IOP Conference Series: Earth and Environmental Science

PAPER • OPEN ACCESS

Improving Wastewater Quality By Using Ceratophyllum Demersum L.

To cite this article: Enas A. Mahdi Al-Nabhan and Dunya A. H. Al-Abbawy 2021 *IOP Conf. Ser.: Earth Environ. Sci.* **910** 012086

Improving Wastewater Quality By Using Ceratophyllum Demersum L.

Enas A. Mahdi Al-Nabhan[1] and Dunya A. H. Al-Abbawy[2]

[1,2]University of Basrah, College of Science, Department of Ecology, Iraq.

[2]Email: dunya.hussain@uobasrah.edu.iq

Abstract

The current study aims to improve the quality of wastewater collected from the secondary sedimentation tank of Hamdan sewage water treatment plant, Basrah, Iraq, using *C. demersum* L. for phytoremediation potential in the laboratory experiment. The experiments were also designed to establish some toxicological effects of wastewater on plant physiology as a response to wastewater stresses. The selected physio-chemical parameters of wastewater were conducted. Plant analyses were also measured before and after 21st day of exposure. The results showed the efficiency of the plant in improving the water quality in a different ratio. The dilution 1:3 (T3) was the most efficient with a significant difference ($p<0.05$) between treatment and control for most of the measured parameters. The result showed the efficiency of *Ceratophyllum demersum* in removing the Total hardness, Ca+2, Mg+2, BOD5, COD, NO3-2, PO4-2, Na+ and Cl- in 1:3 dilution (55.88 , 13.27, 89.55, 90, 47.37, 50.65, 48.51, 16.28 and 40.24)% compared with (17.65, 5.42, 27.31, 50, 15.79, 16.30, 13.86, 6.73 and 15.77)% in control respectively. The *Ceratophyllum demersum* plant has proven to be effective in improving the quality of wastewater, and therefore it can be considered an effective candidate in phytoremediation technology.

KeywordS: Phytoremediation, Wastewater, Heavy metals, Toxicological effect.

1. Introduction

Numerous organic and inorganic contaminants have been released into the environment as a result of developing agriculture, industrial activity, and global population expansion. [1-3]. Moreover, wastewater discharge into water bodies has a significant adverse effect on water quality and creates substantial environmental and health problems [4,5]. Since such conventional methods for cleaning up polluted soils and waters be effective, they are generally very costly and labor-intensive [6]. As a result, researchers have searched for an alternative, environmentally friendly. Low-cost approaches that do not generate secondary waste do not require repair and maintenance and do not necessitate specialized labor and the use of sunlight as a source of energy [7]. This technology is phytoremediation, plants, and their associated microbes to eliminate pollutants from soil, water, and sediments. Many aquatic plants are used to remediate wastewater like *Ceratophyllum demersum*. This species is rootless; submerged dicotyledon's seed belonging to the family Ceratophyllaceae grows well in the subtropical and tropical weather regimes [8,9]. Different studies were done on the efficiency of some aquatic plants to remediate wastewater. [10] studied the removal efficiency of organic load and some nutrients from sewage by [11], studied water hyacinth (*Eichhornia crassipes*) for biotreatment of textile wastewater. [12] examined the ability of *Hydrilla verticillata* for improving domestic wastewater in an artificial culture system for one year. *Hydrilla verticillata*, success in reducing pH, Ec, TSS, BOD5, COD, PO_4, NO_3, NO_2, Ammonia, hardness, calcium, and magnesium confirmed the removal of Nickel using *C. demersum* exposing to different concentrations of industrial wastewater in two-week. [13] noted the bioaccumulation susceptibility and tolerance of *C. demersum* to different lead concentrations in a 21-day laboratory experiment. The results showed increased lead accumulation with increased metal concentrations. The current study aims to assess the ability of *C. demersum* to remove various pollutants from wastewater and minimize wastewater toxicity through phytoremediation technology. As well as investigate some toxicological effects of wastewater on coontail.

2. Materials and Methods

2.1. Wastewater sample collection

Water samples were taken from the secondary sedimentation tank of the Hamdan Central sewage Wastewater Treatment Plant in Basrah Province, Iraq. This sewage wastewater treatment plants containing preliminary treatments as bar screen and primary treatments as primary sedimentation tank to settle the solid particles and finally the secondary sedimentation tank before discharge to surface water. The collected sample was placed in clean, labelled polyethene bottles of 20-litre capacity and transported to the laboratory for analysis.

2.2. Plant Samples and analysis

The labelled plastic bags were used to store aquatic plants. *Ceratophyllum demersum L.* collected from the Hammar Marshland, southern Iraq. It was taken to the laboratory, washed several times with tap water, and distilled water to remove debris. For a week of acclimatization in the laboratory with 18.2 C° of water temperature used and 23.2 C° of air laboratory temperature, the plant was placed in a container filled with 20 litres of tap water. After acclimatization was done, a healthy plant of the same weight was selected for the experiment. Each container provided 25g of the plant. Specific plant parameters were measured before and after the experiments to improve the biochemical changes that occurred. These parameters are fresh weight, relative growth, total chlorophyll, protein content, and a tolerance index.

2.3. Fresh weight

At the end of the experiment, the plant samples were taken from all treatments to measure the changes in fresh weight, washed with tap water and distilled water to remove and debris. The samples were oven-dried at 70 C until constant weight in gram.

2.4. Total chlorophyll content

The total chlorophyll was calculated according to [14]. About 0.2 grams of the plant's fresh weight were taken. Extracted with 20 ml of 80% acetone, centrifuged at 5000 cycles per minute for 5 minutes, the filtrate part was obtained, and the absorbance was recorded with a spectrophotometer at two wavelengths 645 - 663 nm. The formula was used to calculate the total chlorophyll value in milligrams per gram.

$$\text{Total Chlorophyll mg/g} = (12.7 \times \text{OD } 663) + (16.8 \times \text{OD } 645)$$

OD: optical Density on 645 and 663 nm

2.5. Protein content

The protein in the plant was measured according to [15], as a percentage by calculating the total nitrogen in it and multiplying it by a factor of 6.25

2.6. Experimental Setup:

The experiment was carried out in a controlled laboratory condition. A total of 21st days of the investigation were designed to examine the ability of *Ceratophyllum demersum* rhizofilteration to enhance water quality. Even to check at the changes in the plant's morphological and physiological properties as a response to various pollutants. Plastic containers used each container is filled with a two-litter of wastewater as named below:

T1: Raw wastewater treatment without dilution + plant
T2: Diluted (1:1): (one volume wastewater + one volume distilled water) + plant
T3: Diluted (1:3): (one volume wastewater + 3 volumes distilled water) + plant
CT1: Raw wastewater treatment without plant
CT2: Control diluted (1:1) wastewater without plant
CT3: Control diluted (1:3) wastewater without plant

The wastewater height of each container was labeled. After one week of acclimatization, 25 grams of a healthy plant was used for each container. The plant's changes were noticed. To substitute for wastewater losses due to evaporation, distilled water was added to each basin to the remarked previously.

2.7. Removal Efficiency of pollutants

The removal efficiency (%) of pollutants by *Ceratophyllum demersum* was measured according to the equation below [16, 17].

$$\text{Removal efficiency} = \frac{C1-C2}{C1} \text{ X } 100$$

Where C1: initial concentration of parameter before Phytoremediation

C2 is the concentration of the parameter after phytoremediation

2.8. Relative growth rate

The relative growth of the plant was calculated at the end of the experiment, and it was compared with the initial plant weight. The relative growth rate was calculated as an equation [18].

$$\text{Relative growth rate} = \frac{\text{final plant weight (g)}}{\text{Intial plant weight (g)}}$$

2.9. Tolerance index rate

According to the formula mentioned in [19], the tolerance index ratio was calculated, and the result was expressed as a percentage.

$$\text{Tolerance Index Rate} = \frac{Dry\ weight\ of\ the\ plant\ in.Treatment\ g}{Dry\ weight\ of\ plant\ in\ Control\ treatment.\ g} \times 100$$

2.10. Statistical analysis

SPSS model 23 statistical analysis program was used at 0.0.5 significance level. Applying descriptive statistics of results and analyze one-way variance (ANOVA) between wastewater experiment treatments (T1, T2, and T3) and control treatments (CT1, CT2, and CT3) for water and plant parameters.

3. Results

3.1. Reduction of pollutants and removal efficiency calculation

Before starting the experiment, the wastewater samples were analyzed to determine their characteristics in raw and diluted (T2, T3), As described below:

Table 1. Wastewater Characteristics at the beginning of experiments.

Parameter	Raw wastewater T1	Dilution (1:1) T2	Dilution (1:3) T3
pH	6.9± 0.1	7.2± 0.05	7.5± 0.10
Temp.(°C)	17.88± 0.05	17.56± 0.32	18.2± 0.02
EC(mS/cm)	5.20± 0.01	2.55± 0.10	1.77± 0.11
TDS (mg/l)	3392.1± 2.05	1632± 2.0	1132.8± 2.0
Cl^- (mg/l)	1999.23± 0.84	999.24± 1.06	499± 0.99
NO_3^{-2}(mg/l)	24.03± 0.95	14.9± 0.95	9.30± 0.17
PO_4^{-3} (mg/l)	3.88± 0.01	1.92± 0.01	1.01± 0.01
BOD_5 (mg/l)	40 ± 0.26	20± 1.0	10 ± 1.02
COD (mg/l)	134.03 ± 1.13	76± 2.00	38 ± 2.01
Total hardness (mg/l)	1350 ± 0.20	650.1± 0.1	370.02± 0.02
Ca^{+2} (mg/l)	240.48 ± 0.02	124.22± 0.02	60.12± 0.10
Mg^{+2} (mg/l)	182.12± 0.06	82.45± 0.11	46.12± 0.02
Na^+ (mg/l)	1292.5± 0.20	727.50± 2.02	465.83± 0.98

After the 21st day of the plant's exposure, the results demonstrated the plant's capacity to reduce the values of selected parameters in T1, T2, and T3. Figures (1-3) describe the rate of change in pH, temperature, and electrical conductivity. In all treatments, water temperature, pH, and EC were increased. However, the increasing pH values significantly at 0.05 ($p \leq 0.05$) in wastewater (T1, T2, and T3) and control treatments (CT1, CT2, and CT3) were 8.1, 8.1, 8.3, and 7.6, 7.5, 7.7 respectively. However, the increase in water temperature with no significant difference at 0.05 ($p \geq 0.05$) .variation in EC were 5.54, 2.68, 1.88, 5.38, 2.61, and 1.80 mS/cm in T1, T2, T3, CT1, CT2, and CT3, respectively. Fig1, 2, and 3.

The result also indicated a significant reduction in the BOD5 and COD, chloride ion, nitrate salts, phosphate, sodium, total hardness, calcium, and magnesium in the experiment and control treatments at a level of 0.05.Fig (4-10).

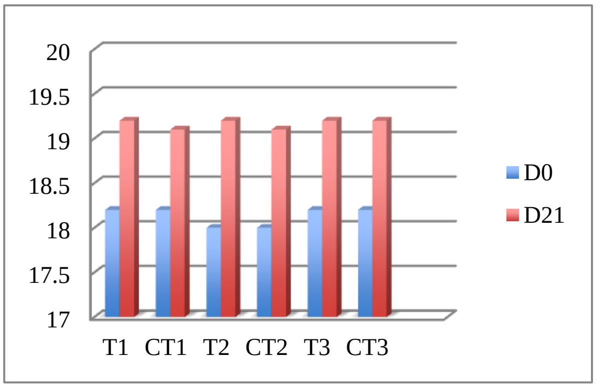

Figure 1. The average water temperature at the beginning and end of the experiment.

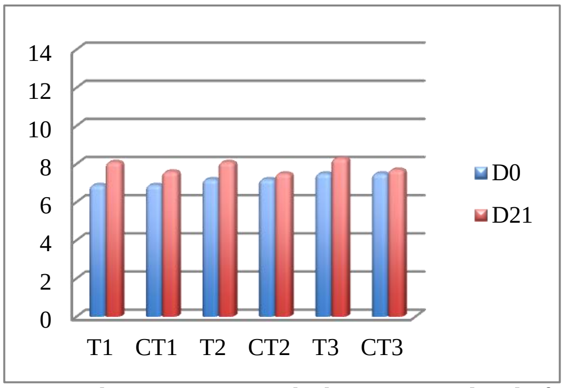

Figure 2. The average pH at the beginning and end of the experiment.

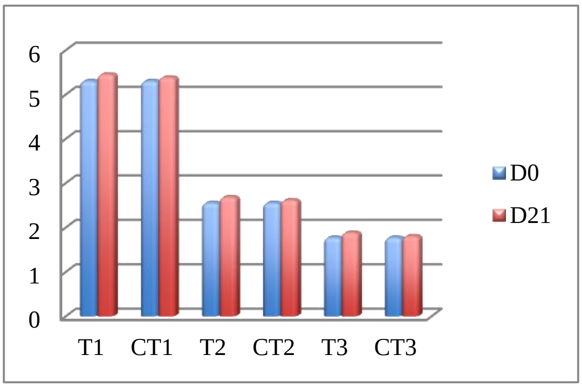

Figure 3. The average Electrical Conductivity at the beginning and end of the experiment.

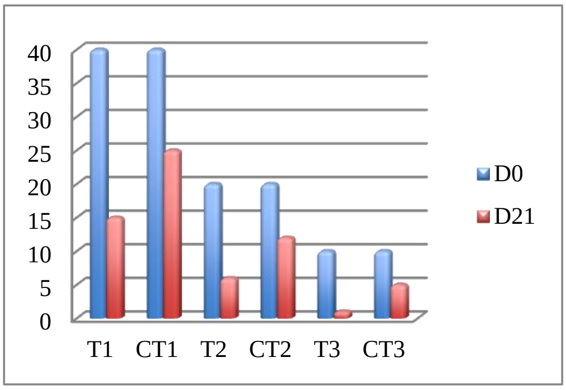

Figure 4. The average BOD_5 at the beginning and end of the experiment.

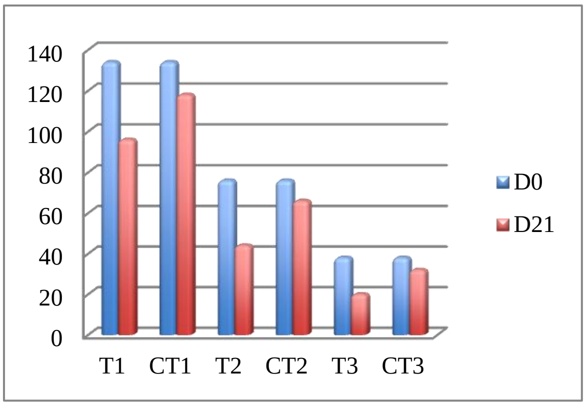

Figure 5. The average COD at the beginning and end of the experiment.

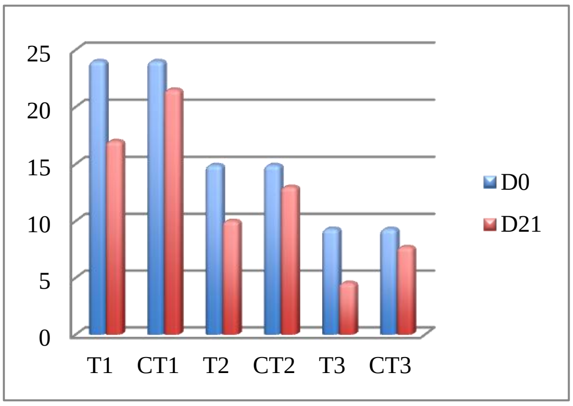

Figure 6. The average nitrite (NO3) at the beginning and end of the experiment.

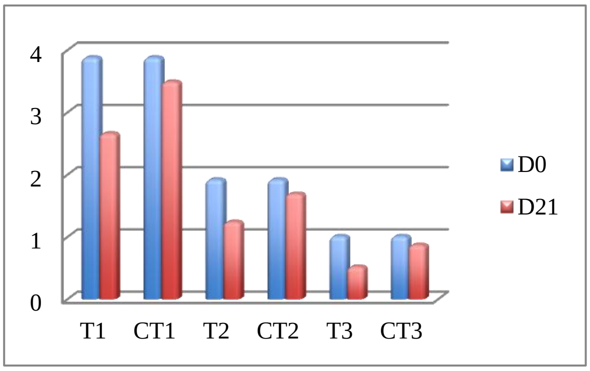

Figure 7. The average PO_4 at the beginning and end of the experiment.

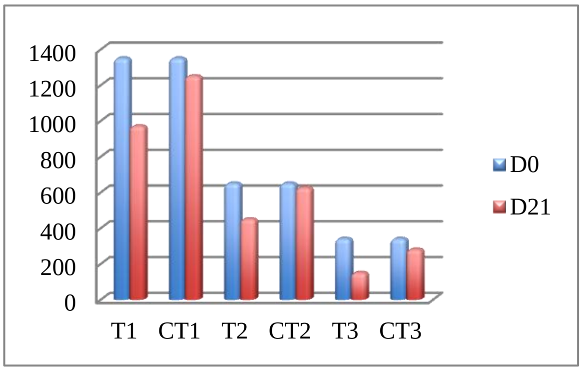

Figure 8. The average total hardness at the beginning and end of the experiment.

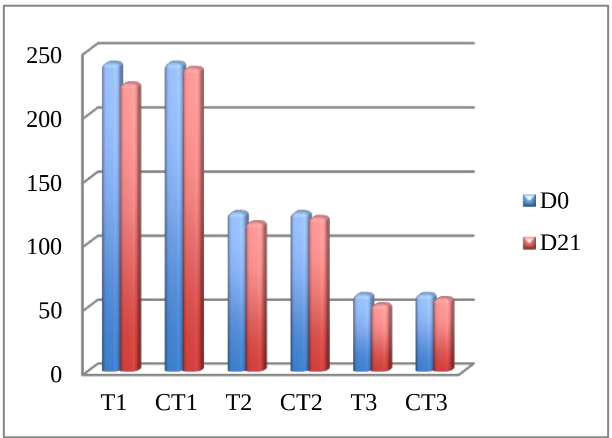

Figure 9. The average calcium ion (Ca^{+2}) at the beginning and end of the experiment.

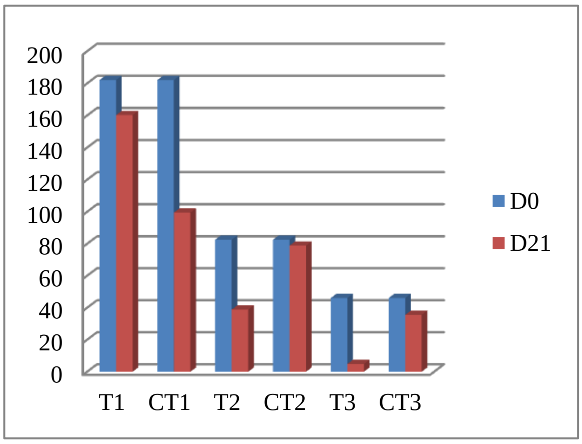

Figure 10. The average Magnesium (Mg) at the beginning and end of the experiment.

Table 2. The removal efficiency of pollutants for all treatments after 21st of exposure.

Parameter	Removal Efficiency %					
	T1	CT1	T2	CT2	T3	CT3
BOD_5 mg/l	62.5± 2.50 a	37.5± 2.25 b	70 ± 5.0 a	40± 5.0 b	90±10.0 a	50± 10.0 b
COD mg/l	28.35± 0.74 a	11.94±2.50 b	42.10± 1.32 a	13.16± 1.31 b	47.37± 2.63 a	15.79± 2.64 b
NO_3^{-2}mg/l	32.01± 0,40 a	13.88± 0.01 b	37.31± 1.01 a	19.55± 0.77 b	50.65± 0.22 a	16.30± 1.08 b
PO_4^{-3}mg/l	31.44± 2.83 a	9.79± 0.25 b	35.75± 0.52 a	14.40± 0.51 b	48.51± 2.97 a	13.86± 0.99 b
T. hardness mg/l	28.14± 0.07 a	7.40± 0.15 b	30.75± 0.01 a	10.11± 0.36 b	55.88± 0.59 a	17.65± 0.59 b
Ca^{+2} mg/l	6.68± 0.04 a	1.66± 0.045 b	6.30± 0.12 a	3.13± 0.01 b	13.27± 0.16 a	5.43± 0.58 b
Mg^{+2} mg/l	45.35± 0.005 a	12.01± 0.005 b	53.48± 0.49 a	16.98± 0.48 b	89.55± 0.04 a	27.31± 0.3 b
Cl^- mg/l	15.92± 0.05 a	6.96± 0.1 b	21.45± 0.1 a	19.93 0.005 b	40.24± 0.02 a	15.77 ± 0.04 b
Na^+ mg/l	9.93±0.001 a	3.35± 0.07 b	13.34± 0.13 a	5.22± 0.27 b	16.28 ± 0.21 a	8.73 ± 0.22 b

*different letter (for each column for each treatment and control alone) refer to significant difference at 0.05 ($p \leq 0.05$)

3.2. Toxicological effect on plant

The initial fresh weight was 25gram and, the initial total chlorophyll was 7.876 mg/ gram, and the Initial protein was 32.56 %. The plant analysis revealed significant differences between treatments at the 0.05 Level ($p \leq 0.05$), as shown in Table 3 and the T3 recorded decreasing in toxicological effects compared to T1 and T2 after 21th of exposure.

Table 3. Toxicological effect of wastewater on plants before and after 21st of exposure.

Parameter	Initial plant analysis	T1	T2	T3
Fresh weight	25.032 gram	11.33± 0.04 c	14.81± 0.06 b	18.91± 0.08 a
Relative growth	-	0.45± 0.01 c	0.59±0.09 b	0.75±0.02 a
T. chlorophyll	7.876 mg/gram	6.73± 0.03 c	7.05± 0.013 b	7.47± 0.06 a
Protein %		20.68± 0.25 c	25.70± 0.06 b	30.37± 0.05 a
Tolerance index rate	-	44.67± 0.38 c	59.75± 0.37 b	76.82± 0.06 a

*different letter (for each column for each treatment) refer to significant difference at 0.05 ($p \leq 0.05$)

4.Discussion

4.1. Reduction of pollutants and removal efficiency

Since it is a controlled laboratory condition, the temperature values were not significantly elevated. This may be to the experiment carried out in control conditions in the laboratory. The pH value increased in all treatments due to the plant's photosynthetic activity. Because the plant consumes dissolved CO2 gas, its absorption increases the pH, rising towards alkalinity [16]. The results showed that the value of pH between 6.9-8.3 supports the growth of the coontail plant. The electrical conductivity slightly increased. Due to the conductivity, the growth of *Ceratophyllum demersum* was good, especially in diluted treatment, due to decreasing conductivity, which is an indicator of water quality. Its presence is evaluated by the volume of salts present in the water. BOD and COD removal in the plant treatments (T1, T2, and T3) was significantly higher than in the control treatments (CT1, CT2, and CT3), as shown in table 2. The anaerobic condition favors the aerobic bacterial activity to reduce the BOD and COD. These aerobic conditions are due to increasing photosynthetic activity, which realizing oxygen in the wastewater and depletion of CO2. Furthermore, aerobic oxidation of organic matter in wastewater, the sedimentation process [11, 20], and the filtration of suspended solids contribute to the enhanced reduction of BOD and COD in planted system treatments [21, 22]. These results agreed with [23- 25].

At the end of the experiments, the decline in NO_3 in plant treatments was more significant than the reduction in control treatments (Figure 6), suggesting that the reduction can be due to plant uptake. Many experiments have improved the knowledge that N removal results from the nitrification/denitrification activity of root-associated bacteria [26]. In contrast, the reduction in PO_4 in plant system treatment may be due to the sorption, complexation, precipitation, biotic assimilation, and plant uptake [27, 28] (Figure 7). This result agreed with [29]. At the end of the experiment, the total hardness, calcium, and magnesium ion declined in all plant treatments and control. This decrease can be due to the total hardness value dependent on Ca, Mg, K, Cl, and SO4, and the plant absorbing this salt for metabolic activity.` This result agreed with [28]. Several aquatic plant species absorb sodium directly from the water on their leaves. Abscission of leaves is a typical result when a plant consumes more sodium than it can tolerate by this pathway [30]. Chloride can be absorbed by plants from their roots and stored in their leaves. Toxic effects can arise depending on the plant's susceptibility to chloride as this occurs. These signs are identical to those experienced when sodium levels rise: scorching of leaf abscission. Chloride foliar absorption is also expected. It can result in the abscission of leaves in sensitive plants.

4.2.Toxicological effect on Ceratophyllum demersum

The pH increase was caused by the photosynthesis activity of plants, which took dissolved CO_2 from water and released oxygen. Thus pH value increased as a result of the CO_2 reduction [22]. The plant's results are reflected that the decrease in total chlorophyll content in raw treatment (T1) (without dilution) is more than in other treatments. This reduction may be due to the high level of salinity, which negatively affects the plant's enzymes activity for causing inhibition of chlorophyll synthesis. This result agreed with [28]. The result also improved the reduction of protein content without significant between treatments. This reduction may be due to high salinity, which harms the cell by affecting amino acid content such as nitrogen and reducing DNA and RNA in plant tissue. And the reduction in protein content may also increase the activity of protaseae responsible for protein degradation.

The result also indicated that the effect on tolerance index in raw treatment is more than in other treatments. This reduction may be due to the negative impact of the dry weight of plants and relative growth due to high salinity and chloride content.

Conclusion

As a result, the *Ceratophyllum demersum* is a good phytoremediater for removing pollutants and improving the wastewater characteristics. More dilution of wastewater with distilled water in the ratio (1:3) improves remediate pollutants than the raw wastewater due to their features.

Acknowledgment

The research work has been done with the collaboration of the Directory of wastewater in Basra Province and supported by the Department of Ecology laboratories.

References

[1] Lalevic, B., Raicevic, V., Kikovic, D., JOVANOVIC, L., SURLAN, M. G., Jovic, J., ... & Morina, F. (2012). Biodegradation of MTBE by bacteria isolated from oil hydrocarbons-contaminated environments.
[2] Gandhi, N., Sirisha, D., & Sekhar, K. C. (2013). Phytoremediation of chromium and fluoride in industrial wastewater by using aquatic plant ipomoea. Science of the Total Environment, 12, 1-4.
[3] Gavrilescu, M., Demnerová, K., Aamand, J., Agathos, S., & Fava, F. (2015). Emerging pollutants in the environment: present and future challenges in biomonitoring, ecological risks, and bioremediation. New biotechnology, 32(1), 147-156.
[4] USEPA, J.(1998). Guidance for data quality assessment: Practical methods for data analysis.
[5] Barceló, D., Ginebreda Martí, A., Acuña, V., Batalla, R. J., Elosegi, A., Guasch, H., & Sabater, S. , (2011). Combined scenarios of chemical and ecological quality under water scarcity in Mediterranean rivers.
[6] Zhang, X. B., Peng, L. I. U., Yang, Y. S., & Chen, W. R. (2007). Phytoremediation of urban wastewater by model wetlands with ornamental hydrophytes. Journal of Environmental Sciences, 19(8), 902-909.
[7] Mustafa, H. M., & Hayder, G. (2020). Recent studies on applications of aquatic weed plants in phytoremediation of wastewater: A review article. Ain Shams Engineering Journal.
[8] Chorom, M., Parnian, A., & Jaafarzadeh, N. (2012). Nickel removal by the aquatic plant (Ceratophyllum demersum L.). International Journal of Environmental Science and Development, 3(4), 372.
[9] DiTomaso, J. M., Kyser, G. B., Oneto, S. R., Wilson, R. G., Orloff, S. B., Anderson, L. W., & Mann, J. J. (2013). Weed control in natural areas in the western United States. Weed Research and Information Center, University of California, 544.
[10] Kulasekaran, A., Gopal, A., & John Alexander, J. (2014). A study on the removal efficiency of organic load and some nutrients from sewage by Ceratophyllum demersum. L J. Mater. Environ. Sci, 5(3), 859-864.
[11] Mahmood Q., Ping, Z., Siddiqi M. R., Islam E. M. Rashid and Hayat, Y. (2005). Lab Scale Studies on Water Hyacinth (Eichhornia crassipes Marts Solms) for Biotreatment of Textile Wastewater. Caspian J. Env. Sci. 3 (.2):: 83~88.
[12] Mustafa, H. M., & Hayder, G. (2021). Recent studies on applications of aquatic weed plants in phytoremediation of wastewater: A review article. Ain Shams Engineering Journal, 12(1), 355-365.
[13] Chen, M., Zhang, L. L., Li, J., He, X. J., & Cai, J. C. (2015). Bioaccumulation and tolerance characteristics of a submerged plant (Ceratophyllum demersum L.) exposed to toxic metal lead. Ecotoxicology and Environmental Safety, 122, 313-321.
[14] Oron, G., de-Vegt, A., & Porath, D. (1988). Nitrogen removal and conversion by duckweed grown on wastewater. Water Research, 22(2), 179-184.
[15] Page, A. L., Miller, R. H. and Keeney, D. R. (1982). Methods of Soil Analysis, part 2.chemical and microbiological properties, 2.
[16] Kumar, S., & Deswal, S. (2020). Phytoremediation capabilities of Salvinia molesta, water hyacinth, water lettuce, and duckweed to reduce phosphorus in rice mill wastewater. International journal of phytoremediation, 22(11), 1097-1109.
[17] Kooh, M. R. R., Lim, L. B., Lim, L. H., & Malik, O. A. (2018). Phytoextraction potential of water fern (Azolla pinnata) in the removal of a hazardous dye, methyl violet 2B: artificial neural network modeling. International journal of phytoremediation, 20(5), 424-431.
[18] Lu, X., Kruatrachue, M., Pokethitiyook, P., & Homyok, K. (2004). Removal of cadmium and zinc by water hyacinth, Eichhornia crassipes. Science Asia, 30(93), 103.
[19] Wilkins, D. A. (1978). The measurement of tolerance to edaphic factors by means of root growth. New Phytologist, 80(3), 623-633.
[20] Shah, M., Hashmi HN. Ali, A.& Ghumman, A.R. (2014).Performance assessment of aquatic macrophytes for treatment of municipal wastewater.J.Environ. Heal. Sci. Eng. 12:106.
[21] Xiang Shi, Haijing Sun, Hongwei Pan, Yitai Chen, Zeping Jiang, Jianfeng Liu & Shufeng Wang., (2015). Growth and efficiency of nutrient removal by Salix jiangsuensis J172 for phytoremediation of urban wastewater. Environ Sci Pollut Res 23, 2715–2723
[22] Rana, V.&Maiti, S.K., (2018). Municipal wastewater treatment potential and metal accumulation strategies of Colocasia esculenta (L.) Schott and Typha latifolia L. in a constructed wetland. Enviro. Monit. Assess.190:328.
[23] Oron, G., de-Vegt, A., & Porath, D. (1988). Nitrogen removal and conversion by duckweed grown on wastewater. Water Research, 22(2), 179-184.

[24] Korner, S., Lyatuu, G. B., & Vermaat, J. E. , (1998). The influence of Lemna gibba L. on the degradation of organic material in duckweed-covered domestic wastewater. Water Research, 32(10), 3092-3098.
[25] Azeez, N. M., & Sabbar, A. A. (2012). Efficiency of duckweed (Lemna minor L.) in phytotreatment of wastewater pollutants from Basrah oil refinery. Journal of Applied Phytotechnology in Environmental Sanitation, 1(4), 163-172.
[26] Vaillan, N. , Monnet, F., Vernay, V., Sallanon, H., Coudret, A. &Hitmi, A.(2002).Urban wastewater treatment by a nutrient film technique system with a valuable commercial plant species (Chrysanthemum cinerariaefolium Trev.).Environ. Sci. Technol.,36:2101-2106.
[27] Zhang, X. B., Peng, L. I. U., Yang, Y. S., & Chen, W. R. (2007). Phytoremediation of urban wastewater by model wetlands with ornamental hydrophytes. Journal of Environmental Sciences, 19(8), 902-909.
[28] Bindu, T., Sylas, V.P., Mahesh, M., Rakesh, P.S.&Ramasamy, E.V. (2008). Pollutant removal from domestic wastewater with Taro (Colocasia esculenta) planted in a subsurface flow system. Ecol. Eng., 33:68-82.
[29] Loveson, A., & Sivalingam, R. (2013). Phytotoxicological assessment of two backwater wetlands in Kannamaly, Ernakulam using aquatic macrophyte Spirodela polyrhiza. Journal of Environmental & Analytical Toxicology, 3(4), 180.
[30] Faroughi, Maryam. (2011). Role of Ceratophyllum demersum in recycling macro elements from wastewater. J. Appl. Sci. Environ. Manage., 15 (2):401 – 405
[31] Al-Abbawy, D. A., Al-Sweid , Z.& Al-Saady, S. A., (2020). Effects of salinity stress on biochemical and anatomical characteristics of Ceratophyllum demersum L. Eurasia J Biosci 14:5219-5225.