4$ . Phosphate was measured by diluting sample with 10 ml of  $HNO_3$  in a vessel then heated inside a Microwave Digester at 200 °C for 15 min. Afterwards, the filtrate was added to a solution of concentration Ammonium hepta molybdate and  $SnCl_2$ . Phosphate concentration was measured with an absorption spectrophotometer (610 nm wave length).

#### 2.5. Data calculations

Treatment efficiency for each of the parameters was calculated as the removal percentage:

$$\text{Removal efficiency (\%)} = (\text{Cin} - \text{Cout}) / \text{Cin} \times 100 \quad (1)$$

where, Cin (mg/l) is the inlet concentration and Cout (mg/l) is the outlet concentration.

Difference (%) of concentration for nutrient contained in above-ground biomass:

$$\text{Difference (\%)} = (\text{Cin} - \text{Cfin}) / \text{Cin} \times 100 \quad (2)$$

where, Cin (mg/kg) is the initial concentration of nutrients and Cfin (mg/kg) is the final concentration of nutrients. The removal efficiency of all the treatments can be seen in Table 3.

#### 2.6. Statistical analyses

Plant's growth and development was analyzed with SPSS (IBM SPSS Statistics 22) before and after greywater treatment in order to examine the plants' capacity to deal with greywater. Data normality was checked by the Shapiro-Wilk test. As the data had normal distribution, the Paired-Samples T-test was used to identify significant differences ( $p < 0.05$ ) in plant height, shortest leaf length, and number of shoots.

### 3. Results and discussion

#### 3.1. Hydraulic Loading Rate (HLR)

Table 4 presents the value of HLR for every treatment. The highest value of HLR was represented by CW1 ( $1.53 \text{ m}^3 \text{ day}^{-1} \text{ m}^{-2}$ ) while the lowest one was represented by CW6 ( $0.65 \text{ m}^3 \text{ day}^{-1} \text{ m}^{-2}$ ). The HLR tends to be in descending order from CW1 to CW6. The plummeted efficiency could also be triggered by the high value of HLR. A study by Çakir et al. (2015) testified that there was decreasing removal rate in line with the increasing of HLR namely 80.4, 74.5 and 62.1% were attained as a result of 0.050; 0.075 and  $0.125 \text{ m}^3 \text{ day}^{-1} \text{ m}^{-2}$  HLR, respectively. While a research conducted by Abou-Elela et al. (2017) revealed that a decreasing HLR from 0.18 to  $0.10 \text{ m}^3 \text{ day}^{-1} \text{ m}^{-2}$  improved the removal percentage on municipal wastewater in pilot scale horizontal subsurface CW.

#### 3.2. Reduction in BOD (Biological Oxygen Demand)

Reduction in BOD by CW1 reached a maximum of 54.4% and a minimum of -52.8% (calculated using Eq. (1)). The negative value appeared only in the first sampling. In CW2, negative values were observed for the first four weeks of sampling, before it reached the maximum value (90.66%) in the last week of sampling. This value is the highest reduction efficiency among all parameters measured from treatment CW2 (Figure 3). BOD in CW3 showed the highest reduction of 65.7% (week 6) and had negative values in the second and third week of sampling. CW4 had the highest value of 80% (week 2) but had negative values in the first and last week of sampling. Reduction of BOD had positive values from the first to fourth of sampling in CW5. The highest reduction of BOD was 82.88% for CW5. CW6 experienced only one negative value (-83.05%) in the fourth week.

In the system, the removal of polluting organic matter is degraded aerobically and anaerobically by microorganisms (Vasudevan et al., 2011). In addition to biochemical reactions, a decrease in BOD can also occur through a physical process, namely through the precipitation and attachment of particulates to gravels (Abou-Elela, 2017). However, the negative removal efficiency can occur due to decay of plant organs inside the substrate (Šereš et al., 2017) and HRT did not allow for adequate contact time and establishment of anaerobic conditions in the beds.

#### 3.3. Reduction in COD (Chemical Oxygen Demand)

CW1 showed only one positive value in the fourth week of sampling (17.86%). CW2 reached 82% of reduction efficiency in the last week of sampling and had three negative values in the first three weeks. CW3 revealed only negative value in the third week and 44.4% was the highest reduction efficiency observed in week 6. CW4 showed negative values until the last week of sampling (-36.75%) with 27.78% of the highest reduction. CW5 reached the highest removal (76.47%) in the fifth week of sampling and had -4.54 and -160 % as two negative values in the third and fourth week. CW6 reached 80.77% of reduction in the third week of sampling with two negative values at the first two weeks.



**Table 1.** Specific dimension of six tanks.

Tank	Length (m)	Width (m)	Surface Area (m <sup>2</sup> )	Depth (m)
CW1	2.4	1.1	2.64	0.6
CW2	2.4	1.1	2.64	0.6
CW3	3	1.1	3.3	0.6
CW4	4	1.1	4.4	0.6
CW5	4.4	1.1	4.84	0.6
CW6	5.6	1.1	6.16	0.6

**Table 2.** Inlet characteristic.

Water Quality Parameter	Average $\pm$ SD	Max	Min
TSS	280.17 $\pm$ 83.71	433	153
Nitrate	1.49 $\pm$ 0.52	2.195	0.639
BOD	197.67 $\pm$ 94.98	391.5	100.4
COD	285.33 $\pm$ 90.14	423.12	199.2
Phosphate	1.76 $\pm$ 1.54	4.9	0.19
Oil and Grease	0.177 $\pm$ 0.085	0.32	0.08
pH	6.6 $\pm$ 0.28	7.2	6.3
Temperature	26.5 $\pm$ 0.35	27	26.1

Decrease in COD can occur through degradation carried out by heterotrophic microorganisms both anaerobically and aerobically (Priya et al., 2013) and also through physical processes such as sedimentation, filtration (Merino-Solis et al., 2015; Sudarsan et al., 2017) and deposition (Tilak et al., 2017). The physical processes are more important in reducing COD than biological processes (Sudarsan et al., 2017).

Increase in COD can be caused by an increase of organic load due to decay of plant organs that are still left in the substrate. It can also be caused by the existence of a non-biodegradable substances which is shown by the BOD/COD ratio value. Values below 0.3 indicate that the organic load in wastewater is non-biodegradable (Abou-Elela, 2017). The wastewater source from cafes/restaurants and potential cleaning chemicals is also presumably contributed the low value of BOD/COD ratio. This study found some data with BOD/COD ratios below 0.3, specifically 0.24 (CW3: 6<sup>th</sup> week of sampling); 0.22 (CW4: 6<sup>th</sup> week of sampling); 0.1 and 0.15 (CW5: 3<sup>rd</sup> and 5<sup>th</sup> week of sampling, respectively); 0.075, 0.08, and 0.29 (CW6: 1<sup>st</sup>, 2<sup>nd</sup> and 3<sup>rd</sup> week of sampling, respectively).

The non-biodegradable components are suspected to be lignin and hemicellulose derived from the wild plant organs left in substrate before the study was carried out. Lignin is one of the constituent components of plants along with cellulose and other fiber ingredients that make up plant cells. Lignin and hemicellulose are organic materials that are not easily degraded by microbes. The solubility of natural lignin in water is less at low pH (Zhu and Theliander (2011). Other factors which can increase COD include the presence of glucose, fructose, ethanol and acetic acid (Welz et al., 2011). However, those substances were not measured in this study.

### 3.4. Removal of nitrate

Removal of nitrate in CW1 showed negative values in weeks 1,2,3, 6 of sampling with only 13.23% of the highest reduction (the removal was calculated by Eq. (1)). CW2 experienced -323% removal efficiency only in the last week but had 50.63% as the highest removal at the fifth week. CW3 reached 83.55% removal in the sixth weeks and had negative value only in the fifth week of sampling. CW4 reached 28.82% as the highest value of removal in at the second week. A negative value also occurred in CW5 (-96.59%) for the last week of sampling. CW6 had 78.1% of removal at the fifth week of sampling.

In many studies, denitrification is considered a major mechanism for nitrate reduction in the CW system (Matheson and Sukias, 2010; Saeed and Sun, 2012; Merino-Solis et al., 2015). Some studies have shown that plants contribute to the reduction of nitrates not only through direct uptake, but also as a source of organic carbon which can trigger denitrification (Lin et al., 2002; Almeida et al., 2017). However, some studies suggest that plants only have a small contribution to reduce nitrate in CW and their significance is still debated (Shelef et al., 2013; Li et al., 2013). Matheson and Sukias (2010) show that plant uptake contributes up to about 9% of the total nitrate reduction in the system, while 71% of nitrate reduction is dominated by denitrification. Plant uptake is just another mechanism for reducing nitrate but when the plant has withered and dies, the nutrients contained in the tissue can return to the system. This is in accordance with the observation of this study that nitrate concentrations at the outlets varied significantly and were not uniformly reduced by the presence of plants (Figure 3e). The uptake of nitrate can be inhibited by various factors, including the presence of competing chemicals. Nitrite can inhibit nitrate uptake because it has the same transporter and binding site. Ammonium is also considered to be an inhibitor by disrupting the affinity of the transport system (Bose dan Srivastava, 2001).

A second possible cause of reduced removal efficiency in nitrate by CW systems is low activity of denitrification or nitrate ammonification. If the aerobic degradation process has high activity, the activity of anaerobic degradation processes such as denitrification will be reduced (Hua et al., 2017). This statement is reinforced by Stein and Hook (2005) who showed that high nitrification activity and COD, which indicates high aerobic activity connected with low anaerobic degradation.

### 3.5. Removal of phosphate

Phosphate removal for CW1 showed negative values in the first four weeks of sampling. The maximum value was 18.79% in the last week. CW2 reached the highest removal efficiency of 90.62%, while CW3 showed a negative value only in the first week and reached removal of 49.27%. CW4 still experienced a negative value in the last week of sampling and reached 71.87% removal in the second week. CW5 showed negative value only in third week (-16.17%), while CW6 had negative

Table 3. Removal effectiveness (%) of all treatments.

Water Quality Parameter	CW 1			CW 2			CW 3			CW 4			CW 5			CW 6		
	Average ±SD	Max	Min	Average ±SD	Max	Min	Average ±SD	Max	Min	Average ±SD	Max	Min	Average ±SD	Max	Min	Average ±SD	Max	Min
Nitrate	-49.4 ± 106.9	13.23	-265	-35.19 ± 142.7	50.6	-323	22.52 ± 40.2	83.6	-42.7	-15.65 ± 66.4	28.8	-143.7	-343.8 ± 384.8	46.63	-859	13.1180 ± 61.5	78.1	-56.54
Phosphate	-120 ± 236.8	18.79	-591	22.26 ± 49	90.6	-26.8	21.55 ± 27.6	49.3	-23.9	-85.28 ± 154.7	34.4	-307.7	30.9817 ± 30.6	62.10	-16.2	6.9540 ± 77.54	89.9	-93.16
TSS	-39.45 ± 30.7	-39.5	33	17.27 ± 38.4	86.9	-31.5	26.9 ± 38.1	63.6	-26	15.9 ± 61.27	63.8	-100.5	36.0917 ± 37.2	68.18	-11.5	-11.4440 ± 68.14	74.3	-114.8
Oil and Grease	29.7 ± 35.17	63.63	-33.3	-12.5 ± 77.77	83.3	-150	-160 ± 367.5	64.3	-900	21.9 ± 51.6	72.2	-60	4.8600 ± 56.5	62.50	-100	-3.3320 ± 60.55	66.7	-100
BOD	54.44 ± 39.01	54.44	-52.8	-16.7 ± 75.7	90.7	-132	8.8 ± 32.7	65.7	-25	6.58 ± 74.14	80	-124.7	38.6083 ± 46.2	82.88	-20.6	16.0140 ± 57.47	62.6	-83.05
COD	-5.59 ± 13.7	17.86	-19.5	7.2 ± 49	82	-59	19.29 ± 23.5	44.4	-19.1	-13.6 ± 6.11	71.9	-114.6	10.1033 ± 88.48	76.47	-160	-64.7160 ± 194.03	80.8	-400

Table 4. Value of HLR for each tank.

Tank	HLR (m <sup>3</sup> day <sup>-1</sup> m <sup>-2</sup> )
CW1	1.53
CW2	1.53
CW3	1.22
CW4	0.92
CW5	0.83
CW6	0.65

values for weeks 4,5 and 6 with 89.93% as the highest removal at the second week of sampling.

Phosphate can be reduced by physical-chemical reactions related to adsorption, precipitation (Burgos et al., 2017) and ion exchange (Mojiri et al., 2017) on CW media. Phosphate reduction can also be caused by uptake by plants (Vera et al., 2014; Haritash et al., 2015) but the plant's uptake capacity is considered limited (Vymazal, 2004).

Increases in dissolved of phosphate may have occurred due to changes in chemistry causing dissociation. If the media is anaerobic, Fe (III) will be reduced to Fe (II) which will result in a weak bond with phosphate (Verhoeven and Meuleman, 1999). As a result, phosphate concentration will increase in the outlet. Increase of phosphate value could also be caused by the release from dead plant organs in the substrate present before the study was carried out. Dead plant material is thought to be a reason since the system was densely populated with wild plants prior to this experiment. Nutrients can return to the system due to wilting plants and can affect the effectiveness of CW reduction (Menon and Holland, 2014). Low phosphorus removal could be also attributed to the fact that the units were not new, hence the gravel media was probably already saturated from previous operation, providing limiting adsorption sites (Tatoulis et al., 2017).

### 3.6. Removal of TSS (Total Suspended Solid)

Removal efficiency of TSS in CW1 showed positive values in weeks 2,3,4 and 5 with 33% as the highest value. CW2 showed a negative value of removal only in the third week with 86.85% as the highest removal for the last week. CW3 reached 63.6% of removal as the highest value of removal in the second week with two negative values at the first and fourth week. CW4 had 63.79% as the highest value for removing TSS with only one negative value at the first week. CW5 and CW6 had 68.18% and 74.34% respectively as the highest removal. Both CW5 and CW6 had negative value for the last week of sampling.

Removal efficiency of TSS was generally low (Figure 3c). Removal efficiency of TSS. The low removal efficiency of TSS could be caused by low HRT (Hydraulic Retention Time). Low HRT causes short duration of contact of wastewater with the substrate, so that the physical process does not occur optimally. Panrare et al. (2015) stated that HRT extension would provide a better reduction in TSS parameters, even though it was applied to non-plant treatments. Merino-Solis et al. (2015) revealed that HRT that is good maximize effectiveness of HSSFCW varies from 3 to 15 days depending on the type of pollutant in the waste.

### 3.7. Removal of Oil and Grease (O&G)

CW1 removed 63.63% O&G in the fourth week as the highest removal value. CW2 reached 83.33% as its highest removal in the last week of sampling. CW3 showed -900% as the lowest removal efficiency in the last week and had 64.28% as the highest value of removal at the third week. CW4 showed 72.22% as its highest value of removal in the second week, while CW5 showed 33.33% of removal both in the first and the last week. However, CW6 still showed negative value until the last week of sampling (-100%).

The removal of Oil and Grease (O&G) can be conducted by plants via translocation and accumulation in the plant's body. A study conducted by Mustapha et al. (2018) provided evidence that *T. latifolia* had the capability to accumulate O&Gs in its tissue and decrease 70–80% of O&G in the system. Plants' root can also physically trap O&G (Winanti et al., 2018).

The drop of O&G efficiency at CW4 and CW5 in the present study could be related to tank leakage. Leakage can lead to greater O&G retention on the surface of the substrate. After leaks in the system were repaired at CW4 and CW5, a higher level of O&G was measured at the outlet. It is suggested that when liquid waste could flow constantly, the remnants of oil and fat left in the system then rose and experienced a wash-out, increasing the concentration of oil and fat at the outlet.

### 3.8. Nutrients in above-ground biomass

Nutrients which are measured in above-ground biomass was Total N and Phosphate concentration (mg/g). It represents the initial and final concentration (before and after the wastewater flowed to the plants). The difference of the concentrations was determined using Eq. (2). *I. pseudacorus* showed an increase 71.6% of Total N and 27.5% of Phosphate. *C. bicolor* had a slight increase of Total N (15.7%), but there was no any phosphate detected in the biomass. *R. discolor* also showed a slight increase of Total N (4.5%) but it had more increase in Phosphate (20.9%). *S. trifasciata* experienced a decrease of Total N (-24.8%) and even a slight increase of Phosphate (5.9%). *H. psittacorum* showed a decrease of Total N and Phosphate namely -33.8% and -45.4% respectively. Further detail of concentration can be seen in Table 5.

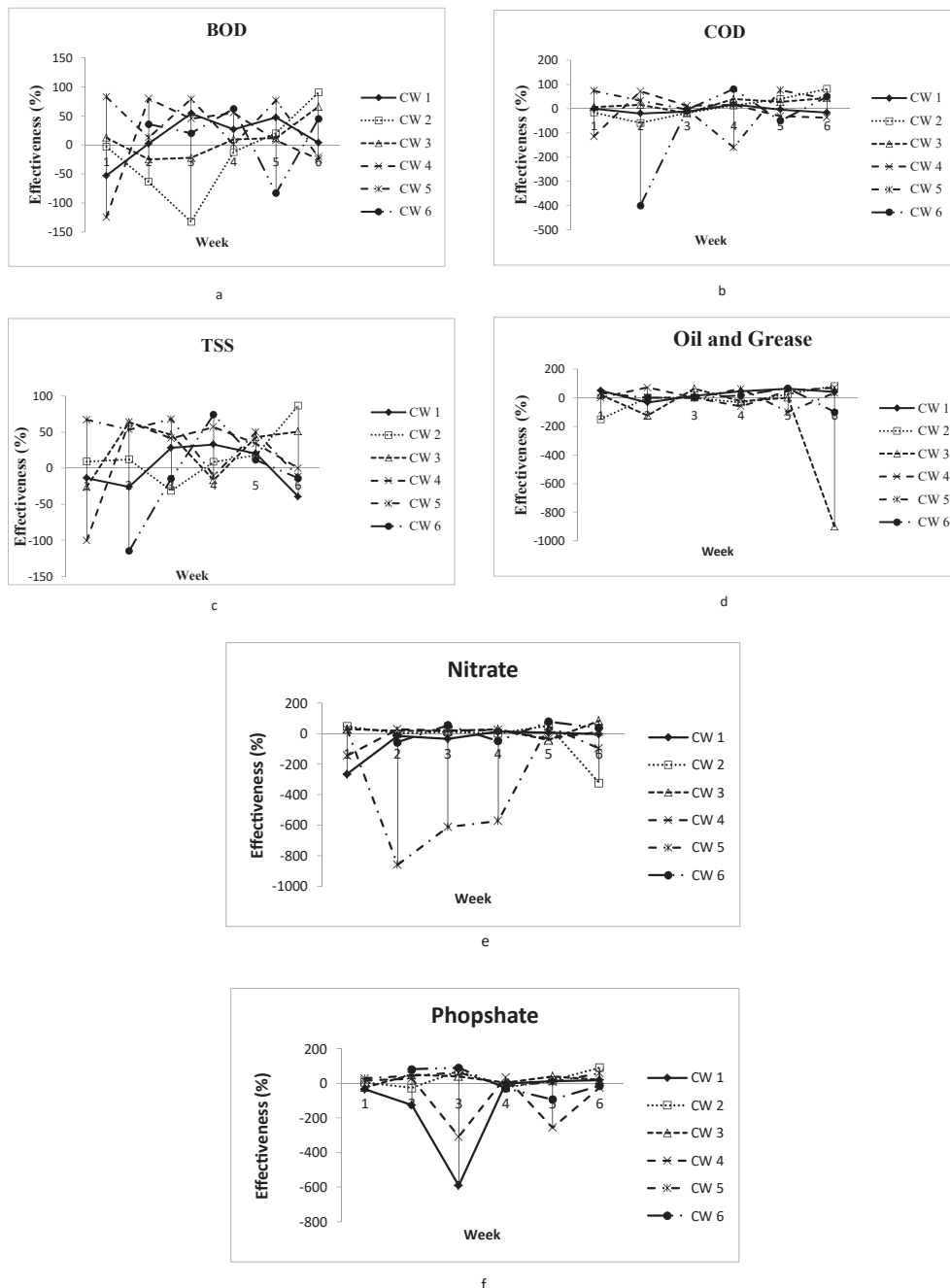


Figure 3. Removal Efficiency in each of treatments for (a) BOD (b) COD (c) TSS (d) Oil and Grease (e) Nitrate (f) Phosphate.

**Table 5.** Concentration of Total N and Phosphate before and after system worked in plants' above-ground biomass.

Treatment	Total N (mg/kg)		Phosphate (mg/kg)	
	Initial	Final	Initial	Final
CW 2	1200.454	2059.986	115.35	147.09
CW 3	3307.562	3828.63	247.87	Not detected
CW 4	2686.7	2808.637	206.7	249.99
CW 5	2069.344	1556.133	123.58	130.96
CW 6	2473.785	1635.287	196.03	106.75

All plants have a higher tendency to uptake N (Figure 4) than Phosphate (Figure 5). Minimum N uptake can reach 10 times higher than phosphate. Barco and Borin (2017) in their study stated that plants tend to store a smaller amount of P compounds than N compounds in their roots, especially *I.pseudacorus* plants. Our research supports this observation, demonstrating that above-ground biomass, N levels are up to 10 times higher than Phosphate (Table 5). Total N was higher than Phosphate in all test species. Wu et al. (2011) also revealed that *I.pseudacorus* has an N uptake capacity greater than P because P was bound tighter by the substrate. In this study, the increase in total N uptake in *I. pseudacorus* before and after the system was 71.6% while the phosphate uptake was 27.5%.

A decrease in total N concentration in above-ground biomass was found in *S. trifasciata* (CW5) and *H.psittacorum* (CW6) (Figure 4) while a decrease in phosphate was found in *H. psittacorum* (CW6) and possibly *C.bicolor* (CW3) (Figure 5). This could be caused by a number of factors. Given that the reduction of N and Phosphate was observed in the final tanks of the system, it is possible that there was a reduction in available N and Phosphate causing deficiency. Because of the previously mentioned reasons for temporal fluctuations in greywater concentrations of Nitrate and Phosphate, it is suggested that the point measurements of Nitrate and Phosphate may not indicate the average concentrations over time. In contrast the total N and Phosphate of plants indicates the accumulation or loss during the experimental period. Another reason for variation could be related to interspecies variation in transpiration.

Sudarsan et al. (2017) pointed out that the uptake of nitrate and phosphate carried out by leaves is the smallest among other parts of the plant because of the process of stomata transpiration in the leaves. Another possibility is that N and P have been used by metabolic processes in the plant. This assumption is supported by Collins et al. (2006) which revealed that substances stored in the body of the plant can be metabolized resulting the reduction of nutrient concentration in plant tissues.

### 3.9. Plant growth and development

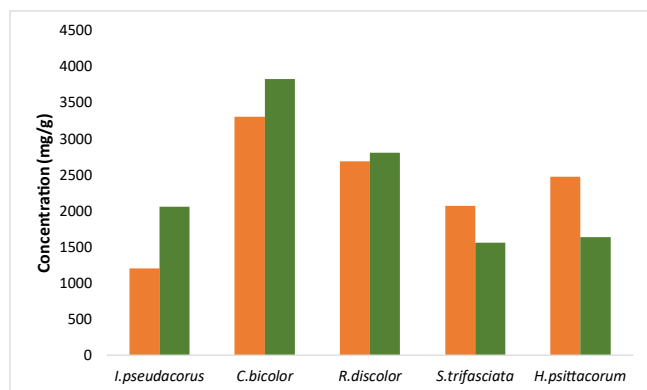
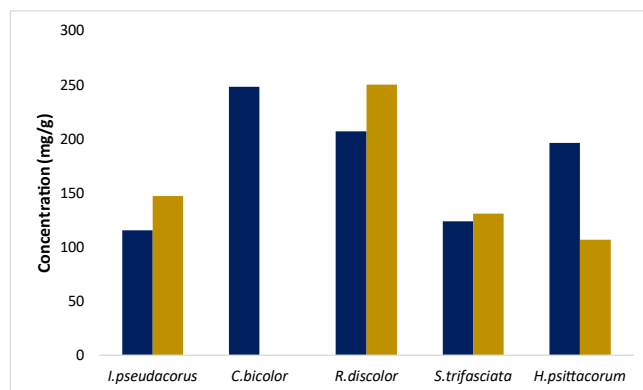
During the study, *I.pseudacorus* showed significant growth (plant height, paired T-Test,  $p < 0.05$ ) and also produced flowers). *I.pseudacorus*

had showed good performance in dealing with greywater in a previous study (Perdana et al., 2018). Similar results were also found for *Caladium bicolor* and *Rhoe discolor* which encountered good growth but no flowering occurred. In the first and second weeks of acclimatization, both of these plants seemed withered. The *R. discolor* showed its wilting marked by brownish leaves on the underside. However, when entering the third week of acclimatization until the end of the sampling, both showed remarkable growth and development. *C.bicolor* and *R.discolor* produced a lot of shoots and clumps. The growth of the leaves reaches almost twice of the initial length. Both plants experienced significant growth ( $p < 0.05$ ).

Unsatisfactory growth and development were found in *S. trifasciata* and *H.psittacorum*. A number of leaves on each plant of *S.trifasciata* were withered. It is suspected that this plant could survive in the CW system but may not reproduce. In another study, Weedon (2016) also found poor development presented by *P.australis* in CW system, showing that certain plants cannot always adapt successfully. However, in this study some of the difficulties in growth experienced by *S.trifasciata* (CW5) and *H.psittacorum* (CW6) may be attributed to the technical problem (leakage) at CW5 at the beginning of acclimatization. This could have caused both reduced water and nutrient availability to the plants in CW5 and CW6. In addition, the T-test showed that there was no significant difference in terms of their growth and development before and after the experimental period. Some of the roots of the wild plants that had grown before the study were suspected to have decayed in the substrate and lead to the increase of organic loads.

## 4. Conclusion

This study denotes fluctuating removal efficiency among parameters measured and the negative removal efficiency was promoted mainly due to lack of HRT and high value of HLR. As less known ornamental plants, *C.bicolor* and *R.discolor* are recommended to be applied in CW for their endurance. Both of them are also promising to be implemented toward other types of wastewater for future research.

**Figure 4.** Total N concentration in above-ground biomass.**Figure 5.** Phosphate concentration in above-ground biomass.

## Declarations

### Author contribution statement

M.C. Perdana: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.

S. Hadisusanto: Conceived and designed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data.

I. L. Setyawan Purnama: Conceived and designed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data.

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### Competing interest statement

The authors declare no conflict of interest.

### Additional information

Data associated with this study has been deposited at Mendeley (Title: Inlet and outlet measurement from WWG in Suluban) under the <https://doi.org/10.17632/btj35rr277.1>.

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