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Performance evaluation and phytoremediation efficiency of selected aquatic macrophytes on aquaculture effluent

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Abstract
An experiment was carried out to evaluate the phytoremediation efficiency of six aquatic macrophytes, three floating (*Ipomoea aquatica, Alternanthera philoxeroides, Eichhornia crassipes*) and three submerged (*Cabomba caroliniana, Hydrilla verticillata, Ceratophyllum demersum*) on aquaculture effluent. Considering the nutrient dynamics of the experimental time period, all aquatic macrophytes were found to be effective when compared with the control. Floating aquatic macrophytes showed better performance till the first 28 days' period when compared with submerged aquatic macrophytes. At the end of the experiment, lowest total solids (TS), turbidity, total hardness, Chemical Oxygen Demand (COD) and Biological Oxygen Demand (BOD) levels, as well as the highest dissolved oxygen (DO) levels were observed in *C. demersum* followed by *H. verticillata*. Considering the overall time period, nutrient removal, and performances of other water quality parameters, *C. demersum* and *H. verticillata* can be suggested as the best macrophytes to treat aquaculture effluent among the selected aquatic macrophytes.

Keywords: Phytoremediation, aquaculture effluent, floating and submerged aquatic macrophytes

Introduction

Growing contamination of soil and water resources has become a major problem worldwide and they become polluted due to the uncertainty level of contaminants. Among the major reasons for this issue, intensive agricultural and unfriendly environmental activities had been identified. Many aquaculture systems, specially intensive farming systems generate high amounts of wastewater containing compounds such as suspended solids, total nitrogen and total phosphorus [1], causing serious environmental problems to the receiving water bodies. Unplanned, wasteful use of water in aquaculture is limiting the development of aquaculture industry and more research is essential for the sustainable water-use in aquaculture by minimizing the water use and reuse of water resources in aquaculture.

When considering the aquaculture wastewater, its composition is directly related to the nature and quantity of feed fed to the species being reared and also depends on the type of operation system of the culture. The major sources of effluent from aquaculture consist of excreta, fecal matter and uneaten feed from fish. Nitrogen and phosphorus are two major elements in aquaculture wastewater due to decomposing the excreta, fecal matter and uneaten feed from fish [2]. Various organic and inorganic compounds such as ammonium, phosphate, dissolved organic carbon and organic matter are also a rich source in aquaculture effluent [3, 4]. Decomposition and reuse of these nitrogenous compounds are especially important in recirculating aquaculture systems due to the toxicity of ammonia and nitrite, and the chance of hypertrophication of the environment by nitrate [5]. The food and fecal waste also constitute in wastewater are responsible for the majority of suspended solids. Thus, the high concentration of suspended solids (SS), Chemical Oxygen Demand (COD), Biological Oxygen Demand (BOD), total nitrogen and total phosphorus can be identified in the outlet of the aquaculture facilities. If these untreated aquaculture wastewater discharged to the water bodies, it will cause environmental deterioration of the receiving water bodies due to high levels of nutrients. Thus, nutrient removal is essential for aquaculture wastewater treatment to protect receiving water from eutrophication and for potential reuse of the treated water.

Aquatic plants play an important role in structural and functional aspects of aquatic ecosystems by various ways and phytoremediation is one of them [6].

Using aquatic macrophytes to remove nitrogen, phosphorus, and other contaminants from water are one way of phytoremediation to reduce negative impacts to the aquatic ecosystem. Phytoremediation can be defined as the use of plants and their associated microbes for environmental cleanup [7]. It uses plants to remove pollutants from the environment [8]. These pollutants, when absorbed by the plants, may be stored in the roots, stems, or leaves and changed into less harmful chemicals within the plant or changed into gases that are released into the air as the plant transpires [9]. Thus, phytoremediation is a cost-effective, efficient and environmentally friendly solution for the treatment of aquaculture wastewater and treated water can be reused for the aquaculture production to minimize the water demand for the aquaculture. The use of plants for nutrient uptake is especially valuable because it is used as site remediation and beside of that, phytoremediation is possible to identify as practical and value-added uses of those bio mass produced by the plant material. These could include conversion of plant biomass to composting, soil amendments, anaerobic digestion with methane production, and processing for animal feed.

The aim of the present study was to compare ability or efficiency of selected species of aquatic macrophytes to remediate aquaculture effluents for potential reuse of the water.

Materials and methods

The experiment was carried out in a period of 56 days at ICAR-Central Institute of Fisheries Education, Mumbai, India. The experimental setup was arranged to evaluate the phytoremediation efficiency of aquaculture effluent using different aquatic macrophytes. Six aquatic macrophytes were divided into two combinations, floating and submerged according to their living habitat. Three floating aquatic macrophytes (*Ipomoea aquatica, Alternanthera philoxeroides*, *Eichhornia crassipes*) and three submerged aquatic macrophytes (*Cabomba caroliniana*, *Hydrilla verticillata*, *Ceratophyllum demersum*) which was arranged as T1, T2, T3, T4, T5 and T6 treatments, respectively and control (C) without plants. Each treatment and the control were arranged with 3 replicates and the experimental setup was arranged in 21 tanks (100 L) with a complete randomized design. As an initial stocking, 400 g plants were taken and volume of aquaculture effluent was made to 90 L in each treatment.

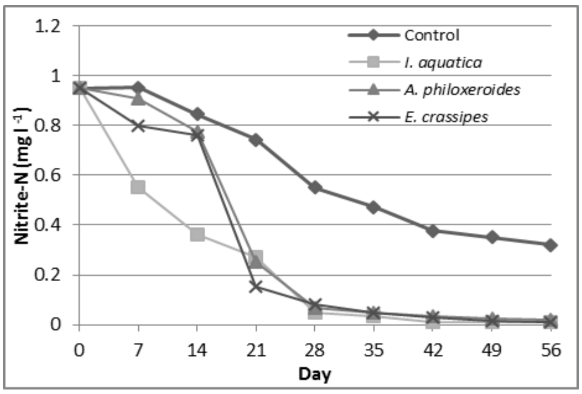
Water quality parameters and plant biomass were measured weekly and fortnightly respectively. OAKTON water proof pH and temperature tester 30 were used to measure pH and temperature. Dissolved oxygen (DO), Biochemical oxygen demand (BOD), Chemical oxygen demand (COD), total hardness, alkalinity, total solids (TS), ammonia-N, nitrite-N, nitrate-N, and phosphate-P were measured according to the standard methods outlined [10].

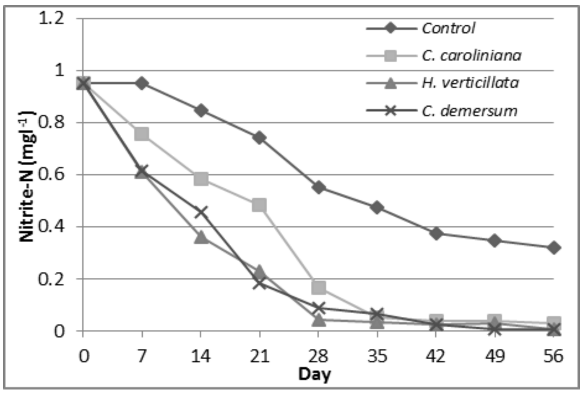
Statistical analysis

The data were analyzed by statistical package SPSS version 16 in which one-way ANOVA and Tukey's multiple range test were performed at a significance level of (P>0.05) 95% confidence limit to know the significant difference between the treatments and control.

Results

All aquatic macrophytes showed high phytoremediation activity when compared with the control. Variations of different nutrients (Nitrite-N, Nitrate-N, Ammonia-N and Phosphate-P) during the experiment are shown in Fig. 1 and 2. On the 28th day of the experiment, lowest nitrite-N remaining was observed in T6, but it was not significantly different from other treatments and the control. At the end of the experiment, the less nitrite-N remaining was observed in T5 and T6 treatments. *C. caroliniana* showed a higher amount of nitrite-N remaining in the system than the other treatments and it was also significantly different from T5 and T6. Also, all treatments showed significant difference with the control considering the nitrite-N concentration remaining at the end of the experiment. A nitrate-N remains in the experiment was depended on the plant species. In first 28 days, lowest nitrate-N remain in the T2 (2.02±0.08 mg L^{-1}) and it was significantly different with all other treatments. When comparing two types during the first 28 days, floating aquatic macrophytes showed fewer nitrate-N remain in the system compared to the submerged aquatic macrophytes. *C. caroliniana* showed the highest amount of nitrate-N remaining in the entire period of the experiment, but it was lower than the control. At the end of 28 days, ammonia-N concentration varied among treatments as T3<T2<T5<T4<T1<T6<C and *C. demersum* showed significant difference with the other treatments. At the end of the experiment, there was no significant difference in ammonia-N concentration between the treatments, and the control. At the end of the 28th day, remaining of phosphate-P concentration showed significant difference within the plant species and also with the control. The lowest phosphate-P concentration was found in T6 followed by T2< T1<T5<T3<T4<C. At the end of the experiment, T6 showed the lowest phosphate-P; but, it was not significantly different from the remaining of phosphate-P concentration in T5 and T1. Treatment 2 showed the 3rd lowest phosphate-P concentration and it was significantly different from other treatments. *E. crassipes* and *C. caroliniana* showed higher remaining of phosphate-P concentrations compared to the others, and also showed a significant difference between these two treatments as well as with the other treatments and the control (Table 1).





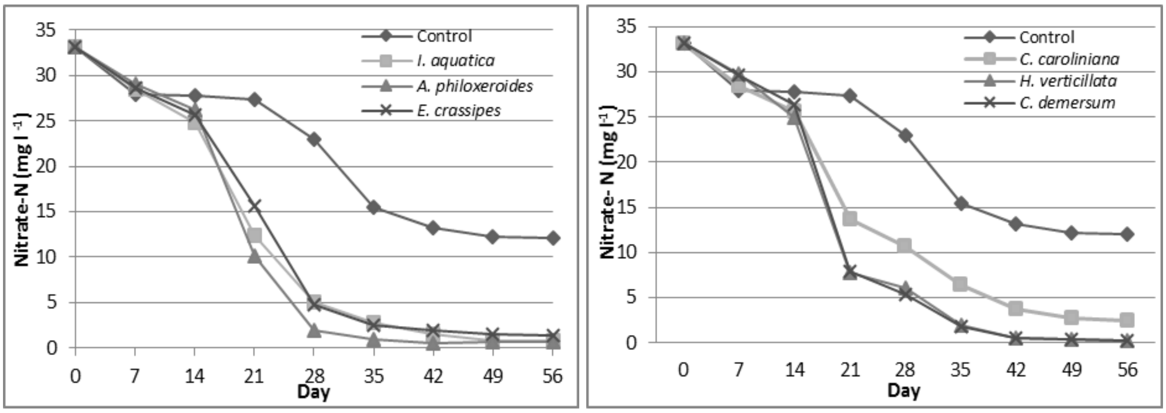


Fig 1: Nitrite-N and Nitrate-N concentration variation within the experimental period

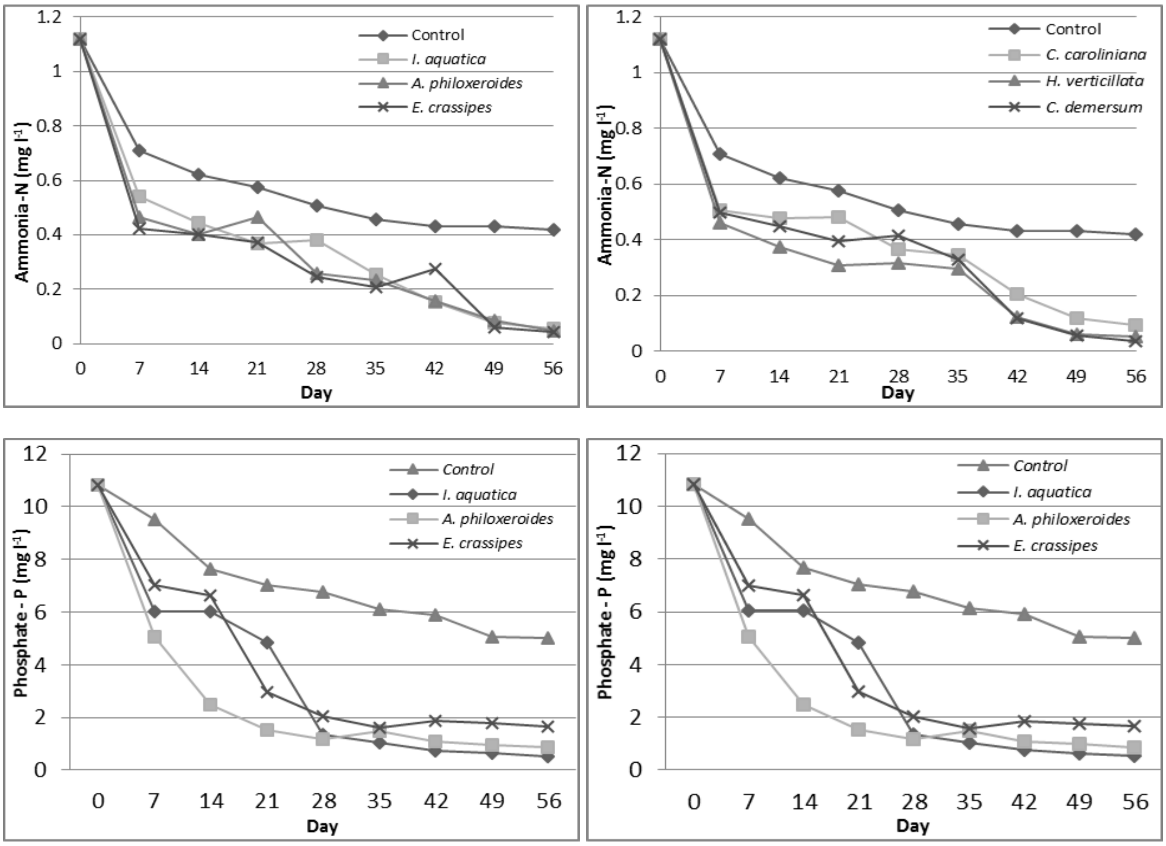


Fig 2: Ammonia-N and Phosphate-P concentration variation within the experimental period

Considering the nitrate-N removal percentage, *A. philoxeroides* showed the highest percentage during the first 28^{th} days and it was also significantly different from the other treatments as well as the control. The high amount of nitrite-N removal percentage at the 28^{th} day was observed in all treatments, and it was not significantly different from treatments other than *C. caroliniana*. Treatment T1 and T5 showed more than 60% of nitrite-N removal after the first 14 days. During the first 28^{th} days, T2 and T3 showed 78% of ammonia-N removal and it was higher than the removal percentage of submerged aquatic macrophytes. The high performance of phosphate-P removal showed by floating aquatic macrophyte T1 and T2, as well as by submerged macrophyte *C. demersum* (T6) during the first 28 days period (Fig. 3).

The removal rate of the nitrite-N, nitrate-N, ammonia-N and phosphate-P nutrients were higher in the first 14 days in all the treatments. Nitrite-N, nitrate-N and ammonia-N nutrient removal rate gradually decreased with the experimental period. The highest nitrite-N, nitrate-N and ammonia-N removal rates were observed in T1 and T5 in the first 14 days. The highest phosphate-P removal rate was observed in T6 followed by T5, T2, T1, T4 and T3 during first 14 days and a rapid reduction was observed after the first 14 days (Fig. 4).

Table 1: Nutrient content in water at the end of 28th and 56th days of different aquatic macrophyte

Nutrient	Control	*I. aquatica*	*A. philoxeroides*	*E. crassipes*	*C. caroliniana*	*H. verticillata*	*C. demersum*
Nitrite-N (mg L^{-1})							
0th day	0.95	0.95	0.95	0.95	0.95	0.95	0.95
28th day	0.55±0.02^{b}	0.046±0.01^{a}	0.065±0.005^{a}	0.083±0.008^{a}	0.16±0.071^{a}	0.044±0.004^{a}	0.09±0.005^{a}
56th day	0.32±0.01^{c}	0.009±0.003^{ab}	0.018±0.001^{ab}	0.009±0.001^{ab}	0.03±0.004^{b}	0.0063±0.001^{a}	0.0056±0.0003^{a}
Nitrate-N (mg L^{-1})							
0th day	33.15	33.15	33.15	33.15	33.15	33.15	33.15
28th day	22.94±0.54^{d}	5.06±0.31^{b}	2.02±0.08^{a}	4.79±0.30^{b}	10.72±0.36^{c}	6.09±0.30^{b}	5.33±0.59^{b}
56th day	12.05±1.02^{d}	0.77±0.06^{ab}	0.68±0.03^{ab}	1.39±0.06^{ab}	2.50±0.25^{b}	0.26±0.01^{a}	0.25±0.02^{a}
Ammonia-N (mg L^{-1})							
0th day	1.12	1.12	1.12	1.12	1.12	1.12	1.12
28th day	0.50±0.06^{c}	0.38±0.002^{abc}	0.25±0.002^{a}	0.24±0.03^{a}	0.36±0.002^{abc}	0.31±0.003^{ab}	0.41±0.036^{bc}
56th day	0.42±0.03^{b}	0.056±0.0031^{a}	0.047±0.002^{a}	0.045±0.002^{a}	0.0093±0.002^{a}	0.053±0.001^{a}	0.036±0.001^{a}
Phosphate-P (mg L^{-1})							
0th day	10.84	10.84	10.84	10.84	10.84	10.84	10.84
28th day	6.75±0.02^{g}	1.35±0.02^{c}	1.18±0.01^{b}	2.03±0.017^{e}	5.17±0.035^{f}	1.62±0.01^{d}	0.85±0.04^{a}
56th day	5.03±0.01^{e}	0.53±0.01^{a}	0.87±0.036^{b}	1.68±0.04^{c}	1.97±0.03^{d}	0.52±0.52^{a}	0.42±0.01^{a}

Values with same superscript did not show any significant difference (*p*>0.05)

Table 2: Water quality parameters at the end of 28 days of different aquatic macrophyte

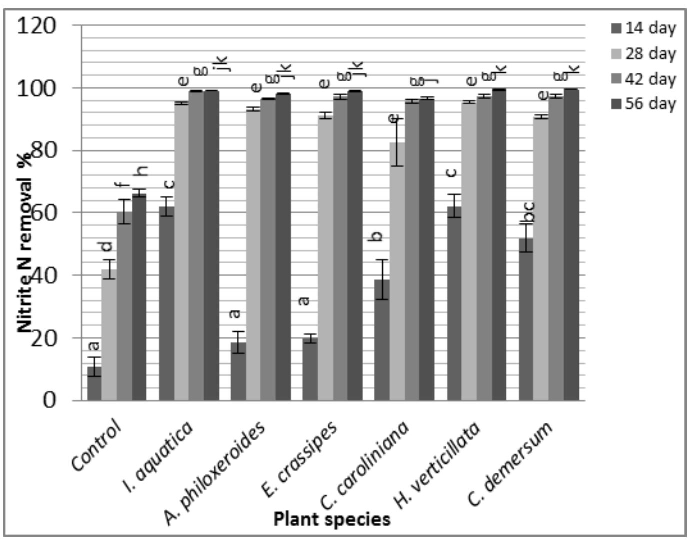
Parameter	Initial	Control	*I. aquatica*	*A. philoxeroides*	*E. crassipes*	*C.caroliniana*	*H. verticillata*	*C. demersum*
pH	8.71	8.20±0.10^{a}	7.60±0.11^{a}	8.15±0.07^{a}	8.01±0.12^{a}	8.11±0.12^{a}	7.80±0.20^{a}	8.10±0.17^{a}
Temperature (^{0}C)	27.20	27.04±0.08^{b}	25.03±0.03^{a}	25.06±0.03^{a}	24.73±0.36^{a}	24.73±0.36^{a}	24.40±0.35^{a}	24.73±0.36^{a}
DO (mg L^{-1})	1.60	2.26±0.14^{a}	3.5±0.03^{b}	2.56±0.06^{a}	3.56±0.12^{b}	4.56±0.03^{c}	6.16±0.08^{d}	6.43±0.06^{d}
BOD (mg L^{-1})	92.00	56.33±1.85^{b}	27.33±1.45^{a}	28.33±0.88^{a}	35.66±3.40^{a}	57.00±1.52^{b}	32.66±1.45^{a}	34.00±2.30^{a}
COD (mg L^{-1})	412.00	302.66±3.71^{d}	247.33±1.85^{c}	242.00±3.60^{c}	123.66±2.72^{a}	250.33±1.45^{c}	288.66±4.91^{d}	203.00±9.07^{b}
Total hardness (mg L^{-1})	134.00	125.66±2.33^{a}	111.00±8.88^{a}	119.33±2.60^{a}	122.33±1.54^{a}	123.00±1.73^{a}	110.66±5.36^{a}	115±1.20^{a}
Alkalinity (mg L^{-1})	192.00	148.66±5.78^{c}	120±2.08^{a}	123.66±2.90^{ab}	139.33±1.66^{bc}	145.66±2.07^{c}	134.00±2.88^{abc}	118.33±4.25^{b}
Turbidity (NTU)	62.00	47.77±1.92^{b}	11.36±0.54^{a}	13.28±0.57^{a}	16.30±0.57^{a}	12.90±1.42^{a}	13.30±0.51^{a}	12.51±0.73^{a}
TS (mg L^{-1})	1.25	0.78±0.41^{c}	0.54±0.02^{ab}	0.49±0.02^{ab}	0.48±0.06^{ab}	0.65±0.028^{bc}	0.58±0.03^{ab}	0.45±0.02^{a}

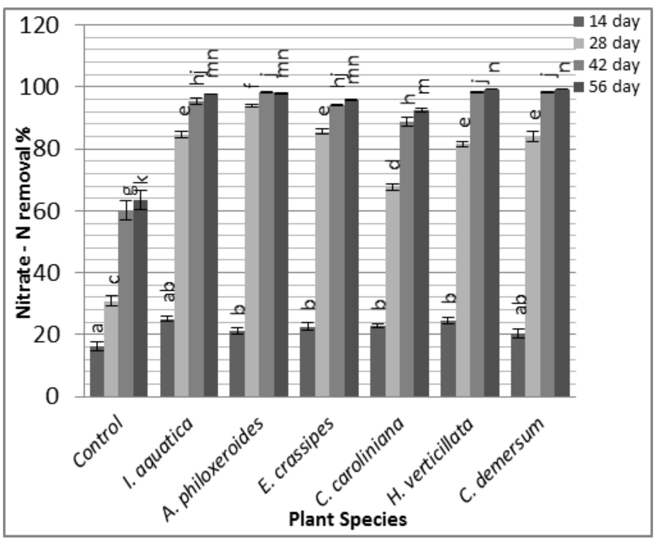
Values with same superscript did not show any significant difference (*p*>0.05)

Table 3. Water quality parameters at the end of 56 days in different aquatic macrophyte

Parameter	Initial	Control	*I. aquatica*	*A. philoxeroides*	*E. crassipes*	*C. caroliniana*	*H. verticillata*	*C. demersum*
pH	8.71	8.00±0.05^{c}	7.82±0.03^{bc}	7.40±0.05^{a}	7.66±0.08^{abc}	8.00±0.06^{c}	7.64±0.07^{ab}	7.52±0.10^{ab}
Temperature (^{0}C)	27.20	26.10±0.15^{b}	25.9±0.20^{b}	25.50±0.29^{ab}	25.00±0.05^{a}	25.03±0.08^{a}	25.06±0.03^{a}	25.03±0.08^{a}
DO (mg L^{-1})	1.60	3.20±0.11^{a}	5.10±0.05^{b}	5.30±0.05bc	5.66±0.06^{c}	6.63±0.08^{d}	7.06±0.06^{e}	9.03±0.08^{f}
BOD (mg L^{-1})	92.00	38.00±1.52^{d}	3.76±0.39^{ab}	3.60±0.20^{ab}	6.50±0.28^{b}	10.66±0.66^{c}	3.24±0.37^{ab}	2.89±0.26^{a}
COD (mg L^{-1})	412.00	139.33±6.35^{d}	70.0±2.88^{b}	68.00±1.52^{b}	68.66±2.4^{b}	89.66±3.17^{c}	65.00±2.88^{b}	44.66±2.60^{a}
Total hardness (mg L^{-1})	134.00	120.00±1.15^{b}	107.66±7.83^{ab}	112.33±2.96^{ab}	112.33±3.71^{ab}	110.00±2.64^{ab}	104.33±4.48^{ab}	97.00±1.52^{a}
Alkalinity (mg L^{-1})	192.00	135.33±4.84^{c}	86.00±1.52^{a}	93.66±2.33^{b}	99.33±0.88^{ab}	107.66±4.48^{b}	92.33±2.02^{a}	87.00±2.08^{a}
Turbidity (NTU)	62.00	23.8±1.94^{b}	1.24±0.10^{a}	1.24±0.10^{a}	1.55±0.10^{a}	2.27±4.48^{a}	1.16±0.049^{a}	0.72±0.095^{a}
TS (mg L^{-1})	1.25	0.52±0.05^{c}	0.40±0.04^{abc}	0.24±0.02^{a}	0.43±0.03^{bc}	0.45±0.04b^{c}	0.28±0.02^{ab}	0.27±0.03^{ab}

Values with same superscript did not show any significant difference (*p*>0.05)





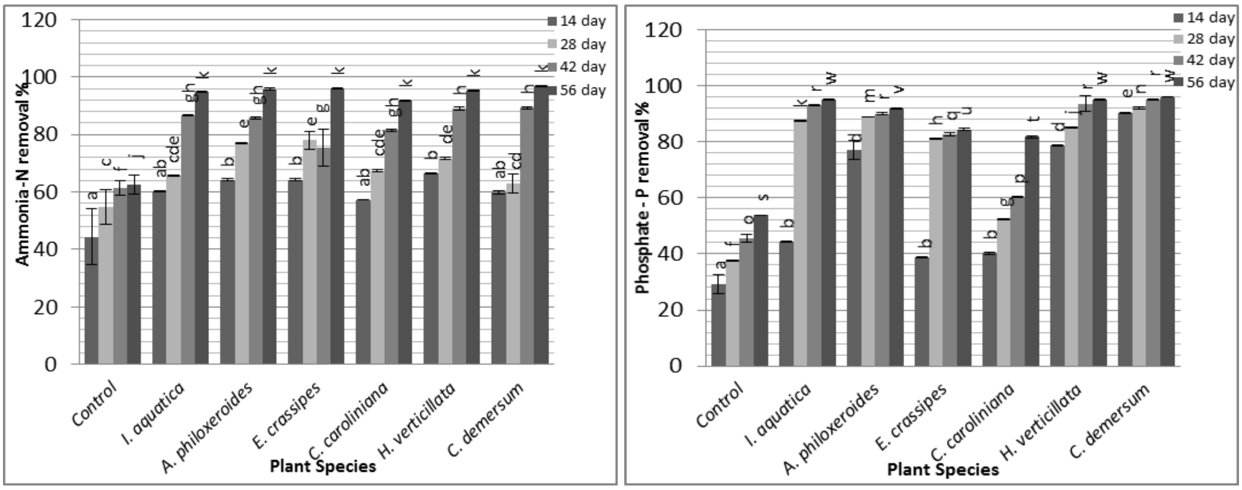


Fig. 3 Different nutrient removal percentage by various plant species at 14th, 28th, 42nd and 56th day of the experiment (Values with same superscript did not show any significant difference (p>0.05))

Considering the plant biomass, a gradual increasing was observed during the experimental time period in submerged aquatic macrophytes other than the *C. caroliniana*. But, in floating aquatic macrophytes, a gradual increase was observed up to the 28th day of the experiment (Fig. 5). Other water quality parameters such as BOD, COD, total hardness, alkalinity, TS showed a gradual decrease with the experimental time period. Dissolved oxygen increased with the experimental time period. This happened in all treatments as well as the control; but, the value of increasing and decreasing of the different parameters was varying depending on the various treatments. According to the result showed at the end of the 28th day, *I. aquatica* showed the lowest turbidity and BOD levels. The lowest COD level was present at *E. crassipes* treatment. The lowest TS and alkalinity levels and the highest DO were present in *C. demersum* (Table 2). At the end of the experiment, lowest values of TS, turbidity, total hardness, COD and BOD levels as well, as the highest, DO level were seen in *C. demersum* treatment (Table 3).

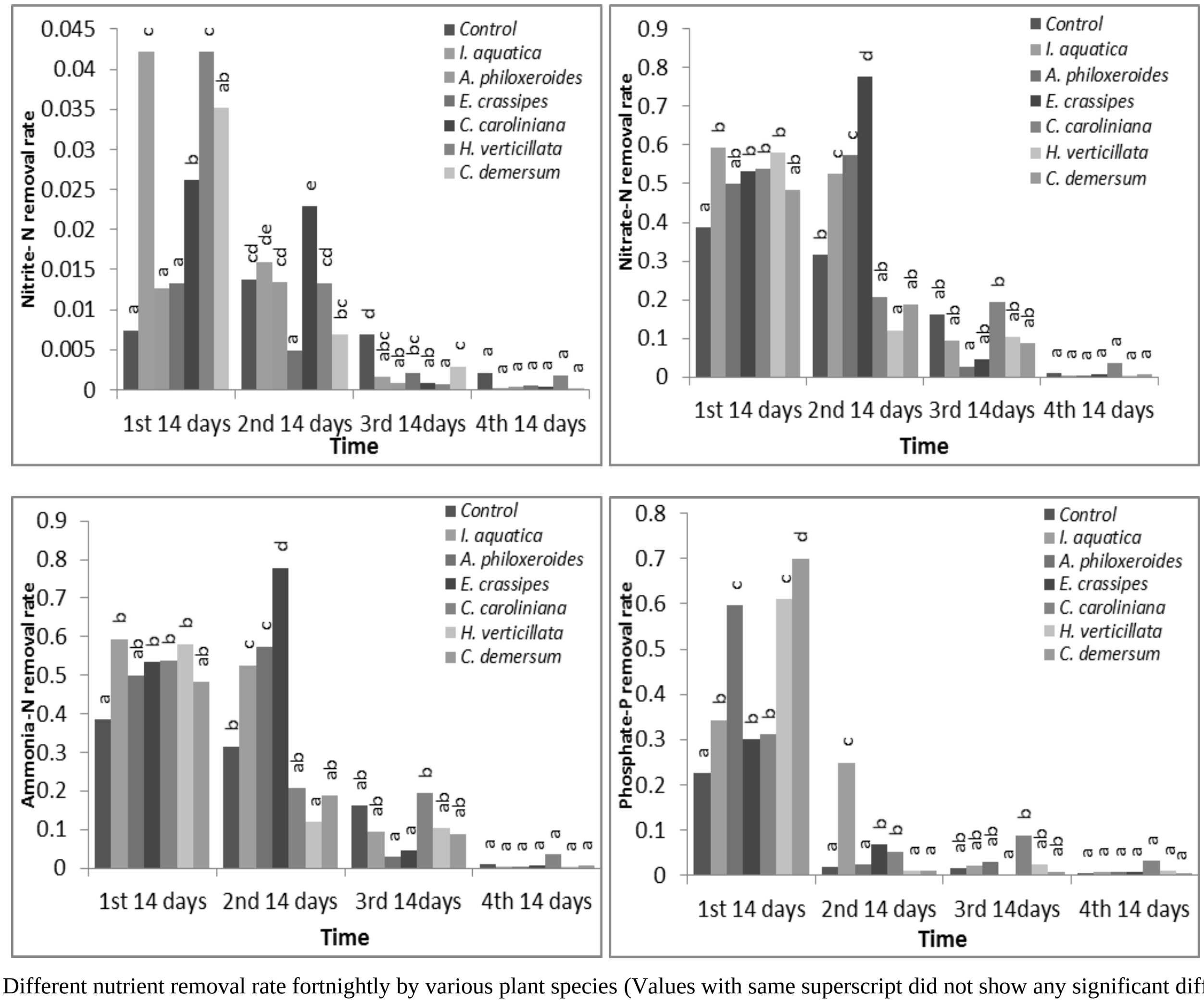
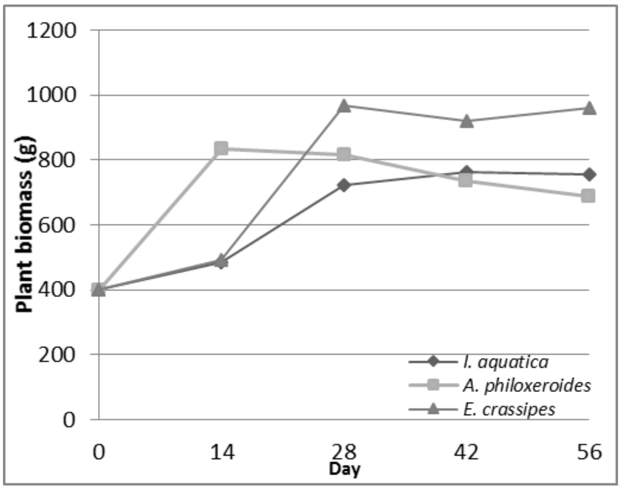


Fig. 4 Different nutrient removal rate fortnightly by various plant species (Values with same superscript did not show any significant difference (p>0.05)



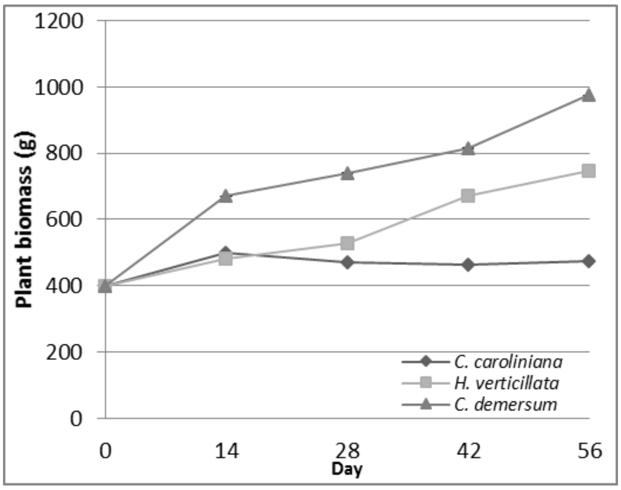


Fig 5: Plant biomass variation within the experimental period

Discussion

Considering the nutrient dynamics during the first 28 days period, floating aquatic macrophytes showed fewer nitrate-N remain in the system compared to the submerged aquatic macrophytes. Rapid plant biomass increasing was observed during the first 28 days of the experiment in floating aquatic macrophytes. *E. crassipes* showed the lowest ammonia-N concentration and the lowest chemical oxygen demand (COD) levels at the end of the first 28 days. Phytoremediation by water hyacinth in sewage and dairy wastewater for a period of 27 days was observed [11]. Results showed that water hyacinth was effective in the treatment of both sewage and dairy wastewater which was significant in reducing dissolved solids, suspended solids, turbidity, COD and BOD reduction were observed, as well as, 50% of nutrient absorption was observed. Also revealed that [6], effective removal of turbidity in dairy wastewater by *E. crassipes* and *Lemna*. Short term experiment was conducted, [12] which treated eutrophic water by *I. aquatica,* after a 48 h period, COD, BOD and TSS in the effluent were reduced by 84.5, 88.5 and 91.1 respectively, and the removal of nutrients (total nitrogen and total phosphorus) varied between 41.5 and 75.5%. In the present study, *I. aquatica* showed considerable nutrient removal during the first 28 days period.

The results [13] revealed an increase in value of pH, DO and percentage oxygen saturation value; while, the other parameters such as turbidity, salinity, electrical conductivity, total dissolved solids, alkalinity, free carbon dioxide, chloride, COD, total hardness, calcium hardness, calcium, magnesium, nitrogen and phosphorus as phosphate recorded a significant decrease in values due to absorption of nutrients during phytoremediation of domestic wastewater by *C. demersum* L. Comparative results were also obtained in the present experiment. Lowest values of TS, turbidity, total hardness, COD and BOD levels, as well as the highest, DO level were observed in *C. demersum* treatment. An increase in biomass of *C. demersum* L. during the culture was indicated by an increase in net primary productivity with the average value of 1.58 g m^{-2} day^{-1} [13]. In the present experiment, a significant increase of biomass was observed in *C. demersum* at the end of the experiment. The Efficiency of *H. verticillata* for absorption of N, P, and K and prevention of eutrophication of storm water was observed [14]. Water quality in experimental ponds with *H. verticillata* was improved by phytoremediation by decreasing turbidity, total solids, and nutrient concentrations. Considering the present experiment, significant nutrient removal was observed by *H. verticillata* among the submerged macrophytes, which was only second to the *Ceratophyllum*. Srivastava *et al*. [15] found that *H. verticillata* and *C. demersum* were one of the best effective combinations for phytoremediation. The present experiment also revealed that best and most effective plants were *C. demersum and H. verticillata* for the phytoremediation among the other aquatic macrophytes.

Conclusion

Floating aquatic macrophytes *I. aquatica, A. philoxeroides* and *E. crassipes* can be suggested as effective plants species for phytoremediation in aquaculture effluents when considering short time period, with the high nutrients removing capability from the aquaculture effluents within the 28 days.

Considering the quantitative reduction of nutrients as well as reduction of most water quality parameters to desirable range in aquaculture effluent and increase in plant biomass during the experimental period, *C. demersum* and *H. verticillata* can be suggested as best aquatic macrophytes among the selected macrophytes for effective phytoremediation in aquaculture effluents.

Acknowledgments

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