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DOI

[10.3390/w17142122](https://doi.org/10.3390/w17142122)

Publication date

2025

Document Version

Final published version

Published in

Water

Citation (APA)

Yen, L. Y., Abdullah, S. R. S., Imron, M. F., & Kurniawan, S. B. (2025). Natural-Based Solution for Sewage Using Hydroponic Systems with Water Hyacinth. *Water*, 17(14), Article 2122.
<https://doi.org/10.3390/w17142122>

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Article

Natural-Based Solution for Sewage Using Hydroponic Systems with Water Hyacinth

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Abstract

Domestic wastewater discharge is the major source of pollution in Malaysia. Phytoremediation under hydroponic conditions was initiated to treat domestic wastewater and, at the same time, to resolve the space limitation issue by installing a hydroponic system in vertical space at the site. Water hyacinth (WH) was selected in this study to identify its performance of water hyacinth in removing nutrients in raw sewage under batch operation. In the batch experiment, the ratio of $COD_{initial}/plant_{initial}$ was identified, and SPSS ANOVA analysis shows that the number of plant size factors was not statistically different in this study. Therefore, four WH, each with an initial weight of 60 ± 20 g, were recommended for this study. Throughout the 10 days of the batch experiment, the average of COD, BOD, TSS, TP, NH_4 , and color removal was 73%, 73%, 86%, 79%, 77%, and 54%, respectively. The WH biomass weight increased by an average of 78%. The plants have also improved the DO level from 0.24 mg/L to 4.88 mg/L. However, the pH of effluent decreased from pH 7.05 to pH 4.88 below the sewage Standard B discharge limit of pH 9–pH 5.50. Four WH plant groups were recommended for future study, as the COD removal among the other plant groups is not a statistically significant difference ($p < 0.05$). Furthermore, the lower plant biomass is preferable for the high pollutant removal performance due to the fact that it can reduce the maintenance and operating costs.

Keywords: domestic wastewater; environmental pollution; phytoremediation; hydroponic; wastewater treatment



Academic Editor: Christos S. Akkratos

Received: 5 June 2025

Revised: 11 July 2025

Accepted: 15 July 2025

Published: 16 July 2025

Citation: Yen, L.Y.; Abdullah, S.R.S.; Imron, M.F.; Kurniawan, S.B.

Natural-Based Solution for Sewage Using Hydroponic Systems with Water Hyacinth. *Water* **2025**, *17*, 2122. <https://doi.org/10.3390/w17142122>

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1. Introduction

Water is one of the important resources on Earth. All living things depend on water to survive. The rapid growth of urbanization and industrialization releases high volumes of wastewater, indirectly increasing the water pollution rate [1]. Water pollution is defined as water condition where the water quality has changed either chemically, physically, or biologically. It is a global issue, and the widespread problems with water pollution are jeopardizing human, plant, and animal health [2].

Sewage is referred to as domestic wastewater, which is generated from household activities. Table 1 shows the characteristics of the sewage effluent from several studies.

The highest contamination from municipal wastewater is organic matter, which will cause organic pollution.

Table 1. Raw sewage characteristics and the study site are compared with the EQA (Sewage) discharge standard.

Parameter	[3]	[4]	[5]	[6]	[7]	EQA (Sewage) Regulations 2009 Malaysia [8]	
						Standard A	Standard B
pH	7.19	8.77	7.38	7.8	7	6.0–9.0	5.5–9.0
TSS (mg/L)	130	780	164.6	88.9	-	50	100
BOD (mg/L)	69.22	80	215.42	-	-	20	50
COD (mg/L)	231.8	520	418.36	278	260	120	200
Nitrates (mg/L)	-	3.84	-	1.4	-	20	50
Phosphate (mg/L)	38.12	1.189	11.43	3.1	-	5	10
TN (mg/L)	-	-	47.15	3.1	-	-	-
Ammoniacal Nitrogen (mg/L)	-	-	33.01	41.6	-	20	50

When organic matter increases in a water body, the number of decomposers will increase to break down the organic matter. The rapid growth of microorganisms will lead to a depletion of dissolved oxygen in the water bodies as the decomposition process occurs. This phenomenon will limit the oxygen availability, endangering aquatic life, generating foul-smelling water, and turning the water black, which is highly toxic for aquatic life. On the other hand, sewage also contains inorganic materials such as phosphates and nitrates, which come from soaps and detergents. The high content of nitrogen and phosphorus in sewage wastewater is the main contributor to triggering water eutrophication. Eutrophication leads to an overgrowth of plants and algae [9].

The dissolved organic compound is commonly removed by employing biological treatment systems [8]. The conventional methods are typically costly, non-eco-friendly [10], and require a large piece of land to build on. The limitation of empty space or land is always an infrastructure issue for the industry. Furthermore, the expansion of production areas in a factory and the increased employee population have resulted in greater quantities of municipal wastewater being generated. Owing to these limitations, innovative and green technology shall be introduced for sewage treatment.

Phytoremediation is a solar-driven biological treatment performed directly in situ [11]. It is an eco-friendly technology and a low-cost method using plants to clean up soil, air, and water contamination. Phytoremediation is commonly used as a tertiary treatment system or polishing system due to the plant's limitations with high concentrations of pollutants. Moreover, the phytoremediation process is slow, and the hydraulic retention time or treatment period used to be longer and required a large area, like a constructed wetland or pond. In the phytoremediation process, there are many aquatic plants, such as water hyacinth, water lettuce, Kariba weed, water chestnut, and vetiver grass commonly used for the phytoremediation process. According to the Iamchaturapatr [12], water hyacinth, water lettuce, and water chestnut obtained the highest nutrient removal rates according to plant weight calculation. However, phytoremediation is not popular in Malaysia. Furthermore, invasive plants like water hyacinth and water lettuce have also been reported as the most problematic aquatic plants, as they have the fastest growth rate and can bloom in water bodies and block rivers in a short time to causing the river to lose evapotranspiration [13].

Hydroponics is the system for plant cultivation that uses nutrient solutions in water without soil [14]. According to Souza et al. [14], hydroponics is an advanced approach for crop production that is not only innovative, eco-friendly, reliable, and flexible, optimizing

resources and production output, but is also a promising method for protecting soil with minimal use of soil. Rana et al. [15] highlighted that the hydroponic principle can also be employed in nutrient reclamation from domestic wastewater for crop production in a controlled environment. In a hydroponic system, phytoaccumulation (plants extracting pollutants) [16] and rhizofiltration (filtration by the plant's root system) [17] are reported to be the main pollutant-removal mechanisms, while phytodegradation (enzymatic degradation by plants) [18] and rhizodegradation (enzymatic degradation by the plant root's microbial communities) [19] were also mentioned.

Owing to the space limitation, the hydroponic technique can be considered an innovative and useful technology to treat raw sewage by phytoremediation. It may require space horizontally instead of vertically. Among the free-floating aquatic plants, water hyacinth consists of large and beautiful flowers, is a native plant in Malaysia, and is also equipped with a robust root system and a high growth rate [20]. It is a promising candidate to be employed for nutrient removal from domestic wastewater. Thus, this research aimed to analyze the capability of WH in treating raw sewage in a hydroponic system. The hydroponic technique provides a controlled environment for water hyacinth. It is expected to be more efficient in removing nutrients from raw sewage and also to shorten the hydraulic retention time. The growth rate of water hyacinth will be monitored, and the treated sewage water quality will be analyzed. The presented results are expected to provide insight into raw sewage treatment using phytotechnology while also contributing to reducing further environmental pollution.

2. Materials and Methods

2.1. Sampling Location

The study was conducted at one of the semiconductor manufacturing plants in Senawang, Negeri Sembilan. The raw sewage was collected at a site away from the mixing of cafeteria wastewater. The coarse solids of the raw sewage were filtered out before transferring into a collection tank for research study purposes.

2.2. Source and Propagation of Water Hyacinth

The plants, water hyacinth, were collected from Universiti Kebangsaan Malaysia's pond near the Engineering Faculty. The plants were cleaned and propagated in a 1000 L tank with raw water placed in an open area (10 h of full sunlight). The plants were cleaned with raw water to remove the dried leaves and insect larvae grown on the plants. The plants were cultivated in raw water with minimal nutrients from sewage effluent under natural sunlight to let them adapt to the new environment for 1 week.

2.3. Experimental Setup for Batch Phytoremediation of Raw Sewage

There are 13 units of plastic boxes prepared for batch treatment. The size of the box is 27.5 cm (width) × 37.5 cm (length) × 42.5 cm (height). The reactor dimension was selected to accommodate the length of the water hyacinth's root. The pilot hydroponic system was placed in the sheltered area (10 h of sunlight with a temperature of 28–35 °C) to prevent rainwater mixing into the solution as a dilution factor.

At first, the water hyacinth was tested under batch phytoremediation. The plants were weighted and arranged into three groups: group 1 (4 WH—total plant weight range of 250–300 g); group 2 (5 WH—total plant weight range of 350–400 g); and group 3 (6 WH—total plant weight range of 450–500 g). The purpose of the batch experiment is to identify the effect of plant quantity on the effectiveness of raw sewage treatment and the duration for organic pollutant removal. Figure 1 shows the batch experiment setup.

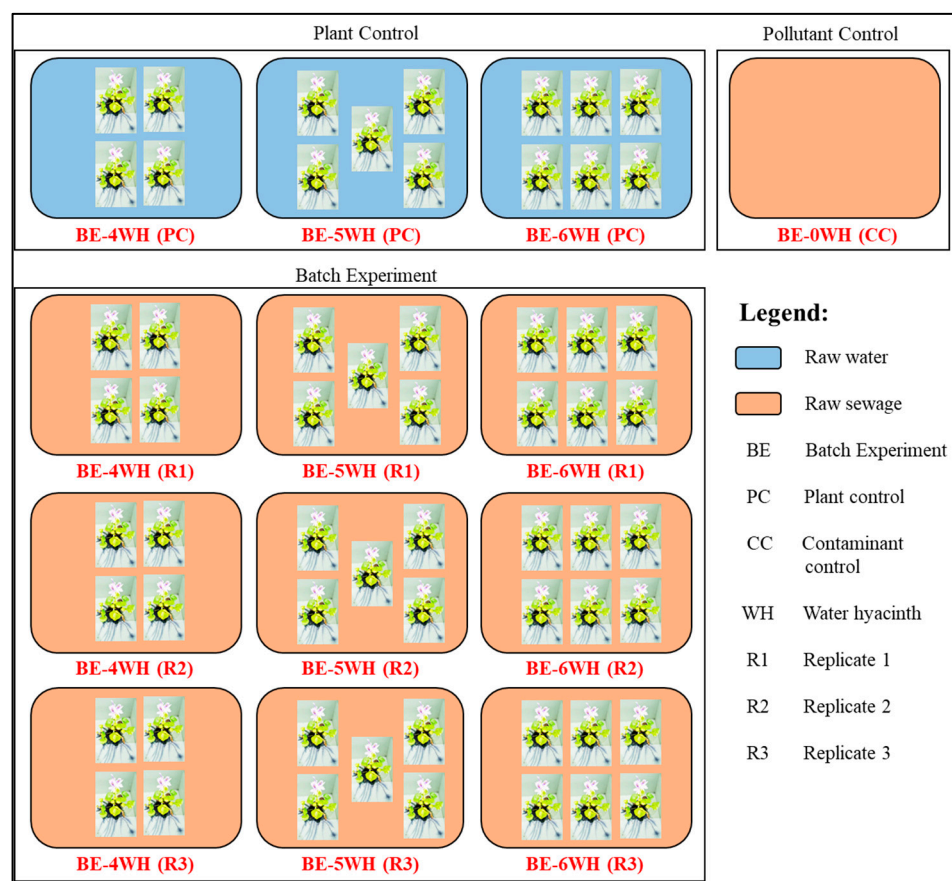


Figure 1. Batch experiment setup.

2.4. Water Sampling and Analysis

Water samples will be collected at each box of the batch experiment. The batch phytoremediation testing was run until one of the plant factor hydroponic systems met the Standard B discharge parameter. Water samples were analyzed for pH (pH Meter, pH72, Japan), DO (YSI DOmeter, ProODO, Yellow Springs, OH, USA), COD (USEPA reactor digestion method 8000 using DRB200, HACH, Loveland, CO, USA), BOD₅ (Winkler method, [21]), NH₄ (USEPA1 Nessler Method 8038 using HACH DR6000, Loveland, CO, USA), NO₃ (USEPA1 Nessler Method 8039 using HACH DR6000, Loveland, CO, USA), TP (USEPA 1,2 PhosVer 3[®] method 8048 using HACH DR6000, Loveland, CO, USA), Total Suspended Solid (TSS) (Photometric method 8006 using HACH DR6000, Loveland, CO, USA), and color (platinum–cobalt method 8025 using HACH DR6000, Loveland, CO, USA). All analysis was conducted in triplicate and presented as mean \pm SD. The data were statistically analyzed using the Statistical Package for Social Sciences (SPSS) software version 21.0.

2.5. Plant Sampling and Analysis

Water hyacinth plant total weight was measured on alternate weekdays. The plants were dried for 10 min before weighing. After being weighed, the plant was replaced in the same location in the pilot hydroponic system. An appointed water hyacinth plant was selected to monitor the physical growth; the root length, weight of the plant, and size of the plant were recorded [22].

2.6. Statistical Analysis

The data from water analysis and plant growth weight were analyzed to examine the performance of the water hyacinth phytoremediation on raw sewage. The data were statistically analyzed using the Statistical Package for Social Sciences (SPSS) software. The one-way analysis of variance (ANOVA) using SPSS was performed to determine whether there are any statistically significant differences between the means of two or more independent groups [23].

3. Results and Discussions

3.1. Characteristics of Raw Sewage Wastewater at the Study Site

The raw sewage wastewater was collected directly from the sewage pump sump for the batch experiment. Table 2 shows the characteristics of raw domestic wastewater used in this study. It shows that the COD and BOD concentration of raw sewage wastewater is far higher compared to the Environmental Quality Act 2009 for sewage [8]. Total suspended solids (TSS) concentration is below the discharging limit of Standard B. However, it is higher compared to the discharge limit of Standard A. The COD, BOD, and TSS are considered significant parameters for raw sewage.

Table 2. Raw sewage wastewater characteristics at the site.

Parameter	Raw (Tap) Water	Raw Sewage Wastewater	EQA (Sewage) Regulations 2009 Malaysia [8] *	
			Standard A	Standard B
pH	7.28 ± 0.16	7.31 ± 0.05	6.0–9.0	5.5–9.0
DO	5.95 ± 1.62	0.25 ± 0.04	NA	NA
BOD (mg/L)	8.26 ± 4.31	277 ± 15.4	20	50
COD (mg/L)	74.53 ± 8.6	516 ± 12.7	120	200
Ammoniacal Nitrogen (mg/L)	3.03 ± 2.87	18 ± 0.34	20	50
Nitrates (mg/L)	1.39 ± 0.12	1.6 ± 0.34	20	50
Total Phosphate (mg/L)	1 ± 0.0	3.95 ± 0.18	5	10
Total Suspended Solid (mg/L)	4.13 ± 1.17	76.5 ± 5.30	50	100
Color (ADMI)	-	34.5 ± 2.35	NA	NA

Note: * NA: not available.

3.2. Plant Growth Monitoring

Domestic wastewater is full of nutrients, which are essential for plant growth. As mentioned by Rezanian et al. [24], the nutrient-enriched water is the most favorable for water hyacinth, in which this plant can grow very well in domestic wastewater. Table 3 represents the 4 WH, 5 WH, and 6 WH, respectively, as variables to test the interaction between the number of plants factor and raw sewage contaminant removal. The tables show the plant growth observations from each group in batch phytoremediation versus the plant control. As observed, the water hyacinth grew healthily throughout the Day 10 study in both water conditions (sewage wastewater and tap water). When the plants reached the congested density in the experiment box, some of the plants' leaves started to turn yellow.

During the 10-day batch experiment, water hyacinth wet weight growth was monitored. In Figure 2, the solid line represents plants weight gained in sewage wastewater, whereas the dashed line represents plants weight in tap water. The final weight of the plants grown in sewage was statistically higher for 4 WH and 6 WH ($p < 0.05$), as marked in Figure 2. The water hyacinth is favorable to grow in high-nutrient solutions [25]. In the 10-day experiment, the total wet weight gained for the 4, 5, and 6 WH plant groups increased from 282 g to 575 g (104%); 381 g to 670 g (76%); and 488 g to 746 g (53%), respectively. WH plants are able to tolerate low concentrations of nutrient water as well [24]. The

total wet weight gained for 4, 5, and 6 WH plant control was 250 g to 486 g (94%), 395 g to 648 g (64%), and 471 g to 695 g (48%), respectively. A similar result was also obtained from previous research, in which water lettuce was grown positively in sewage wastewater after 35 days in a hydroponic system [26]. Aquaculture effluent is also reported to result in positive biomass growth for water lettuce grown in a hydroponic system for over 30 days [27].

Table 3. Plant growth observation.













	Day 0	Day 10
4 WH Plant		
4 WH Plant—Control		
5 WH Plant		
5 WH Plant—Control		

Table 3. *Cont.*

	Day 0	Day 10
6 WH Plant		
6 WH Plant—Control		

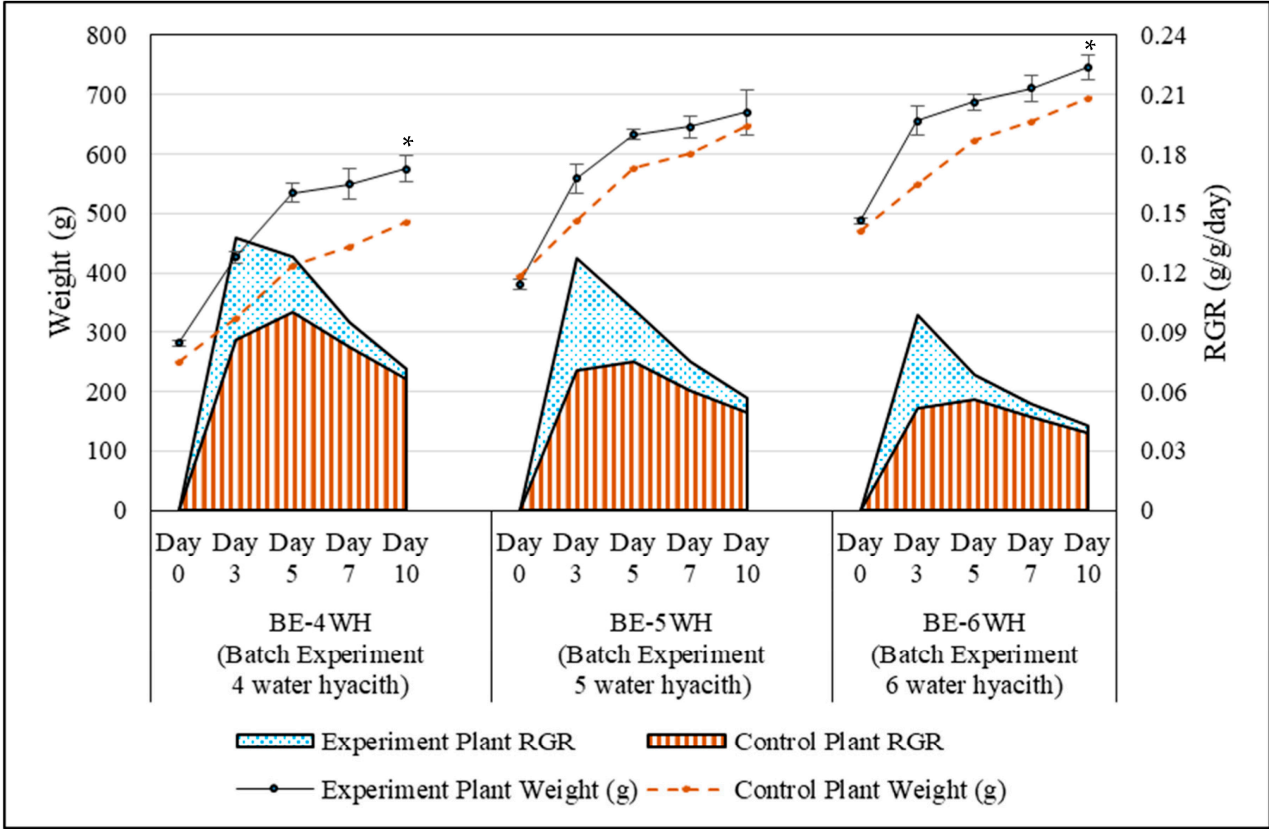


Figure 2. Variation of plant weight. Vertical error bars indicate mean \pm SD of three replicates. An asterisk (*) indicates a significant difference in plant weight between control and experimental plants.

A high relative growth rate (RGR) of plants that grew in sewage wastewater was observed when comparing the RGR of plants in the control that grew in tap water. Furthermore, the 4 WH plants group, which grew in either sewage wastewater or tap water,

gained the highest RGR of 0.14 g/g/day and 0.1 g/g/day, respectively, if compared to the 5 WH plants group (0.13 g/g/day and 0.07 g/g/day) and the 6 WH plants group (0.10 g/g/day and 0.06 g/g/day), respectively, as shown in Figure 2. The chart also shows that the RGR is in a reducing trend after Day 3 in the experimental group and after Day 5 in the plant control group. This shows that the water hyacinth growth rate is higher when there is more space for plants to expand and grow horizontally. This phenomenon can be related, as water hyacinth is able to grow at 12 acres per day [28]. However, the growth rate is not significantly different after Day 3 since the box space was already congested.

Overall, the RGR of water hyacinth grown in sewage (experiment) was significantly higher compared to tap water (control). These results were expected due to the availability of more nutrients in sewage as compared to tap water. In accordance with the obtained results, water lettuce also showed higher RGR while grown in nutrient-supplemented greywater (0.07 g/g/day) as compared to the tap water (0.06 g/g/d) [29]. In addition to that, the hydroponic system allowed a better nutrient provision as compared to the conventional method (soil-based), which can result in a better RGR for pak choi (0.18 vs. 0.16 g/g/d) [30].

3.3. Water Quality Monitoring

In Table 4, the sewage wastewater was monitored from the phytoremediation study and the contaminant control box. It is obviously observed that the clouded yellowish color from the original sewage wastewater was reduced and turned to slightly clear water, whereas the control sewage wastewater sample was increasingly cloudy on Day 3. This may be due to the exponential (log) phase and the stationary phase, where the bacteria developed and formed a layer of slime on the surface of raw sewage [31]. On Day 5, algae were also observed growing in the control box, and the algae fully covered the box on Day 7. On Day 10, the control wastewater's suspended solid was reduced, and a clear greenish solution could be observed.

Table 4. Water changes monitoring.











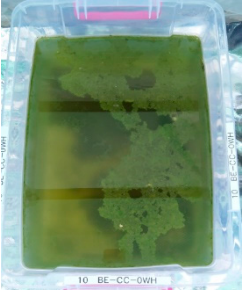







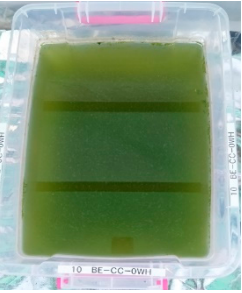
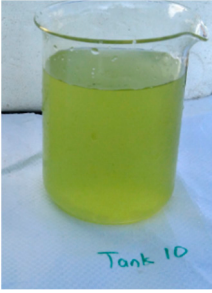
Setup Day	With Water Hyacinths	Water Sample with Water Hyacinths	Control	Water Sample Control
Day 0				
Day 3				

Table 4. Cont.

Setup Day	With Water Hyacinths	Water Sample with Water Hyacinths	Control	Water Sample Control
Day 5				
Day 7				
Day 10				

3.3.1. Biochemical Oxygen Demand (BOD)

The BOD concentration in the sewage wastewater was 233 ± 30.3 mg/L. In Figure 3, the initial BOD concentration removal from Day 0 to Day 3 is only <10% for the 4 and 5 WH plant groups and 22% for the 6 WH plant groups ($p < 0.05$). This may be due to the exponential phase for bacteria to activate and degrade BOD. However, there is statistically significant ($p < 0.05$) BOD removal from Day 3 to Day 5 in all three sets of batch hydroponic setups. From Day 3 to Day 5, the BOD removal rate significantly increased by 71% in the 4 and 5 WH plant groups; the 6 WH plant group managed to reduce 80% of BOD. Rezanian et al. [24] found that water hyacinth was able to reduce the BOD by 61%, which is from 9 mg/L to 3.5 mg/L in 14 days. With the additional introduction of bio-hedge material in the phytoremediation process, water hyacinth is able to reduce the BOD up to 88% removal from 215 mg/L to 26 mg/L in 35 days [5]. Another hydroponic plant of *Amaranthus campestris* also showed 90% removal of BOD from secondary wastewater effluent after 50 days in hydroponic treatment [32]. Nevertheless, from Day 5 to Day 10, a minor BOD rising trend was observed ($p < 0.05$). This may be due to some of the water hyacinth starting to wither, organic degradation, and being dissolved into the body. Even though the BOD removal had reached the highest removal of 70–80%, the BOD level of the raw sewage was still maintained at the borderline of the sewage Standard B discharge limit of 50 mg/L.

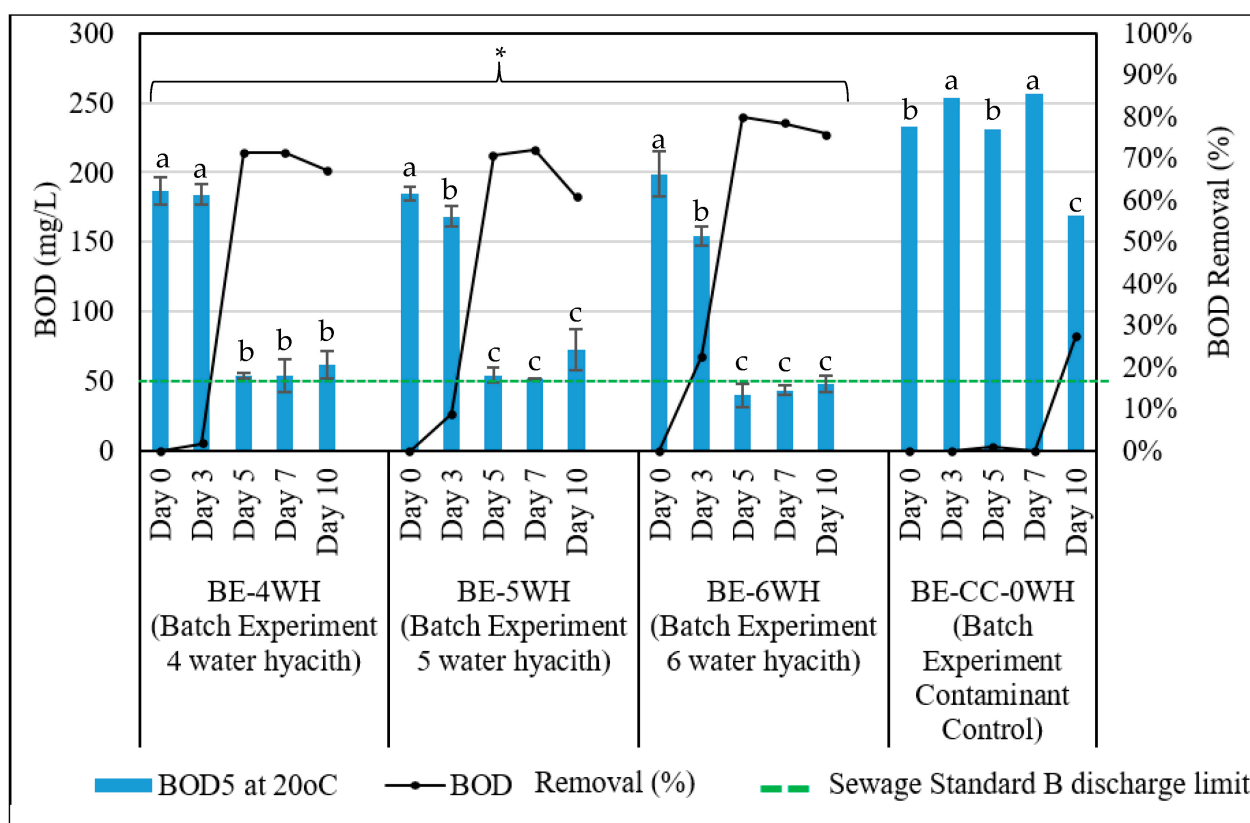


Figure 3. BOD concentration response with time exposure and plant factor monitoring. Vertical error bars indicate \pm SD of three replicates. Different letters above the graphs (a-b-c) indicate significant differences in BOD value between observation days under the same treatment condition. An asterisk (*) indicates a significant difference in final BOD value between the control and the experiment.

The BOD trend in the control setup is significantly different from the phytoremediation setup. The BOD level from Day 0 to Day 3 increased by 10%, where the BOD concentration was 223 mg/L to 254 mg/L. This may be due to the exponential (log) phase and stationary phase, where the bacteria develop and form a layer of slime on the surface of raw sewage [31] and contribute to the BOD concentration. The slime layer can be clearly seen in Table 4. However, on Day 5, the BOD level was reduced by 10%, and the BOD concentration was reduced from 254 mg/L to 231 mg/L. The removal of BOD may be caused by algae forming, and the algae play a role in organic degradation. Despite this, the BOD has again increased by 10% from 231 mg/L to 256 mg/L. The measured BOD may be contributed by the algae themselves, as the water sampling was in a greenish form with large suspended solids. There was 37% BOD removal from Day 7 to Day 10, where the BOD concentration was 256 mg/L to 169 mg/L. Although the water sample (Table 4) still looked greenish in Day 10. However, there were no suspended solids that could be found in the solution. Algae may also degrade the suspended solids for their growth.

3.3.2. Chemical Oxygen Demand (COD)

COD has a similar trend as BOD, and the correlation between COD and BOD is statistically significant ($r = 0.986$; $p < 0.05$). Both COD and BOD analyses are used to determine the oxygen demand in the solution. The key difference between BOD and COD is that the BOD is the oxygen demand of microorganisms to oxidize organic matter in the water under aerobic conditions, while the COD is the oxygen demand to oxidize all the pollutants in the water chemically [33].

Based on Figure 4, water hyacinth was able to reach the highest COD removal of 75%, 78%, and 77% by 4, 5, and 6 WH plant groups, respectively, on Day 5. The efficiency of COD removal is not statistically significantly different ($p > 0.05$) between the numbers of plants factor throughout the 10 days of the phytoremediation process, but it was significantly different as compared to the control ($p < 0.05$). This was observed in Rezanian et al.'s [34] study that the water hyacinth reduced 80% of COD during the 1st week of batch phytoremediation. *A. campestris* was reported to reduce COD by up to 58.5% after 10 days of hydroponic treatment, with an extension of treatment period to 50 days resulting in an increment of 18.36% removal [32]. Figure 4 also shows that the three sets of WH plant groups have reduced the COD concentration below the sewage Standard B discharge limit of 200 mg/L on Day 5.

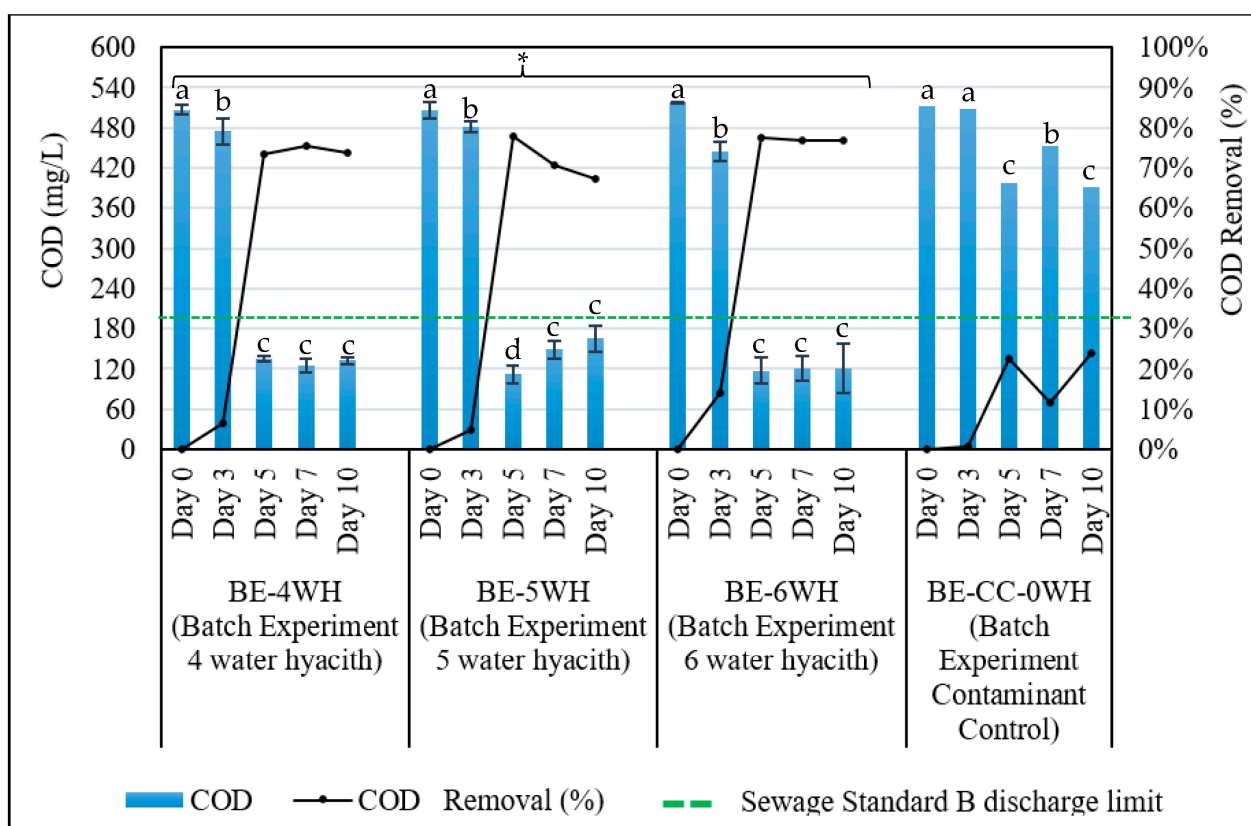


Figure 4. COD concentration response with time exposure and plant factor monitoring. Vertical error bars indicate \pm SD of three replicates. Different letters above the graphs (a-b-c-d) indicate significant differences in COD value between observation days under the same treatment condition. An asterisk (*) indicates a significant difference in the final COD value between the control and the experiment.

In the control reactor, the COD removal rate is not statistically significant ($p > 0.05$) from Day 0 to Day 3. When the algae grew on Day 5, the COD in the sewage wastewater was reduced by up to 20%. According to Vymazal [35], the water hyacinth root system creates the environment for microbial growth and indirectly provides oxygen at the rhizosphere for microbial degradation of organic pollutants. This is the main reason, as phytoremediation obtained higher COD and BOD removal compared to the contaminant control setup.

3.3.3. Ammoniacal Nitrogen (NH_4)

Nitrogen is one of the common elements in living things. It is one of the fundamental elements to build proteins and nucleic acids in an organic molecule [9]. Nitrogen is also one of the important nutrients to sustain plant growth [36]. In Figure 5, the initial ammoniacal

nitrogen (NH_4) for the three sets of WH plant groups and contaminant control on Day 0 is in the range of 1.80–2.80 mg/L. NH_4 was significantly reduced on Day 3, where the 4 and 5 WH plant groups gained a removal rate of 75% and 85%, respectively ($p < 0.05$). This is mainly due to immediate nutrient uptake by plants after the 2-week propagation period in tap water conditions. Akinbile and Yussof's [37] study shows that ammoniacal nitrogen was reduced by 96.12% by water hyacinth and 91.82% by *Pistia stratiotes* in the 3rd week under continuous conditions. In another research, water hyacinth also showed a capability of ammoniacal nitrogen removal up to 77.48% after 9 days of treatment [38].

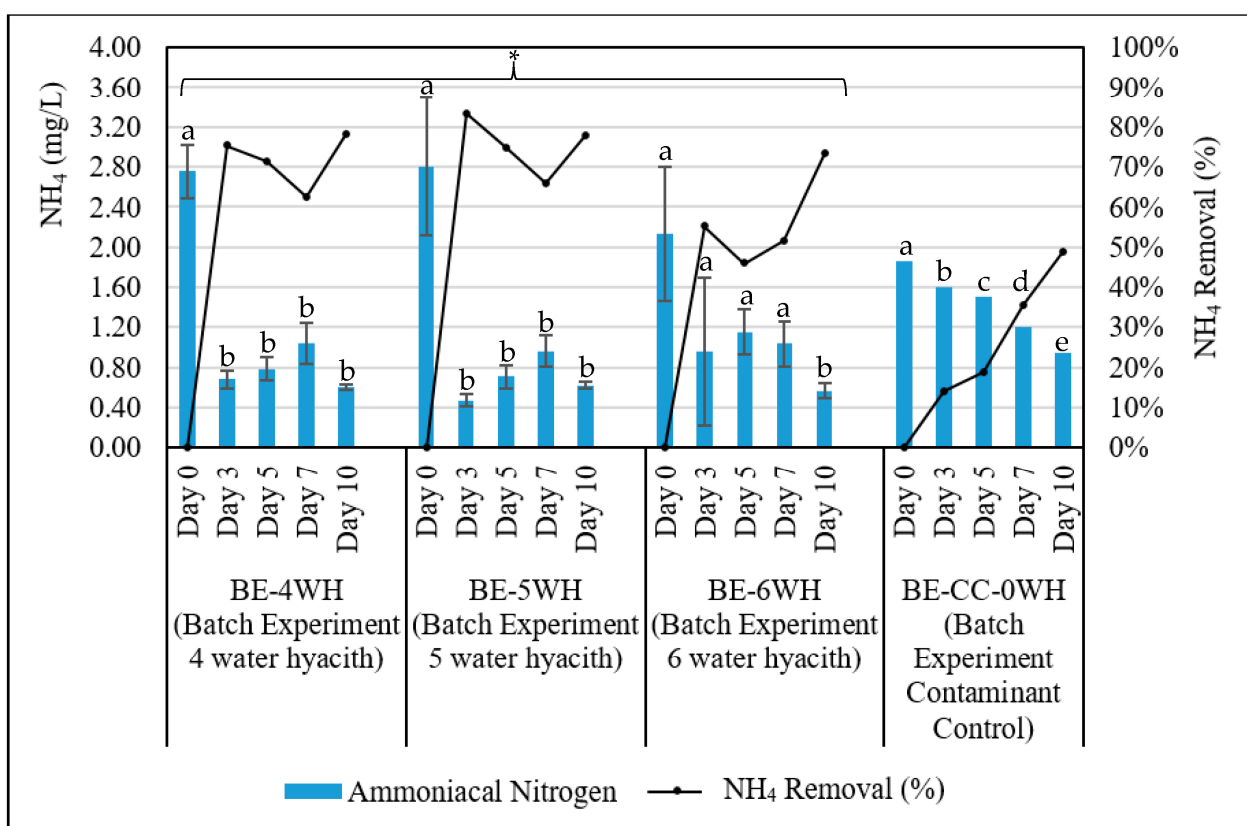


Figure 5. NH_4 concentration response with time exposure and plant factor monitoring. Vertical error bars indicate \pm SD of three replicates. Different letters above the graphs (a-b-c-d-e) indicate significant differences in NH_4 value between observation days under the same treatment condition. An asterisk (*) indicates a significant difference in final NH_4 value between control and experiment.

However, the NH_4 removal was slightly reduced from Day 3 to Day 7. The reducing removal rate trend of NH_4 similarly occurred in the three sets of the WH plant group setup. This may be due to the decomposition of the soluble organic nitrogen and the urea and uric acid, as well as the suspended solids of organic nitrogen from the filamentous bacteria [36,37]. Again, there was a removal of NH_4 from Day 7 to Day 10, indicating that the plants' nutrient uptake supported biomass growth and development during this period [9].

The removal of NH_4 in the contaminant control experiment setup only achieved 14% on Day 3. From Day 5 to Day 10, the NH_4 removal in the control sharply increased from 17% to 54%. This was due to the algae forming as shown in Table 4. Algae also prefer to take up NH_4 directly compared to NO_3 [9].

3.3.4. Nitrate (NO_3)

Nitrogen can be presented in several forms in wastewater treatment according to the oxidation states [9]. Nitrates are the conversion of NH_4 through nitrification by nitrifier bacteria when oxygen is present. Nitrates will be further converted to nitrogen gas through denitrification by heterotrophic bacteria by using nitrates as an oxygen source when under anoxic conditions [39]. At the rhizosphere, microbial activity accelerates the nitrogen decomposition, enhances the other nutrient elements, and improves the bioavailability of nitrogen for plant uptake [9].

In Figure 6, nitrate levels reduced significantly ($p < 0.05$) from Day 0 to Day 3 in the three sets of WH plant group setups. This may be due to the low oxygen presence, which created the environment for the denitrification process. However, the nitrate concentration increased significantly ($p < 0.05$) from Day 3 to Day 10 when DO levels increased in the three sets of WH plant groups, as discussed in Section 3.3.8. There is a slight fluctuation of nitrate concentration from 2.40 to 2.70 mg/L, 2.17 to 2.47 mg/L, and 2.10 to 2.37 mg/L in the 4, 5, and 6 WH plant groups, respectively, from Day 5 to Day 10, which may be due to the nitrogen uptake by water hyacinth and bacteria in the solution. Nitrogen is only available to be absorbed by plant roots in two forms: NH_4 and nitrates. However, plants prefer to take up NH_4 compared to nitrate, according to Ting et al. [9]. Reddy and Tucker's [40] research findings further supported that water hyacinth will consume nitrates when the NH_4 content is nearly depleted in the water medium. Other plants such as *Epipremnum aureum*, *Codiaeum variegatum*, and *Syngonium podophyllum* were mentioned to be capable of nitrate removal from aquaculture effluent with efficiency $> 70\%$ [41].

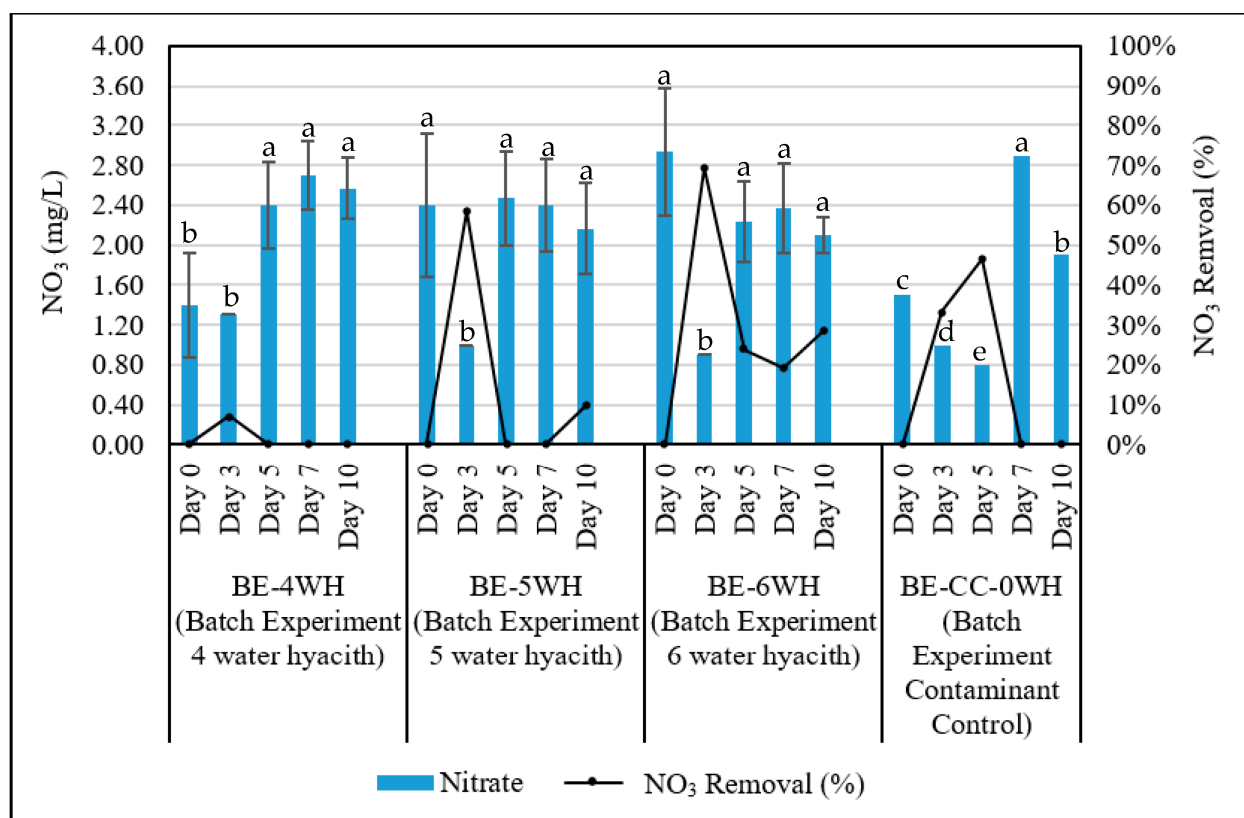


Figure 6. NO_3 concentration response with time exposure and plant factor monitoring. Vertical error bars indicate \pm SD of three replicates. Different letters above the graphs (a-b-c-d-e) indicate significant differences in NO_3 value between observation days under the same treatment condition.

In the control experiment, there was nitrate removal from Day 0 to Day 5, mainly due to the denitrification process, as the DO level is low. As discussed in Section 3.3.8, the DO level in the control experiment slightly increased on Day 5 and maintained until Day 10. A very obvious observation in Table 4 is that the algae forming on Day 7 caused the high nitrate level in the control sampling. From Day 7 to Day 10, the nitrate concentration again drops from 2.8 to 1.8 mg/L. Overall, there was no significant difference in final nitrate concentration between the experimental and control groups ($p > 0.05$).

3.3.5. Total Phosphate (TP)

Phosphate is one of the most important elements for plant biomass growth. Figure 7 shows that the total phosphate removal rate is statistically significant ($p < 0.05$) on Day 3. The removal rate reached 75%, 72%, and 76% in the 4, 5, and 6 WH plant groups, respectively, on Day 3. However, the trend of total phosphate fluctuated and was not statistically different from Day 3 to Day 10 ($p > 0.05$), especially in the 4 WH and 5 WH plant groups. According to Vymazal [35], most of the phosphorus is absorbed by plant roots, with the highest uptake rate occurring at the beginning of the plant's growing season. However, the storage of phosphorus might not be long term, and it can be leached out during decomposition. Akinbile and Yussof [37] reported that the phosphorus trend was reduced by 85.03% from the 1st week to the 3rd week by water hyacinth under the aquaculture influent. However, the phosphorus concentration was increased from the 3rd week (4.87 mg/L) to the 4th week (7 mg/L) due to the accumulation of detritus plants.

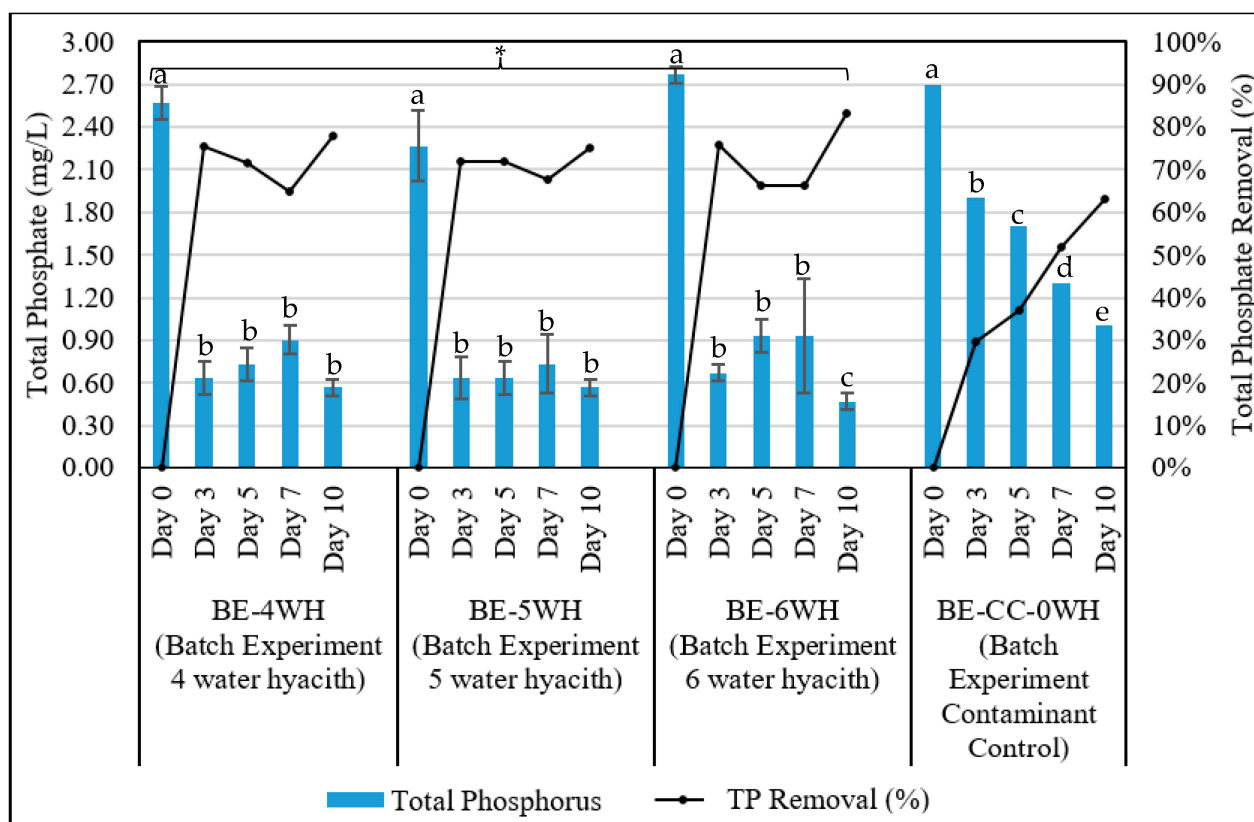


Figure 7. Total phosphate concentration response with time exposure and plant factor monitoring. Vertical error bars indicate \pm SD of three replicates. Different letters above the graphs (a-b-c-d-e) indicate significant differences in TP value between observation days under the same treatment condition. An asterisk (*) indicates a significant difference in final TP value between control and experiment.

The total phosphate concentration removal gradually increased in the control group, as shown in Figure 7, compared to the three sets of WH plant groups. According to Vymazal [35], the total phosphate uptake rate by microbial is very fast. However, the amount of storage is very low. In contrast, the uptake rate by microbiota (fungi, algae, etc.) is rapid due to this organism's growth multiplying at a higher rate. The total phosphate removal rate shows statistical significance ($p < 0.05$) in Days 0 to Day 3 from 2.70 to 1.90 mg/L due to the initial microbial uptake, and from Day 5 to Day 7 from 1.70 to 1.30 mg/L when the algae are forming. Overall, the experiment groups showed significant differences in the final TP concentration as compared to the control group ($p < 0.05$).

3.3.6. Total Suspended Solid (TSS)

Figure 8 shows that TSS removal is minimal from Day 0 to Day 5 for the 4 and 5 WH plant groups (11% and 3%, respectively). The TSS removal rate increased and was statistically significant ($p < 0.05$) from Day 5 to Day 7 for the 4 and 5 WH plant groups (68% and 83%, respectively). The TSS removal trend shows a slight improvement in the 6 WH plant group, but it is not significant ($p > 0.05$) compared to the 4 and 5 WH plant groups. The high removal rate of nutrients and TSS in wastewater during the treatment period in the hydroponic system was due to the development of biomass and root mats [42]. When the root mats are fully developed, they will increase the filtration capacity and cause the removal of TSS, and the nutrient absorption rate will increase [42]. WH is able to remove the TSS from raw sewage wastewater to reach below the sewage Standard B discharge limit of 100 mg/L.

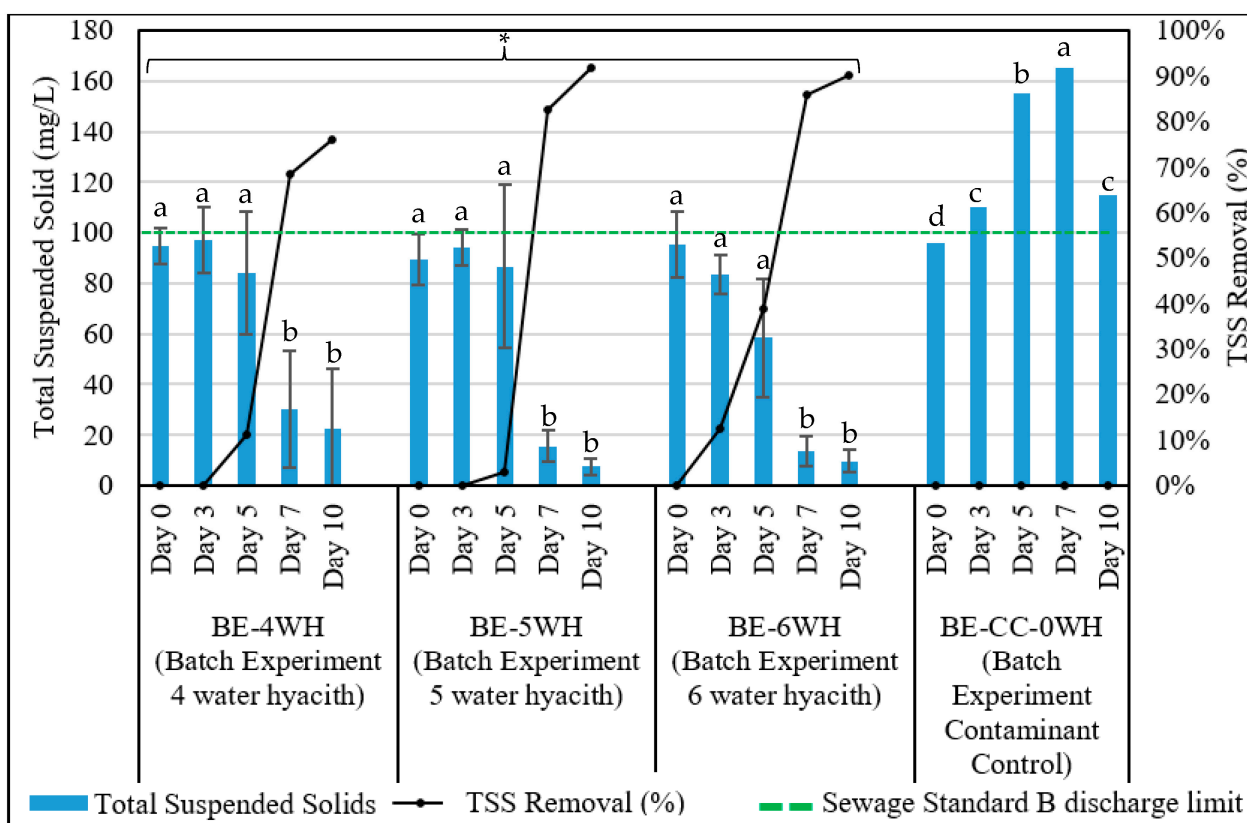


Figure 8. TSS concentration response with time exposure and plant factor monitoring. Vertical error bars indicate \pm SD of three replicates. Different letters above the graphs (a-b-c-d) indicate significant differences in TSS value between observation days under the same treatment condition. An asterisk (*) indicates a significant difference in final TSS value between control and experiment.

The TSS removal shows a statistically significant difference ($p < 0.05$) in the control group compared to the WH plants group. The TSS concentration increased from Day 0 to Day 7 from 96 mg/L to 165 mg/L. This phenomenon is clearly shown in Table 4, where the formation of the bacteria slime layer on Day 3 and the algae formation on Day 5 are shown. The TSS is mainly contributed by microbial and algae. However, the TSS reduced to 115 mg/L on Day 10, when most of the TSS was degraded by algae.

3.3.7. Color

Many researchers commented that water hyacinth is able to remove different types of dye and has gained high attention for textile wastewater [43]. It was highlighted by Mahmood et al. [44], as water hyacinth's root is a cheap source of biosorbent for color. The statement is supported by the findings as shown in Figure 9. The overall color removal in raw sewage gained 40%, 59%, and 63% in the 4, 5, and 6 WH plant groups, respectively. Previous research mentioned the effect of plant biomass weight on color removal by *Juncus effusus*; the increase in initial wet weight from 1 g to 3 g resulted in an increment of color removal of up to 17.5% (81.5% to 99%) [45]. However, the color parameter is not listed in the sewage discharge standard limit. Therefore, the color removal of sewage wastewater is a positive value to ensure the minimal pollution of sewage effluent into the water body.

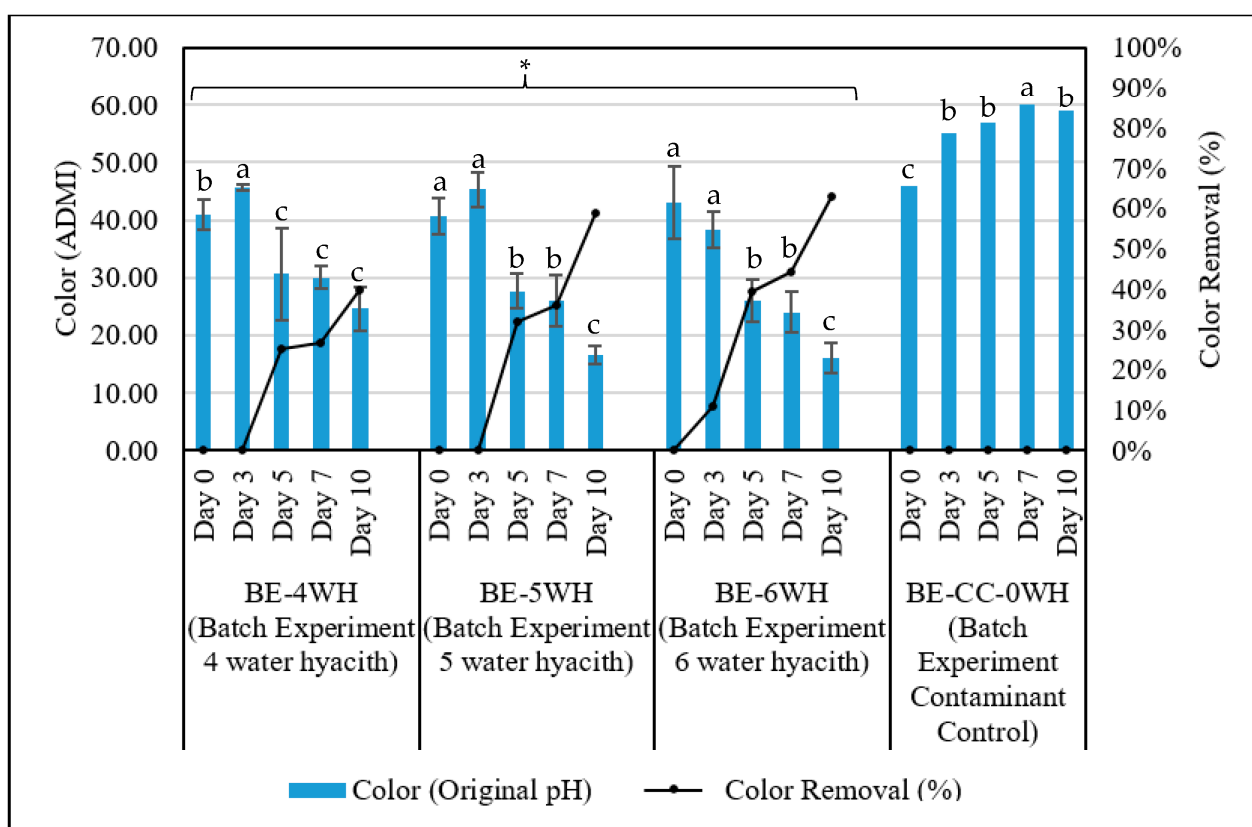


Figure 9. Color concentration response with time exposure and plant factor monitoring. Vertical error bars indicate \pm SD of three replicates. Different letters above the graphs (a-b-c) indicate significant differences in color concentration between observation days under the same treatment condition. An asterisk (*) indicates a significant difference in final color concentration between control and experiment.

The color removal trend showed a significant difference compared to the WH plant groups ($p < 0.05$). However, there was no significant difference in final color concentration among different numbers of WH plants ($p > 0.05$). The color concentration increased

throughout the 10-day experiments. The main contributors are the filamentous forming on Day 3, followed by algae forming on Day 5 onwards, as clearly shown in Table 4. Water hyacinth has the ability to promote algae interception from sewage wastewater, as reported in Qin et al. [11]. Therefore, the effluent's color from the experimental groups is significantly reduced.

3.3.8. Dissolved Oxygen (DO)

Oxygen is a fundamental element for living things to survive. Aerobic bacteria also require oxygen to survive and break down organics in wastewater treatment. Figure 10 shows the DO level trend in the three sets of phytoremediation and contaminant control. In the 6 WH plant group, it gained the highest DO rate, increasing from 0.21 mg/L to 5.1 mg/L. The increasing trend of DO levels in the 4, 5, and 6 WH plant groups was similar ($p > 0.05$). The observed DO in the experimental systems was significantly higher than compared in the control ($p < 0.05$). This is due to the feathery and fibrous root systems of the water hyacinths that provide an environment for bacterial activities [43,46]. When the plants go through photosynthesis, the oxygen concentration in the solution may increase due to the oxygen transport and release through the air spaces of stems and root zones of macrophytes [47]. Similar findings were reported by Akinbile and Yussof [37], where DO was increased from 3.83 mg/L to 5.23 mg/L by using water hyacinth for wastewater treatment within the 3-week experiment.

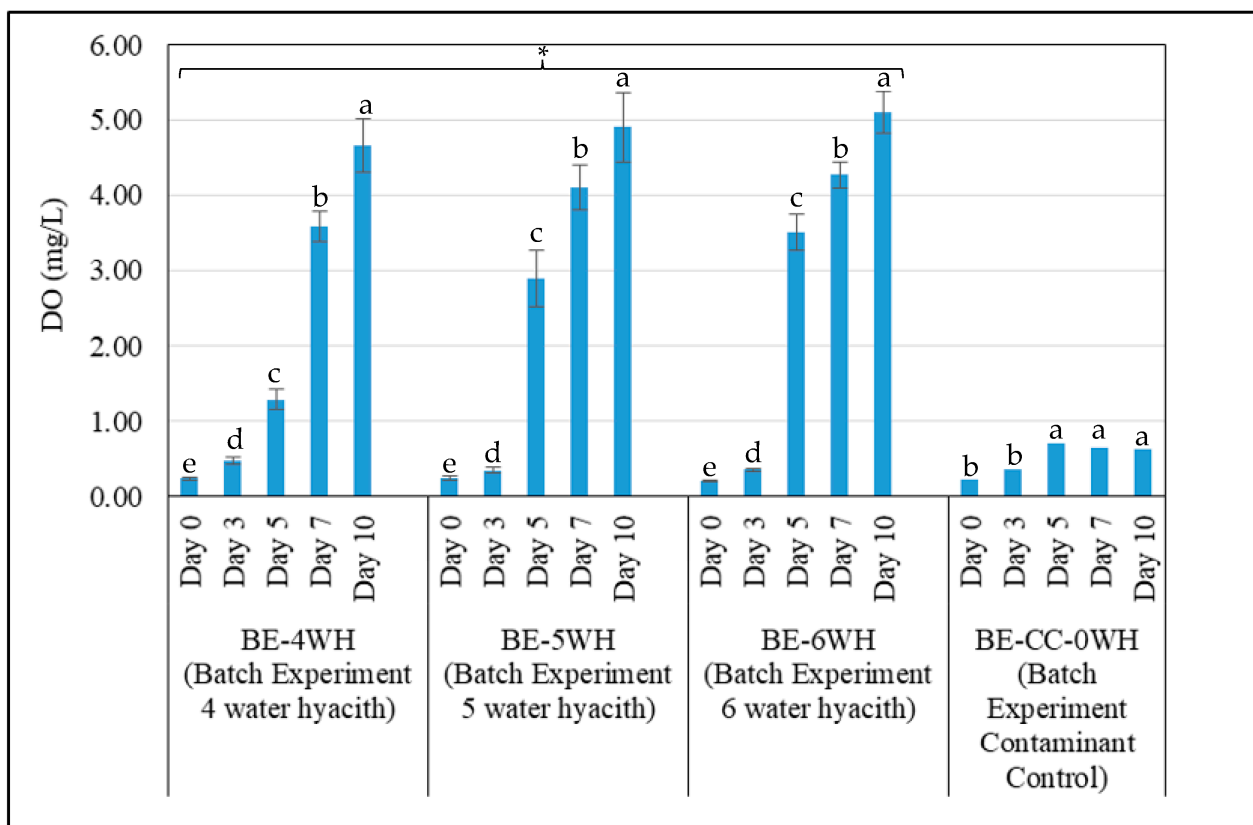


Figure 10. DO response according to time exposure and plant factor monitoring. Vertical error bars indicate \pm SD of three replicates. Different letters above the graphs (a-b-c-d-e) indicate significant differences in DO value between observation days under the same treatment condition. An asterisk (*) indicates a significant difference in final DO value between control and experiment.

The DO level in the control system is not statistically significant from the 0-day to the 3-day experiment. There is a slight increase in DO level from Day 5 to Day 7, as the algae

were observed to start forming. However, from Day 5 to Day 10, the DO level shows a slight declining trend. This may be due to the algae forming that consumed oxygen significantly, covered the water surface, and prevented oxygen from diffusing into the water body [48].

3.3.9. pH

The initial pH of domestic wastewater is almost neutral at pH 7.02–7.11 for the three sets of WH plant groups and contaminant control. As shown in Figure 11, the pH value dramatically reduced by 34% in the 4 WH plant group (from pH 7.06 to pH 4.66), 31% in the 5 WH plant group (from pH 7.05 to pH 4.90), and 28% in the 6 WH plant group (from 7.04 to pH 5.10) throughout the 10-day experiment. The pH change trends were statistically different from Day 0 to Day 5 ($p < 0.05$). However, the pH value in the control group was slightly more stable, but still, the pH dropped from pH 7.11 to pH 6.04. It was not statistically significantly different ($p > 0.05$) from Day 3 to Day 5, then Day 7 to Day 10 in the control group. Similar findings were reported by Akinbile and Yussof [37], where the pH dropped to 4.45 in the air circulation treatment system. pH is important for microbial activities, and the optimum pH values for the nitrification process may vary from pH 6.6 to pH 8.0 [49].

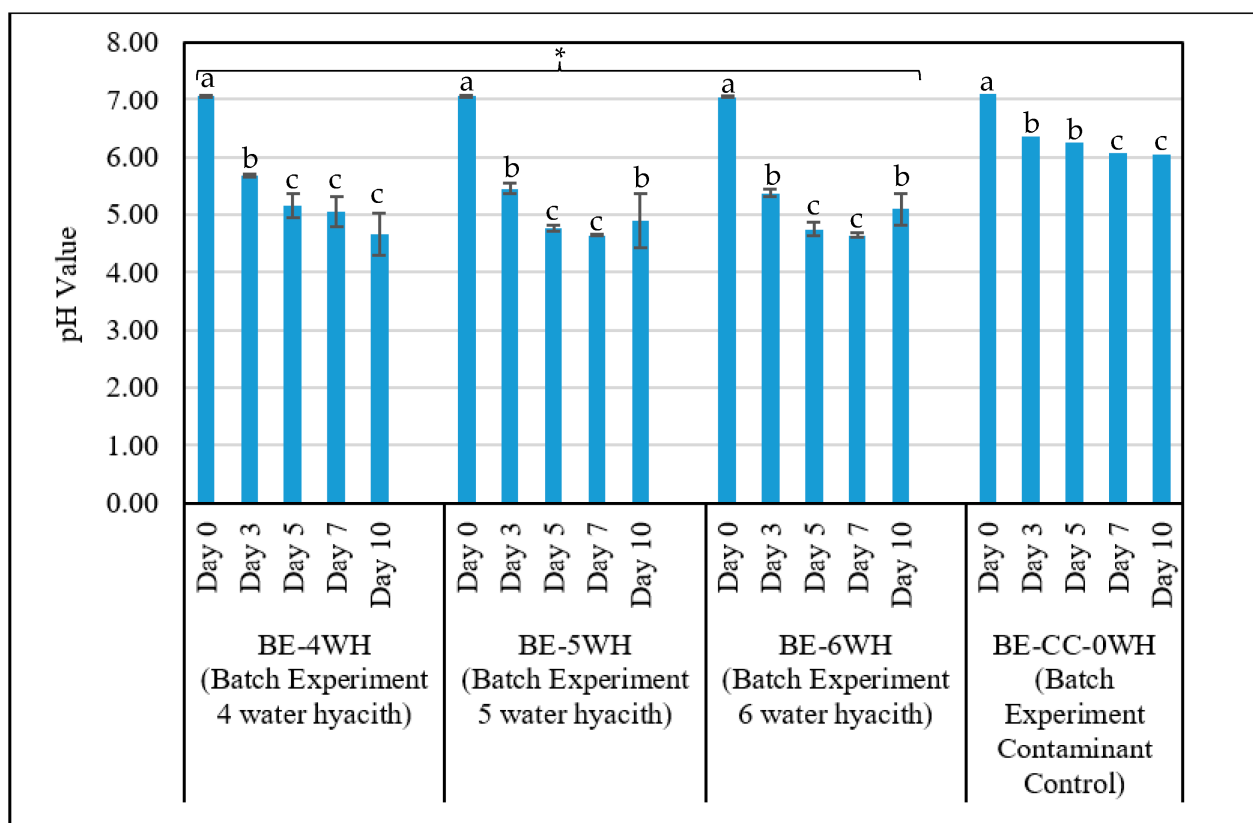


Figure 11. pH response with time exposure and plant factor monitoring. Vertical error bars indicate \pm SD of three replicates. Different letters above the graphs (a-b-c) indicate significant differences in pH value between observation days under the same treatment condition. An asterisk (*) indicates a significant difference in final pH value between the control and the experiment.

In SPSS correlation analysis, the interaction effect between pH and ammoniacal nitrogen shows a moderately strong strength of linear relationship ($r = 0.783$) and is statistically significant ($p < 0.05$) [35]. This is mainly due to the alkalinity of the wastewater. For every one part of ammonia (NH_4) converted to nitrate (NO_3), there will be 7.1 parts of alkalinity depleted. For every part of nitrate (NO_3) conversion to nitrogen gas, there will be a gain of

3.6 parts of alkalinity in the solution [39]. The pH trend decreased from pH 7.0 to below pH 5 from Day 0 to Day 10 when the nitrification process occurred, converting the NH_4 to nitrates discussed earlier. The nitrification process occurred with the presence of oxygen, as shown in Figure 10 of the increasing DO trend. The nitrification rate declines when the pH level is below pH 6, which was not experienced in the study due to the buffering effect of water hyacinths [47].

The pH drop in the solution may also be due to the organic nutrient degradation by the microbial action in domestic wastewater. Bacteria degrade the organic compound in the presence of oxygen and release CO_2 and water molecules [50]. CO_2 dissolves in the water and reduces the pH value in the solution. The interaction effect between pH towards COD and BOD is also statistically significant ($p < 0.05$; $r_{\text{COD}} = 0.825$ and $r_{\text{BOD}} = 0.841$) as simulated in the SPSS report, as shown in Table 5. In the control setup, the pH reduced very slowly compared to the other three sets of WH plant groups, mainly due to the low oxygen level in the control solution, as shown in Figure 10.

Table 5. Analysis of from two-way ANOVA Duncan post-hoc test on the interaction between two independent variables (number of plant factors and exposure period) versus the dependent variable (COD removal).

Exposure Period	Plant Group Comparison			Sig. (<i>p</i>)
Day 0	BE-4 WH	Compare to	BE-5 WH	0.899
	BE-4 WH	Compare to	BE-6 WH	0.432
	BE-5 WH	Compare to	BE-6 WH	0.363
Day 3	BE-4 WH	Compare to	BE-5 WH	0.594
	BE-4 WH	Compare to	BE-6 WH	0.031 **
	BE-5 WH	Compare to	BE-6 WH	0.009 **
Day 5	BE-4 WH	Compare to	BE-5 WH	0.078
	BE-4 WH	Compare to	BE-6 WH	0.161
	BE-5 WH	Compare to	BE-6 WH	0.703
Day 7	BE-4 WH	Compare to	BE-5 WH	0.078
	BE-4 WH	Compare to	BE-6 WH	0.722
	BE-5 WH	Compare to	BE-6 WH	0.037 **
Day 10	BE-4 WH	Compare to	BE-5 WH	0.017 **
	BE-4 WH	Compare to	BE-6 WH	0.350
	BE-5 WH	Compare to	BE-6 WH	0.002 **

Notes: ** Statistically significant when $p < 0.05$. Noted: BE = Batch experiment; WH = Water hyacinth.

According to the Environmental Quality Act 2009 for Sewage, the Standard B discharge limit for pH is pH 5.5–pH 9.0. The sewage effluent after the phytoremediation process may need to be adjusted before discharge to the environment.

3.4. SPSS Correlation Analysis for Water Hyacinth Phytoremediation Performance in Batch Experiment

In the batch experiment, the number of plant factors was a variable to be determined if it had a significant impact on COD removal. From the two-way ANOVA Syntax post-doc (Duncan) analysis, it shows that the number of plants is not statistically significant between the two independent variables (number of plant factors and exposure time) on the dependent variable (COD removal), as shown in Table 5. Only five data points out of fifteen showed that the interaction in different numbers of plants had a significant impact on COD removal. Therefore, 4 WH plants of water hyacinth were recommended for the hydroponic system. The low plant biomass towards a high pollutant removal rate is more preferable as well [25,28].

In Table 6, the pH value shows a statistically significant correlation to most of the variables except plant growth rate and NO₃ removal. This analysis supports the discussion earlier on the contaminants removal due to pH, which may be a significant variable affecting the microbial activities and the plant's pollutant uptake rate [46].

Table 6. The summary of the SPSS correlation on the interaction between two variables.

Variables	pH	
	Pearson Correlation (<i>r</i>)	Sig. (<i>p</i>)
Number of Plant Factors	−0.067	0.663
Exposure Period	−0.810	0.000 **
Plant Weight	−0.806	0.000 **
DO	−0.731	0.000 **
BOD	0.841	0.000 **
COD	0.825	0.000 **
TSS	0.607	0.000 **
NH ₄	0.783	0.000 **
NO ₃	−0.090	0.556
TP	0.871	0.000 **
Color	0.653	0.000 **

Note: ** Statistically significant when $p < 0.05$.

3.5. Future Research Direction

Important avenues for the continuation of this research are advancement into continuous systems, scaling up to larger scales, and valorization of the resultant biomass. Advancing from batch-scale to continuous systems represents a critical step toward real-world application [51]. The obtained optimum condition under batch scale can be adopted and adapted in the continuous system. For a continuous system, several aspects need to be considered, including the potential of automation [52] and maintaining consistent efficacy as an effort to integrate wastewater treatment [53]. Furthermore, scaling up into pilot or even real field scale should also be prioritized to reveal the real potential of this system. Scaling up research can be focused on the modular design approaches, hybridization, and cost-benefit analyses to bridge the gap between laboratory-scale experiments and real-world practical application.

In parallel to scaling up the treatment, future research should also focus on the valorization of the resultant biomass after hydroponic treatment. Depending on the pollutant type and level, resultant biomass can be explored for bioenergy production, organic fertilizer, and other value-added bioproducts. Digestion of water hyacinth juice after phytoremediation yielded 237.37 L of methane/kg VS [54]. Co-digestion of water lettuce with pig manure resulted in a maximum of 292.9 L/kg VS biogas, with methane content reaching 54% [55]. Water hyacinth's biomass contains high cellulose and hemicellulose, which can be hydrolyzed into reducing sugar to produce bioethanol. Previous research mentioned a yield of 1.289 g/L of ethanol produced from dried water hyacinth biomass [56]. Co-composting of aquatic plant biomass of *Pistia* sp. with cow dung (1:1) showed better chemical quality of biofertilizer as compared to cow dung only compost [57]. In the case of non-toxic, low pollutant accumulation, plant biomass can be used as animal feed [58] and mushroom cultivation substrates [59]. The use of a hydroponic system and its resultant biomass valorization is a good initiative toward circular economy and to support the Sustainable Development Goals Numbers 6 (clean water and sanitation), 12 (responsible consumption and production), and 14 (life below water).

4. Conclusions

Raw sewage effluent contains high amounts of COD, BOD, and TSS. The application of hydroponic systems using water hyacinth showed a promising natural-based solution to mitigate sewage pollution. The considerably high removal of BOD (73%), COD (73%), TSS (86%), NH_4 (77%), TP (79%), and color (54%) demonstrates the effectiveness of this method. Water hyacinth grown in wastewater showed an increasing biomass for up to 78% and significantly higher RGR as compared to the control (tap water). Statistical analysis indicates that the number of plant components did not yield a statistically significant change in all pollutant parameter removals, suggesting that 4 WH (4 plants) is adequate to obtain effluent after treatment below the sewage Standard B discharge limit, except for BOD (51 mg/L), which is slightly above the discharge limit of 50 mg/L. Water hyacinth also showed a capability of increasing dissolved oxygen level from 0.24 to 4.88 mg/L. Future research is suggested to optimize the treatment condition to comply with the regulatory standard while also advancing the treatment into a continuous system and pilot scale. Valorization of the resultant biomass after hydroponic treatment is also suggested.

Author Contributions: Conceptualization, S.R.S.A.; Formal analysis, L.Y.Y.; Investigation, L.Y.Y.; Resources, M.F.I.; Data curation, S.B.K.; Writing—original draft, L.Y.Y. and S.B.K.; Writing—review & editing, S.R.S.A., M.F.I. and S.B.K.; Visualization, S.B.K.; Supervision, S.R.S.A.; Project administration, S.R.S.A.; Funding acquisition, S.R.S.A. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by National University of Malaysia under research grant number DIP-2024-001 and the APC was funded by Delft University of Technology.

Data Availability Statement: The original contributions presented in this study are included in the article. Further inquiries can be directed to the corresponding authors.

Acknowledgments: The authors would like to thank National University of Malaysia for their support under DIP-2024-001.

Conflicts of Interest: The authors declare no conflict of interest.

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