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ProEnvironment

ProEnvironment 13 (2020) 60 - 70

Original Article

Capacity of Micro and Macrophytes to Remediate Wastewater for Irrigation

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Received 16 May 2020; received and revised form 27 May 2020; accepted 15 June 2020
Available online 30 June 2020

Abstract

Phytotechnology was applied to assess the capacity of microphytes (algae) and macrophytes (duckweed) to remediate wastewater from the Tanjaro River in order to meet irrigation standards. The results showed clear differences between the initial and treated wastewater. Physiochemical measurements include: temperature, pH, dissolved oxygen, biological oxygen demand, total dissolved salts, electrical conductivity, turbidity and chlorophyll *a*, NO_3^-, PO_4^{3-}, SO_4^{2-}, HCO_3^-, CO_3^{2-}, Cl^-, Ca^{2+}, Mg^{2+}, Na^+, K^+, Fe, Co, Cd, Pb, Cr, Cu, Zn and Mn. Wastewater treated by algae and duckweed showed declines in each variable, except for dissolved oxygen and chlorophyll *a*. Nutrient removing efficiencies of the algae and duckweed indicated the ability to remove 100% of the Fe, Cd, Pb, Cr and As in the wastewater. Algae showed a higher efficiency to remove Co, Cu, Zn and Mn in comparison with duckweed; duckweed showed adequate efficiency to remove PO_4^{3-}, Cl^-, HCO_3^-, CO_3^{2-}. The calculated value of irrigation water quality index (IWQI), 11, of the initial wastewater (low suitability for irrigation) improved after treatment with algae and duckweed to 14 and 15, respectively. Thus, irrigation water quality index (IWQI) improved 36.4 and 27.3% over initial values by duckweed and algae, respectively. These findings indicate the potential of phytotechnology to be applied in environmental remediation in an agriculturally important region in Iraq.

Keywords: macrophytes (duckweed), microphytes (algae), phytotechnology, Tanjaro River, wastewater.

1. Introduction

Land application of wastewater (sludge and excreta) is widespread around the world with countries, using sludge as fertilizer or directly using wastewater for irrigation [14, 44]. Nearly 7% of the total irrigated land in world uses polluted water or wastewater without treatment for irrigation [49] amounting to nearly 20 million ha globally [47]. According to world health organization [48], the benefit of using wastewater for irrigation depends on the quality of wastewater.

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Phytotechnology is the use of living photosynthetic organisms to mitigate environmental pollutants [16]. Phytotechnology includes the use of macrophytes and microphytes, the byproducts of which can be used in compost fertilizer and biofuel production [20, 24].

The microphytes (algae) represent various groups of organisms, which have the capability to grow under different conditions. Algae can grow in both conditions of high and low temperature, pH, and salt concentration [42].

Remediation of wastewater has been documented for the past 40 years [11]. Algae has the capability to remove both organic and inorganic pollutants, especially macronutrients and heavy metals in the wastewater. They also have the ability to destroy organic compounds in the polluted water through bio-transformational processes [23, 43].

The growth rate of the algae depends on several conditions such as chemical (nutrient concentration, carbon dioxide) physical (light for photosynthesis processes, temperature and humidity), and biological (virus infections and competition between species) [37].

In addition to microphytes, green algae and blue green algae, aquatic macrophytes (e.g., duckweed) can also be used to improve the water quality and remediation of wastewater. Macrophytes accumulate heavy metals and other toxic nutrients, and most of them can aerate the water providing oxic or suboxic conditions. They also have the ability to grow quickly and are easy to collect and cultivate [41]. Duckweed species have been used to recover nutrients in wastewater for the past 30 years [10, 30]. Furthermore, because duckweed is a source of protein and starch for animal feed, it has also been used in the application of bioethanol and compost [30].

In addition, macrophytes have potential to naturally purify water, converting wastewater and sewage into pure water and edible duckweed with little resulting sludge. Moreover, several environmental factors such as light intensity, salinity, temperature, pH, nutrient, competition with other plants and toxins in the water have been shown to influence the distribution and growth rate of duckweed, macrophytes *Lemnaceae* species, respectively [13].

Plants (macrophytes) irrigated by wastewater have been shown to accumulate heavy metals in root and shoot parts as well as bringing that wastewater closer to irrigation water quality standards [18, 27]. Although these studies showed the potential of plant bioremediation of wastewater before mixing with Tanjaro River water, it did not emphasize the bioremediation capacity of microphytes (algae) nor compare it with macrophytes (duckweed). Thus, our study aims to use algae and duckweed in the phytoremediation of Tanjaro River water. Algae and duckweed grow naturally in the Tanjaro River especially in the summer season under high sewer discharge from the Sulaymaniyah city.

The current research study was conducted to test the ability of phytotechnology application - specifically, micro/macrophyte, to remediate wastewater in order to meet international irrigation standards. In addition, the ultimate goals of this research were to: contribute to reversing the degradation of natural water and soil resources in Kurdistan Region Governorate (KRG); improve the sustainability of water resources; strengthen water resource management, enhance environmental quality, and expand and improve the health and well-being of KRG citizens through effective science-based wastewater treatment. Our study also aims to contribute to better managing, remediating, and reusing of Tanjaro River wastewater as a source for irrigation.

2. Material and Method

2.1. Study area and sampling location

The study focused on the Tanjaro River in Sulaymaniyah Governorate, Kurdistan, Iraq. The Tanjaro River starts between the Azmar and Baranan Mountains and runs near the NW to SE border of Sulaymaniyah City towards Darbandikhan Lake. The River passes through the Tanjaro valley crossing many urban and agricultural regions with a catchment area of 1167.3 km², a length of 66.7 km, and an average slope of 11% (Fig.1) [31].

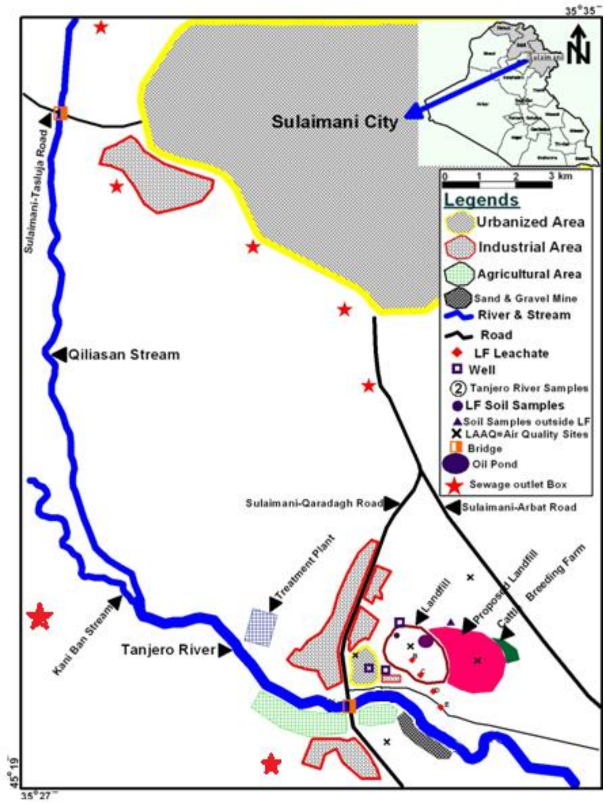

Figure 1. Details of area surrounding the Tanjaro River [36]

Darbandikhan Lake water discharges to the Diyala River one of the major tributaries of the Tigris River. The City of Sulaymaniyah supplies the Tanjaro River with approximately 265,000 m^3 of wastewater daily and mixes with sources from the Dokan Dam (112,000 $m^3\ d^{-1}$) and Sarchnar spring(48,000 $m^3\ d^{-1}$).The domestic water consumption per capita including water losses is 0.42 $m^3\ d^{-1}$ nearly, all of which is converted into sewage. This sewage is eventually combined with rainwater and discharged through sewer pipelines into the Tanjaro River [36]. The collection of water sample from Tanajaro River is showed in the Fig. 2.

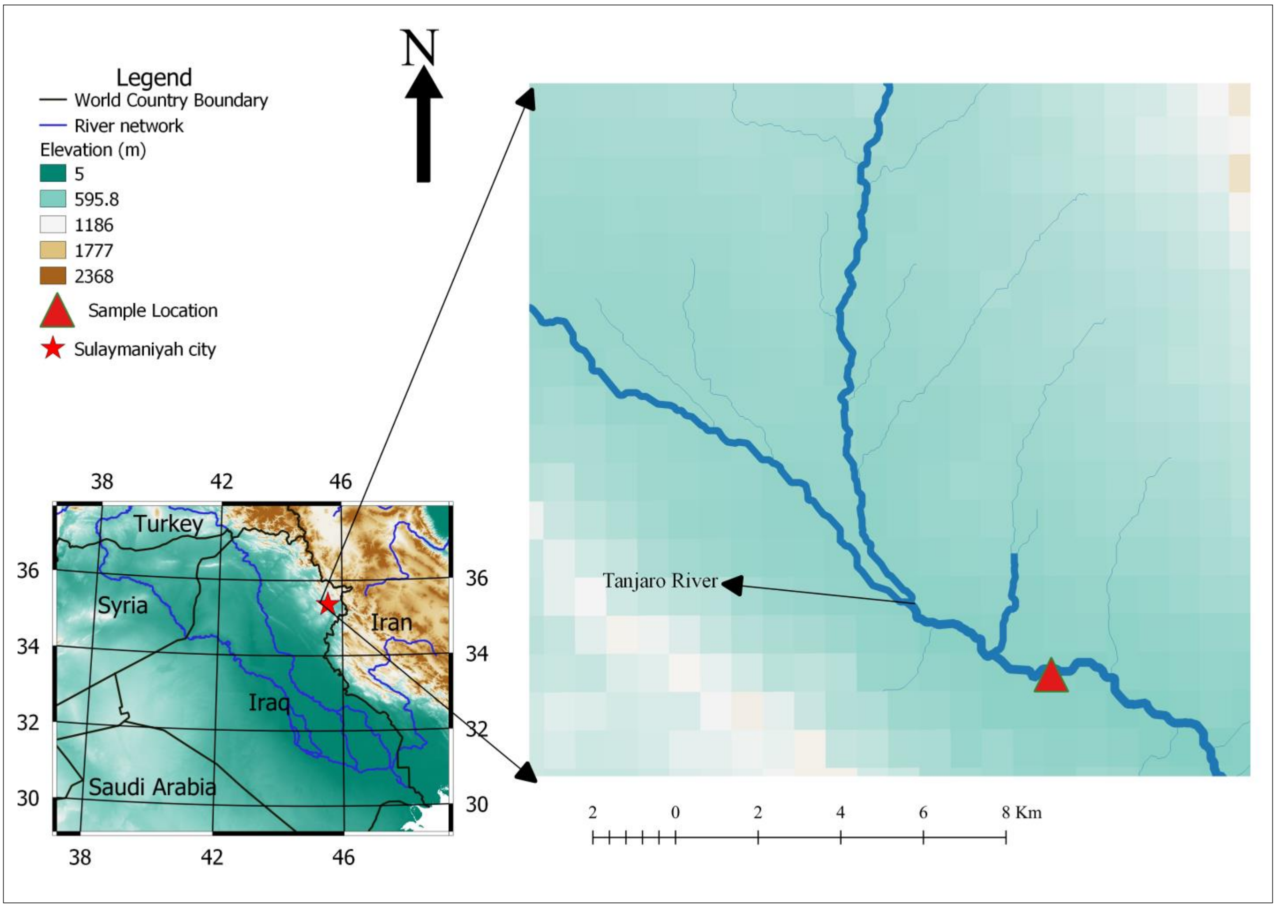

Figure 2. Tanjaro River in Sulaymaniyah city. Sampling location of water samples is shown in the red triangle prepared by ArcGIS, 10.8.

2.2. Experimental Design

The study was setup as a completely randomized design repeated measures experiment with three replicates, in October 2018. Statistical analyses was conducted using the R 3.2.3 software [34] to compare the efficiency of the microphytes, green algae (Chlorphyta) and blue-green algae (Cyanobacteria), and macrophyte, duckweed (*lemmna Gibba*). Six glass basins (50 × 35 × 50 cm) were used to grow the algae and duckweed in triplicate during the experiment.

These basins were filled with 50 L of wastewater from the Tanjaro River. Light and temperature were semi-controlled in the green house. Algae and duckweed were harvested 3 times (0, 5, 10 and 15 days) over the course of the experiment in order to measure and evaluate the effect of harvesting frequency on wastewater remediation [29].

2.3. Wastewater laboratory measurements

Water laboratory measurements consisted of the following physiochemical measurements: pH, total dissolved solids (TDS), dissolved oxygen (DO), biological oxygen demand (BOD), temperature (°C), turbidity (NTU), NO_3^-, PO_4^{3-}, SO_4^{2-}, HCO_3^-, CO_3^{2-}, Cl^-, Ca^{2+}, Mg^{2+}, Na^+, K^+, Fe, Co, Cd, Pb, Cr, Cu, Zn, and Mn. The laboratory measurements were conducted in the College of Agricultural Engineering Science in the Department of Natural Resources and at the Kurdistan Institution for Strategic Studies and Scientific Research following APHA (1998).

Temperature and pH were measured using a portable pH meter HANNA HI 8314 as described by APHA (1998). Electrical conductivity (EC) and total dissolved solids (TDS) were measured at the 25°C using an EC meter (WTW, Multi197i) at the time of sampling following APHA (1998) [3]. Turbidity was measured with a turbidity meter (WTW, pHoto Flex/photo Flex Turb 430) after calibration with turbidity standards; the results were expressed in terms of nephelometric turbidity unit (NTU) following APHA (1998). Dissolved oxygen was measured at the time of sampling using a special oxygen-sensitive membrane electrode (WTW, Multi197i) and the results were expressed in (mg O_2 L^{-1}) following APHA (1998) [3].The biological oxygen demand was determined from dissolved oxygen decreases in a 5-day incubation at 27°C using an OxiTop Control 6 measurement system (German

Standard Method, 2000)[19]. Carbonate and bicarbonate in the water sample were determined using an acid-base titrimetric method following APHA (1998) [3]. Potassium and sodium were measured by PFP7 flame photometer JENAWAY also following APHA (1998) [3].

Water samples were run by inductively coupled plasma (ICP) spectroscopy for determination of calcium (Ca^{2+}), magnesium (Mg^{2+}), and heavy metals (Fe, Co, Cd, Pb, Cr, Cu, Zn and Mn) using a Perkin- Elmer Optical Emission Spectrometer Optima 2100 DV following APHA (1998). Anions (NO_3^-, PO_4^{3-}, SO_4^{2-} and Cl^-) were determined by ion chromatography (IXS1500 DIONEX) as recommended by APHA (1998) [3].

We calculated the ***irrigation water quality index (IWQI)*** before and after the experiment following Eq. 1 and 2 [4]. The percentage of IWQI improvement was calculated using Eq. 3.

$$\mathrm{Wi} = \frac{w}{N} \sum_{i=1}^{N} \mathrm{Ri} \quad (1)$$

$$\mathrm{IWQ\ Index} = \sum \mathrm{Wi} \quad (2)$$

$$\%\ \mathrm{IWQI\ improvement} = \frac{\mathrm{fIWQI} - \mathrm{iIWQI}}{\mathrm{iIWQI}} \times 100 \quad (3)$$

where:

W - the involvement of each one of the water measurements, w is the weight of the water measurements,
N - the total number parameters, and
R - the rating value.
iIWQI - initial Irrigation Water Quality Index
fIWQI - final Irrigation Water Quality Index.

The water suitability for irrigation was assessed using three classes of IWQI for irrigation; IQWI values less than 19 were specified as having a low suitability for irrigation, between 19 and 32 as medium suitability, and greater than 32 as high suitability for irrigation. Also, the ***metal removal efficiency (MRE)*** was calculated from on Eq. 4:

$$\mathrm{MRE} = \frac{\mathrm{iC} - \mathrm{fC}}{\mathrm{iC}} \times 100 \quad (4)$$

where:

i_C - initial concentration of metals in the water sample from Tanjaro River,
f_C - final concentrations of metal after algae and duckweed treated water.

3. Results and Discussions

3.1. Physiochemical measurements in the wastewater during algae and duckweed treatment

The relationship between pH and harvest time is shown in Fig. 3. Algae showed a positive relationship between pH and harvesting time, duckweed showed no relationship with harvesting time. Duckweed showed a noticeable increase in pH on day 5 of cultivation. In general, a weak relation can be seen in Fig. 3 between pH and harvest time. The reason for this is because CO_2 comes to equilibrium at night from respiration of algae and duckweed producing HCO_3^- stabilizing the pH. These results are consistent with those reported by Cole (2009) [12].

Azov (1982), Bai et al. (2017), and Porath and Pollock (1982) [5, 6, 33] show that pH can increase during microalgae and duckweed cultivation via photosynthetic CO_2 assimilation [37]. There are significant differences between the influence of algae and duckweed on pH increasing during harvest time especially in days 5 and 10; nonetheless, the positively correlation with algae was greater than with duckweed according to the Spearman coefficient, which was equal to 0.56 and 0.01 for algae and duckweed, respectively. The results agree with research performed by Zimmo (2005) who found pH stability in wastewater when treated by algae and duckweed [50].

However, we found negative relationships between TDS and turbidity with harvest time for both algae and duckweed.

Also, the properties of the experiential fits were not significantly different between algae and duckweed for ether TDS or turbidity. Duckweed produces high biomass with an increase in salinity and has the capability to remove and uptake salt in the wastewater [26].

Furthermore, Raiz et al. (2018) [38] showed that decreasing turbidity (especially decreasing colloidal particles) corresponds to salt reduction in wastewater.

We found a positive relationship between turbidity and TDS following remediation by algae. In addition, Ozengin and Elmaci (2007) suggested that the duckweed has the ability to reduce turbidity through absorption of colloidal materials [32].

Figure 3 shows a positive relationship between DO and harvest time for both algae and duckweed. There is a positive relationship between BOD and harvest time for both algae and duckweed although day 10 shows values that are somewhat higher than the trend our predict. Bhat and Hiremath (2003) [8] discussed BOD increasing through organic matter increases in the aquarium; since day 15, the biomass of both algae and duckweed started to decompose, causing a decrease in the nutrient in the aquarium and an increase in microbial activity. Algae and duckweed show a similar response with time for BOD. Negative correlations between DO and BOD were observed (Fig. 4). This is in agreement with Waziri et al. (2010) who found a negative association between BOD and DO [47].

The concentration of chlorophyll *a* in the duckweed increased at the beginning of the experiment with a maximum concentration by the first harvest and decline to near zero by the second harvest.

Chlorophyll *a* in the experiment is an indication of the photosynthetic activity in the aquarium and biomass production of the algae and duckweed. Algae increased until the second harvest and declined significantly by 15 days.

There is a significant difference between algae and duckweed in the chlorophyll *a* concentration during harvesting time, except day 5 when both algae and duckweed recorded the maximum value of chlorophyll *a* concentration.

Concentrations HCO_3^-, PO_4^{3-}, Cl^-, and NO_3^- decreased with time for duckweed and algae, both with significant differences in the rate of that decrease between the microphyte and macrophytes. In addition, Al-Nozaily et al. (2000) and Goopy and Murray (2003) [2, 21] report reductions of NO_3^- in wastewater using algae to remediate water. Zimmo et al. (2005) found that both algae and duckweed was able to remediate NO_3^- in wastewater [50].

The results of HCO_3^- shown are in agreement with El-Kheir et al. (2007) and Jahan et al. (2014) who found that HCO_3^- reduced in water when treated by algae and duckweed [15, 25].

However, there are little differences between the capacity of algae and duckweed to reduce Cl^-. Jahan et al. (2014) and Ramirez et al. (2018) [25, 35] found that algae and duckweed was not able to reduce chloride concentrations in wastewater.

The decrease in PO_4^{3-} concentration with time is likely caused by the uptake by algae and duckweed as an essential nutrient. Agarwal et al. (2019) and Saikumar (2014) [1, 39] discussed the P ion as a source for enzymatic processes; it may be that the decrease in P is due to the adsorption by microalgae or precipitation on colloids and metalloids in the wastewater.

Divergent trends were seen in CO_3^{2-} between algae and duckweed.

Duckweed CO_3^{2-} decreased with time while algae slightly increased with time. However, Sun et al. (2016) investigated root exudation and found that fatty acid in duckweed root systems may affect microbial activity and ability to use CO_3^{2-} as a source of carbon and oxygen [45].

Positive trends were observed between SO_4^{2-} concentration and harvest time for both algae and duckweed. There are significant differences, however, in the rate of increase for algae and duckweed. These results do not show agreement with previous research by Gupta et al. (2015) who found a reduction of SO_4^{2-} [22]. The differences of our finding may be due to decompositions of algae and duckweed cell walls during the experiment that contain sulfate polysaccharides.

Both Ca^{2+} and Mg^{2+} concentrations showed clear relationships with harvest time in opposite directions. Also, Ca^{2+} concentrations were similar between algae and duckweed except for day 10 of harvesting (Fig. 3).

Furthermore, algae and duckweed reduced Ca^{2+} concentrations at a similar rate in the wastewater. As with Ca^{2+}, Mg^{2+} concentrations were similar between algae and duckweed except for day 10 of harvesting.

These results agree with Muradov et al. (2014) who found a decline of Ca^{2+} in wastewater and an increase in the biomass of microphytes and macrophytes that were used in the petrochemical application of their study [30]. The finding that Mg^{2+} increases with time agrees with Krems et al. (2013) [28] who found that Mg^{2+} can be replaced by heavy metal in chlorophyll particles, causing a decrease in chlorophyll *a* and metals with time and an increase in Mg^+ concentration.

We found a negative correlation between Mg with heavy metals and chlorophyll *a* (Fig. 5).

Both algae and duckweed K^+ concentration decreased with time, with little differences between their efficiency to remediate the K^+ in wastewater during the bioremediation experiment (Fig. 3).

The results were similar to El-Kheir et al. (2007) and Muradov et al. (2014) [15, 30] who investigated the capability of algae and duckweed to remove K^+ concentration in wastewater. Iyer et al. (2015) found K^+ concentrations in the algae biomass [24]. Positive relationships between Na^+ and harvest time were observed (Fig. 3) with an increase until day 5 and a subsequent stabilization of Na^+ concentration. El-Kheir et al. (2007) found Na^+ increased in the duckweed pond during day 6 of their experiment [15], and Muradov et al. (2014) discussed the algae and duckweed capacity to reduce Na^+ concentration in the wastewater [30].

Sodium increased until day 5 likely caused by competition between K^+ and Na^+ through up-take mechanisms by microphytes and macrophyte. Castillo et al. (2015) described competition mechanisms between K^+/Na^+ and concluded that K^+ was faster than Na^+. It is likely for this reason that Na declined after day 5 [9].

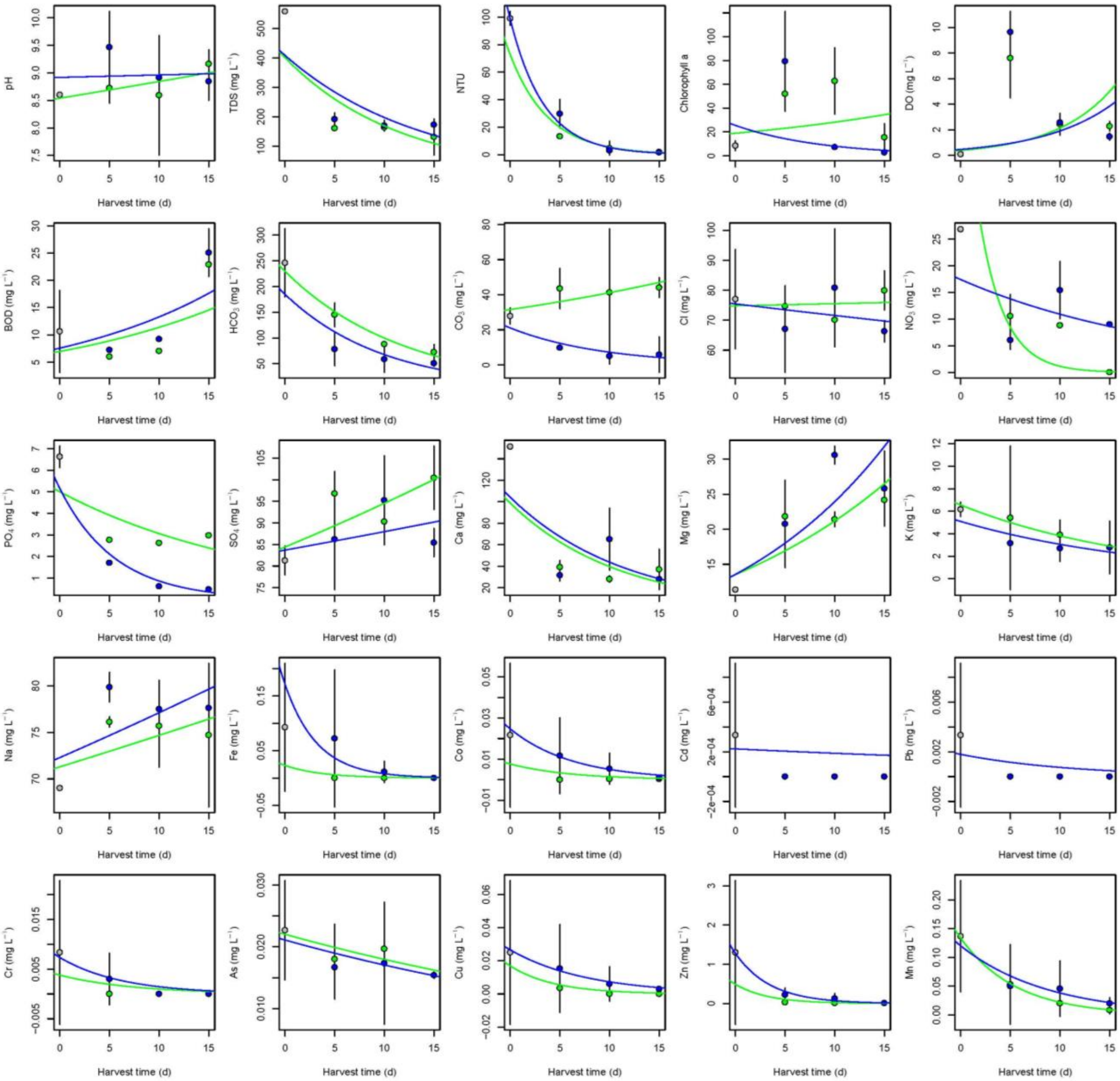

Figure 3. Relationship between physiochemical measurements in wastewater from Tanjaro River during algae and duckweed cultivation with harvesting time (0, 5, 10, 15) per day, by using linear regression

Correlation coefficient - r is calculated using Spearman test. The color illustrates the followings: initial wastewater - gray, algae - green and duckweed - blue

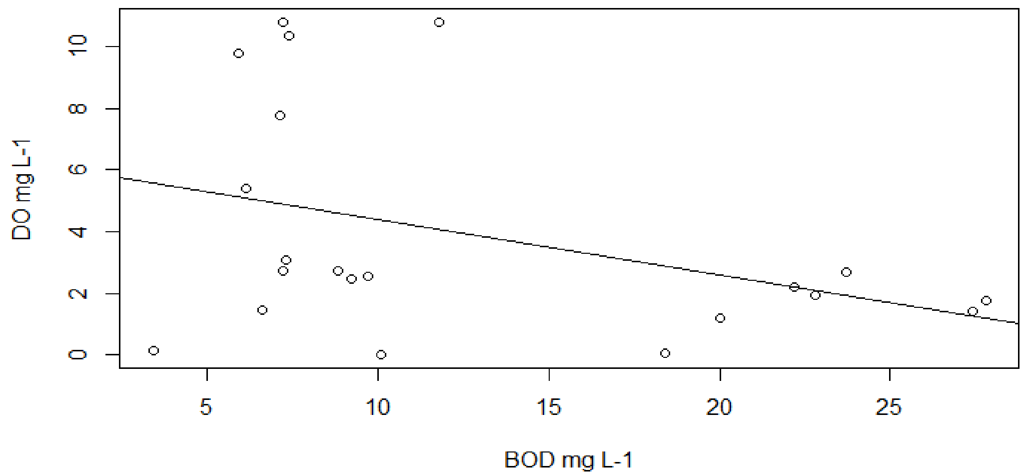

Figure 4. DO regressed against BOD

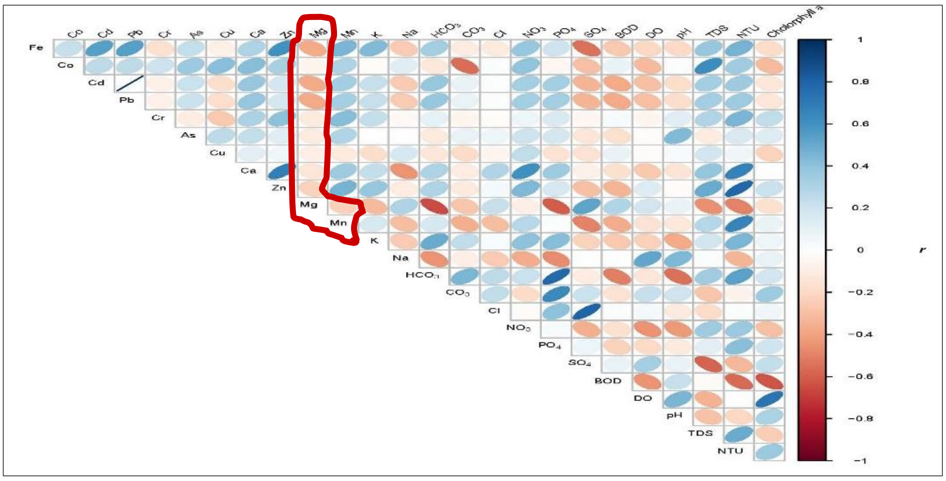

Figure 5. Spearman correlation between ions in the wastewater from the Tanjaro River. The Mg^{2+} correlations are highlighted with the red line

Fig. 3 shows a negative relationship between metal (Fe, Co, Cd, Pb, Cr, As, Cu, Zn, and Mn) concentrations and harvest time. The rate of which metal concentrations decreased were different for algae and duckweed in Fe, Co, Cd, Pb, Cr, As, Cu, Zn, and Mn. Results indicate that increasing algae and duckweed cultivation times strongly affect the reduction of metal concentration in the wastewater from the Tanjaro River. Sekomo et al. (2012) determined algae and duckweed ability to remove wastewater from metals. Krems et al. (2013) showed that metals were included in the metabolic system in both algae and duckweed and were uptaken from the growth medium as a source of micronutrients [27, 40].

3.2. Metals Removal Efficiency (MRE)

The percentages of MRE of algae and duckweed are different with respect to different metals, as shown in Fig. 6. Algae showed efficiencies to remove the Co, Cu, Zn, and Mn in comparison with duckweed.

Algae and duckweed showed similar efficiencies to remove Fe, Cd, Pb, Cr, and As. Agarwal et al. (2019), El-Kheir et al. (2007), and Falabi et al. (2002) concluded that the effect of algae and duckweed on wastewater treatment is the remediation of metal pollution through accumulation in their biomass [1, 15, 17].

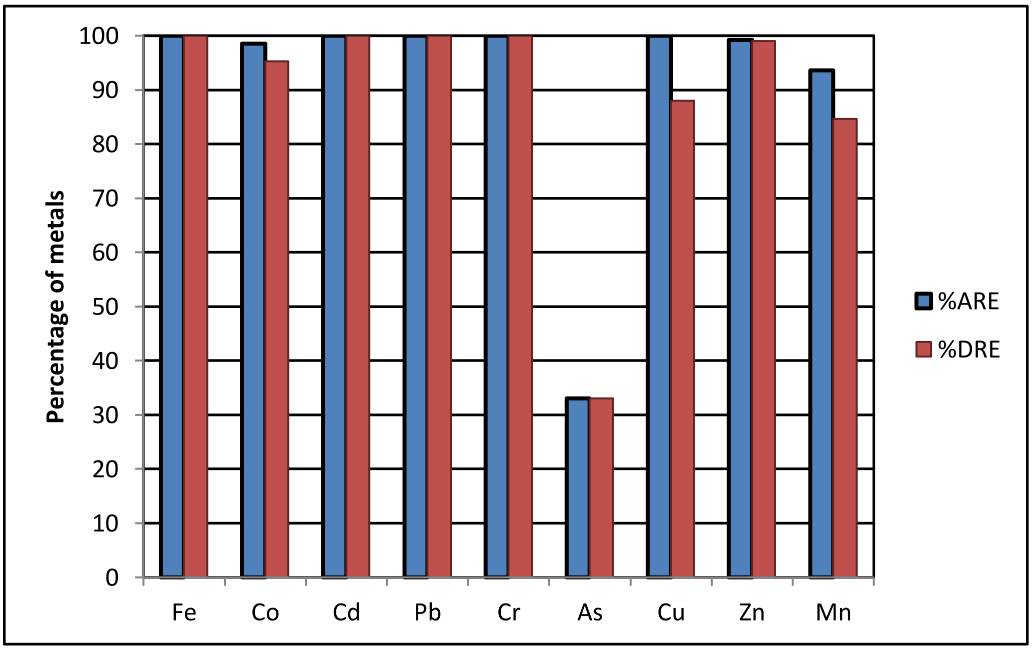

Figure 6. Percent of metals removal efficiency by algae and duckweed (ANRE=Algae nutrient removal efficiency and DNRE= Duckweed nutrient removal efficiency)

3.3. Irrigation Water Quality Index (IWQI)

The IWQI is shown in Fig. 7. However, the comparison between the initial and final IWQI of wastewater after treatment by algae and duckweed showed a significant increase in value from 11 to 14 and 15, respectively, during the 15 day of experiment.

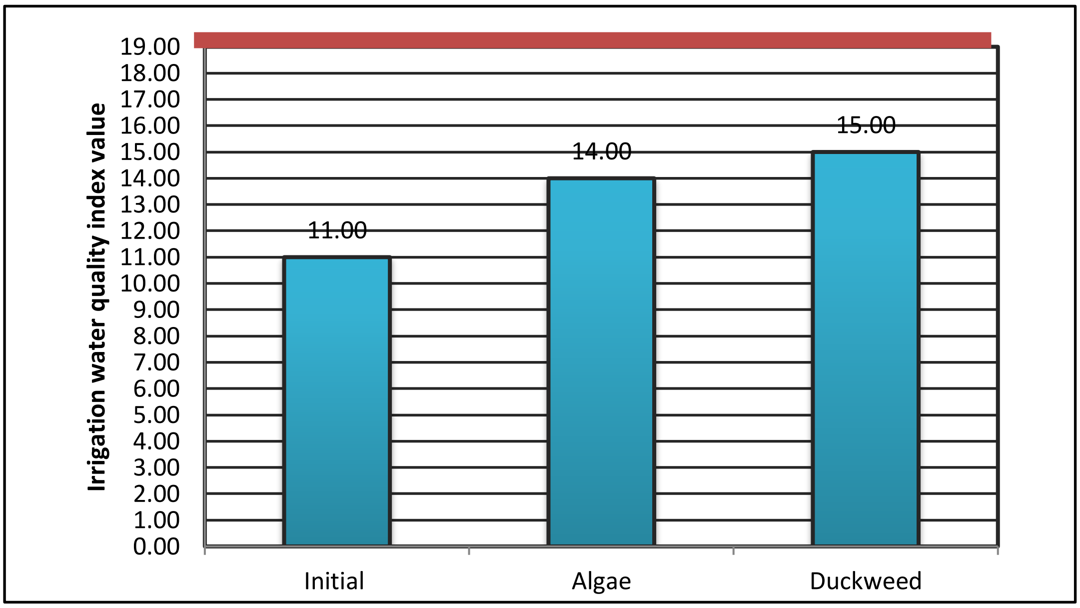

Figure 7. Initial IWQI of wastewater from the Tanjaro River and final IWQI after having been treated by algae and duckweed

The red line signifies the value of water suitability for irrigation, as follows: low suitability < 19, medium 19-32 and high suitability for irrigation

According to Asadi et al. (2020) the water quality for irrigation is classified into three categories depending on suitability, as follows: low < 19, medium 19-32, and high > 32. Results showed the improvement in water quality during the experiment with the duckweed able to improve water for irrigation to a greater degree than algae as shown in Fig. 8. Percentages of improvement by algae were 27.3% and duckweed were 36.4% for 50 L of wastewater within 15 days. These results are sustained by those obtained by Sekomo et al. (2012) and Ramirez et al. (2018) who found that algae and duckweed were able to improve the quality of wastewater for irrigation [35, 40].

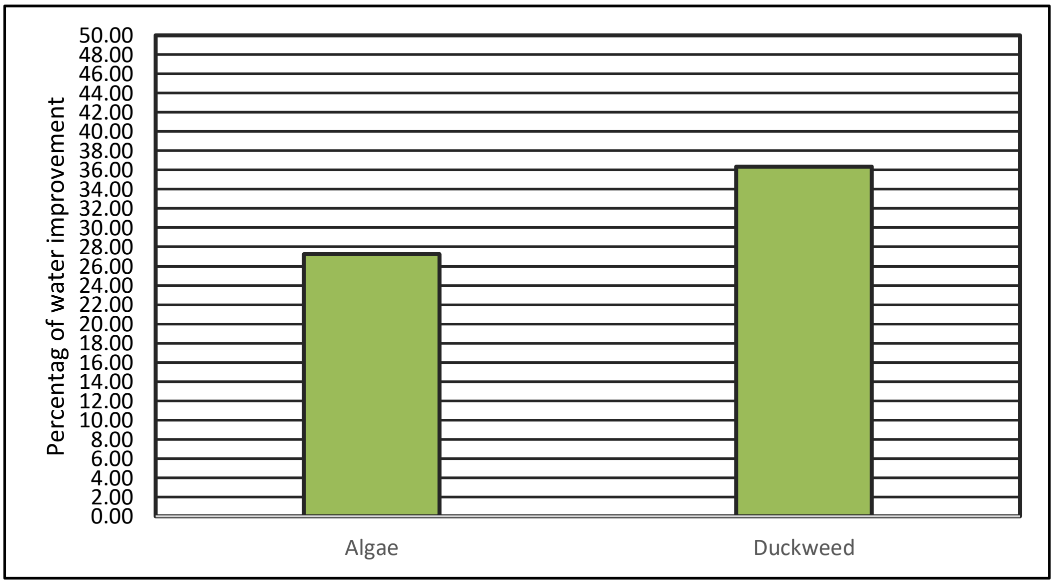

Figure 8. Percentages of improvement for algae and duckweed capacity for 50 L of wastewater in 15 days of cultivation

4. Conclusions

The present study concluded that the Tanjaro River water contains excess nutrients for growth of both algae and duckweed. Our findings suggest that in order to remediate this excess metal and nutrient concentration there is a need to expand the time for the remediation processes to meet international standards for irrigation. The results specifically point out the capacity algae to remove NO_3^-, Co, Cu, Zn, and Mn and duckweed to remove PO_4^{3-}, Cl^-, HCO_3^-, and CO_3^{2-}, although, generally, both algae and duckweed have a similar ability to remove Fe, Cd, Pb, Cr, As, and K^+. Tanjaro River wastewater after treated by algae and duckweed showed a 27.3% and 36.4% improvement, respectively, toward irrigation water quality standards. This study provides further evidence that phytotechnology is a promising avenue for wastewater remediation in this important agricultural region of Iraq.

Acknowledgements: We thank D.R. Hirmans for his help with and review of this manuscript.

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