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THE EFFICIENCY OF DUCKWEED (*LEMNA MINOR L.*) IN TREATING ROW SEWAGE WATER

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Abstract

Because of what the country is going through now from the water poverty, it has become a pressing need for the need to use all available water sources after improving their efficiency. So it was necessary to pay attention to the sewage treatment to determine how they can be used in the farming of timber trees or roses and others.

The efficiency of duckweed (*Lemna minor L.*) for improving the quality of row sewage water was assessed in a laboratory scale experiment. Total nitrogen and total phosphorus as well as some heavy metals (Pb, Cu, Fe, Zn and Cd) were monitored in row sewage before and after being treated with two dosage (5 or 10 g/l) of *Lemna minor*. Pathogenic status of the sewage water had been considered through monitoring total bacterial count, total coliform, fecal coliform, *Aeromonas sp., Pseudomonas sp.,* and total fungi. Obtained results indicated that the investigated practice resulted in removing some pollutants from the tested sewage water. The highest removal efficiencies percentages of total nitrogen, total phosphorus, Cu, Fe, Zn and Cd were 97.45, 35.51, 93.33, 99.63, 98.59 and 100 %, respectively. All tested bacteriological parameters values were reduced at the end of the experiment period. The present work revealing that the application of *Lemna minor* into sewage water, effectively improve its quality.

INTRODUCTION

Duckweeds (*Lemnaceae*) due to their small size, high multiplication rates, susceptibility to pollutants and duckweeds importance in the aquatic food web, are one of the most used aquatic plants in toxicity testing procedures of various inorganic and organic chemicals and their mixtures. Studies showed that duckweeds are very sensitive in various mixtures (such as, wastewater, leachates, etc.) (Radic *et al*., 2011 and Horvat *et al*., 2007).

Duckweeds able to remove and accumulate large amounts of heavy metals, principally through the fronds (Zayed *et al*., 1998).

The application of *Lemna gibba* L (duckweed) in wastewater treatment was found to be very effective in the removal of nutrients, soluble salts, organic matter, heavy metals and in eliminating suspended solids, algal abundance and total and fecal coliform densities (Abou El-Kheir *et al* ., 2007).

Duckweed is a floating aquatic macrophyte belonging to the botanical family *Lemnaceae*, which can be found world-wide on the surface of nutrient rich fresh and brackish waters (Zimmo, 2003). Duckweed is a variety of aquatic plant free-floating at the water surface. It is fast growing and adapts easily to various aquatic conditions. The plants can grow at temperature ranging from 5 to 35°C with optimum growth between 20°C and 31°C and across a wide range of pH (3.5-10.5) (Cayuela *et al*., 2007). Wetlands and ponds are the most common sites to find duckweed.

The nutrients taken up by duckweed are assimilated into plant protein. Under ideal growth conditions more than 40% protein content on dry weight basis may be achieved (Skillikorn *et al*., 1993).

Urban sewage contains toxic heavy metals, which are not removed properly during the traditional treatment of sewage (Chen *et al*., 2005). Therefore, removal of these toxic heavy metals from primary and secondary treated sewage has drawn the attention of workers (Weis and Weis, 2004 and Brix and Arias, 2005).

An understanding of the survival of faecal indicators is basic to the meaningful interpretation of sanitary water quality data. This is because the isolation of coliform bacteria is commonly used to indicate the potential presence of intestinal pathogens (McFeters *et al*., 2001).

Many studies have discussed the potential of aquatic plants, for reducing N and P levels in waste water. Most of these studies were limited to the physiochemical characteristics of the water. Attention has not been given to study of the microbiological changes accompanying the introduction of macrophyte to the water body (Onuoha, 2012).

Non-traditional biological treatment systems including wetlands are known to effectively remove enteric bacteria such as Escherichia coli from sewage waters (Karpiscak *et al*. 1996; Gerba *et al.* 1999; Perkins and Hunter 2000).

The present study was carried out to evaluate the suitability of *Lemna minor* L. in municipal wastewater treatment. It aimed to establish the ability of the aquatic plant to remove TP (total phosphorus), PO4 (artho-phosphate, OP), TN (total nitrogen), bacterial count and fungi from wastewater.

MATERIALS AND METHODS

The present work had been carried out in the Central Laboratory for Aquaculture Research, Abbassa, Abou-Hammad, Sharkia, governorate, Egypt during the period extended from September 2014 until June 2015

to investigate the efficiency of the aquatic duckweed; *Lemna spp*. for treating row sewage water through improving the water quality parameters.

Sewage water collection and preparation:

Raw sewage water was collected from different sites of the sewage drain which located in Hoda Shaarawy village, Abou-Hammad, Sharkia, governorate, Egypt. The collected raw sewage water was transferred to the lab, until being treated with *Lemna spp*.

Plant collection and preparation:

Lemna minor plants were collected from the nearby water surfaces, air dried in the shadow, and then placed in newspapers for the absorption of excessive water.

Experimental design:

Six treatments in triplicates were distributed in 18 glass aquaria each of 20 l water volume (40 x 50 x 10 cm) as follow:

T1: 100% raw sewage water with 5 g *lemna minor*/l.

T2: 50% % raw sewage water with 5 g *lemna minor*/l.

T3: 25% raw sewage water with 5 g *lemna minor*/l.

T4: 100% raw sewage water with 10 g *lemna minor*/l.

T5: 50% % raw sewage water with 10 g *lemna minor*/l.

T6: 25% raw sewage water with 10 g *lemna minor*/l.

Water sampling:

Subsurface (under duckweed mat) water samples for chemical and bacteriological parameters were collected in polyethylene bottles from each aquarium. Water samples were taken at 0, 1, 3 and 6 days. All water samples were analyzed for different chemical and bacteriological parameters.

Analytical techniques:

Chemical characteristics:

Total nitrogen and total phosphorous were detected as described in APHA (1989), where their values determined by using spectrophotometer (model, WPA Linton Cambridge UK).

Heavy metals detection:

Concentrations of Pb, Cu, Fe, Zn and Cd in sewage drainage waters were detected after being prepared according to (Parker, 1972) and then measured by using Atomic Absorption Spectrophotometer (Model Thermo Electron Corporation, S. Series AA spectrometer, UK).

Microbiological examinations:

For microbiological examination, the suitable number of each bacteria was reached, three different tenfold dilution prepared from each sample are used for the enumeration of each bacteria. For total viable count, poured plate method according to APHA (1989) was used, while for enumeration of fungi, on sabouraud dextrose agar medium as described by Oxoid Manual (1982).

Statistical analysis:

Comparison of treatment means using one-way analysis of variance (ANOVA) followed by Duncan's Multiple Range Test (DMRT) was performed to compare the different treatment means at 5% level of significance. The software SPSS, version 10 (SPSS, Richmond, USA) was used as described by Dytham (1999).

RESULTS AND DISCUSSIONS

High levels of N and P are known to cause the enrichment of our natural water bodies and cause eutrophication. Nutrients (N, P) are generally accumulated in the plant biomass and are removed through harvesting (Gregory, 1999). N and P losses can be attributed to uptake by duckweed, its attached biofilm, the biofilm attached to the walls of the systems and sedimentation of particular N and P (Korner *et al*., 2003).

Nitrogen (N) is a major component of municipal wastewater, stormwater runoff from urban and agricultural lands and wastewater from various types of industrial processes (DeBusk, 1999). The nitrogen is composed of various forms that can exist in water, such as particulate and dissolved organic N, ammonium, nitrite, and nitrate. These various forms can transform and serve as sources or end products for each other within the nitrogen cycle (Dotch and Gerald, 1995). For this reason, only TN is considered.

Initial total nitrogen concentrations in different sewage water concentrations (100, 50 and 25 %) were 26.24, 24.01 and 20.92 mg/l, respectively (Table 1). These values were decreased as a result of applying *lemna minor* for improving sewage water characteristics. The decrease rate in total nitrogen concentrations directly proportionated to either *lemna minor* dosage or contact period. As shown in Figure 1, total nitrogen removal efficiency percentages in T1 after 1, 3 and 6 days of contact were 88.67, 90.41 and 92.14 %, respectively. These values in T2 were 92.01, 95.64 and 96.08 % after 1, 3 and 6 days, respectively. In T3 total nitrogen removal efficiencies were 90.38, 95.27 and 96.83 % after 1, 3 and 6 days of applying *Lemna minor*, respectively. These values in T4 after 1, 3 and 6 days were 94.84, 96.55 and 92.48 %, respectively. Total nitrogen removal efficiencies after 1, 3 and 6 days of contact between *Lemna minor* and sewage water in T5 and T6 were 96.35, 97.45, 97.25, 95.73, 96.72 and 92.47 %, respectively.

Körner and Vermaat (1998) reported that *L. gibba* was itself directly responsible for 30% and up to 52% of the total N- and P-loss, respectively. The indirect contribution of *L. gibba* to the total nutrient removal was through algae and bacteria in biofilm on the plant surface which accounted for 35 and 32 % of the total N- and P-loss, respectively.

Table 1. Mean ± SE of total nitrogen concs. (mg/l) in initial and treated different dilutes (100, 50 and 25 %) of sewage water.

Adsorbent conc. →	**5 g/l**				**10 g/l**			
sewage water conc. ↓	**Contact period (day)**				**Contact period (day)**			
	0	**1**	**3**	**6**	**0**	**1**	**3**	**6**
100 %	26.237± 0.283^Aa	2.972± 0.15^Ab	2.518± 0.207^Abc	2.062± 0.045^Ac	26.237± 0.283^Aa	1.354± 0.067^Ac	0.906± 0.021^Ac	1.972± 0.092^Ab
50 %	24.014± 1.057^Ba	1.919± 0.067^Bb	1.048± 0.053^Bb	0.941± 0.058^Bb	24.014± 1.057^Ba	0.876± 0.053^Bb	0.612± 0.001^Cb	0.661± 0.007^Cb
25 %	20.921± 0.028^Ca	2.012± 0.103^Bb	0.99± 0.179^Bc	0.663± 0.006^Cc	20.921± 0.028^Ca	0.894± 0.007^Bc	0.686± 0.007^Bd	1.576± 0.101^Bb

Data shown with different small letters in the same row for each *lemna* concentration separately or different capital letters in the same column are statistically different at P < 0.05 level.

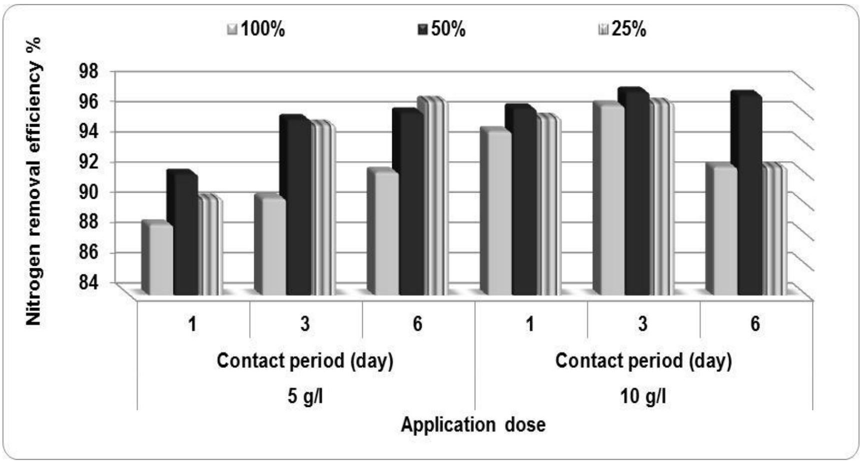

Figure 1. Total nitrogen removal efficiency after different contact periods between 5 or 10 g/l of *Lemna minor* and different dilutes (100, 50 and 25 %) of sewage water.

Phosphorus (P), like N, is a major plant nutrient, hence, addition of P to the environment often contributes to eutrophication of lakes. Phosphorus removal from aquatic macrophyte systems is due to plant uptake, microbial immobilization into detritus plant tissue, retention by underlying sediments and precipitation in the water column (Anonymus, 1998). Like nitrogen, only TP was considered for analysis during the present work, as previously mentioned by Ozengin and Elmaci (2007). Initial total phosphorous concentrations in 100 %, 50 % and 25 % sewage water were 1.6, 1.37 and 1.33 mg/l, respectively. Table 2 revealed that the application of *Lemna minor* into sewage water decreased its total phosphorous concentrations than initial values. As shown in Figure 2 the efficiency of *Lemna minor* for treating sewage water toward total phosphorous was much lower than its efficiency toward total nitrogen. Figure 2 showing that total phosphorous removal efficiency percentages in T1 after 1, 3 and 6 days of contact were 0.78, 0.55 and 8.74 %, respectively. These values in T2 were 1.43, 9.40 and 35.51%, after 1, 3 and 6 days, respectively. In T3 total phosphorous removal efficiencies were 0.22, 8.89 and 27.7 % after 1, 3 and 6 days of applying *Lemna minor*, respectively. These values in T4 after 1, 3 and 6 days were 13.42, 0.96 and 5.08 %, respectively. Total phosphorous removal efficiencies after 1, 3 and 6 days of contact between *Lemna minor* and sewage water in T5 and T6 were 1.65, 10.02, 0.1, 3.63, 6.04 and 8.85 %, respectively. In contrast to the obtained results obtained during the recent study, **Žaltauskaitė *et al*. (2014)** reported that Phosphorous was the most efficiently removed nutrient from wastewater by using *Lemna minor*.

Table 2. Mean ± SE of total phosphorous concs. (mg/l) in initial and treated different dilutes (100, 50 and 25 %) of sewage water.

Adsorbent conc. →	5 g/l				10 g/l			
sewage water conc. ↓	Contact period (day)				Contact period (day)			
	0	1	3	6	0	1	3	6
100 %	1.598± 0.022^{Aa}	1.586± 0.019^{Aa}	1.589± 0.02^{Aa}	1.459± 0.023^{Ab}	1.598± 0.022^{Aa}	1.384± 0.019^{Ab}	1.583± 0.014^{Aa}	1.517± 0.036^{Aa}
50 %	1.371± 0.02^{Ba}	1.351± 0.025^{Ba}	1.242± 0.016^{Bb}	0.884± 0.008^{Bc}	1.371± 0.02^{Ba}	1.348± 0.005^{Aa}	1.233± 0.041^{Bb}	1.369± 0.018^{Ba}
25 %	1.331± 0.011^{Ba}	1.323± 0.012^{Ba}	1.212± 0.058^{Bb}	0.962± 0.008^{Cc}	1.331± 0.011^{Ba}	1.282± 0.02^{Bab}	1.25± 0.037^{Bc}	1.213± 0.001^{Cc}

Data shown with different small letters in the same row for each *lemna* concentration separately or different capital letters in the same column are statistically different at P < 0.05 level.

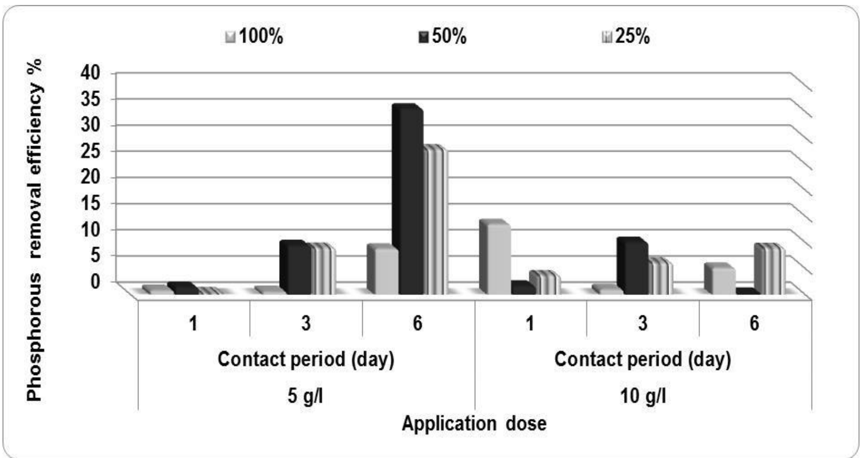

Figure 2. Total phosphorous removal efficiency after different contact periods between 5 or 10 g/l of *Lemna minor* and different dilutes (100, 50 and 25 %) of sewage water.

Heavy metals:

Aquatic plants are known for accumulating and concentrating heavy metals (Outridge and Noller, 1991) and metal fluxes rough those ecosystems (Jackson *et al*., 1994 and St-Cyr *et al*., 1994).

There are several studies that have shown that most *Lemna* spp. Show an exceptional capability and potential for the uptake and accumulation of heavy metals (Szabo *et al*. 1999; Axtell *et al*. 2003; Miretzky, *et al*. 2004). Hammouda *et al*. (1995) evaluated the efficiency of duckweed aquatic treatment in heavy metals removal in various water systems data obtained suggested a maximum reliability of systems with mixtures containing high ratios of wastewater.

Pb: Lead is one of the most abundant toxic metals that pose a serious threat to human beings, animals and phytoplanktons. In human, it is absorbed directly into the blood stream and is stored in soft tissues, bones and teeth (95% in bones and teeth) (David *et al*., 2003).

Data obtained during the present work showed that there were no detectable concentrations of lead in the row sewage water before being treated with *Lemna minor* plant.

Cu: Copper is a micronutrient and an essential component of enzymes involved in redox reactions and is rapidly accumulated by plants and animals. It is toxic at low concentrations in water and is known to cause brain damage in mammals. The toxicity of copper is dependent on local water quality conditions. Copper toxicity increases with a decrease in water hardness; dissolved oxygen; and when present in combination with other metals (South African Water Quality Guidelines, 1996).

Initial copper concentrations before applying 5g/l of *Lemna minor* into 100, 50 and 25 % of sewage water were 0.068, 0.045 and 0.023 mg/l, respectively, while these concentrations before applying 10 g/l of *Lemna minor* were 0.08, 0.03 and 0.01 mg/l, respectively (Table 3). These concentrations decreased after applying *Lemna minor* into sewage water with a rate directly increased with contacting period.

Table 3. Mean ± SE of copper concentrations (mg/l) in initial and treated different dilutes (100, 50 and 25 %) of sewage water.

Absorbent conc. →	**5 g/l**				**10 g/l**			
Sewage water conc. ↓	**Contact period (day)**				**Contact period (day)**			
	0	**1**	**3**	**6**	**0**	**1**	**3**	**6**
100 %	0.0677± 0.0015 Aa	0.053± 0.005 Aa	0.03± 0.006 Ba	0.013± 0.007 Ba	0.08± 0.012 Aa	0.023± 0.01 Ba	0.009± 0.003 Ba	0.013± 0.0017 Ba
50 %	0.045± 0.0015 Ab	0.027± 0.005 Bb	0.021± 0.0012 Bab	0.0138± 0.0083 Ba	0.03± 0.01 Ab	0.006± 0.0012 Bb	0.002± 0.0004 Bb	0.0029± 0.0025 Bb
25 %	0.023± 0.0035 Ac	0.023± 0.0033 Ab	0.015± 0.0015 ABb	0.007± 0.0015 Ba	0.01± 0 Ab	0.003± 0.001 Bb	0.0035± 0.0003 Bb	ND

Data shown with different capital letters in the same row for each *lemna* concentration separately or different small letters in the same column are statistically different at $P < 0.05$ level. ND = non detectable

Figure 3 showing that Cu removal efficiency % in treatment T1 after 1, 3 and 6 days were 21.71, 55.69 and 80.80 %, respectively. These percentages in T2 and T3 were 40, 53.33, 69.33, 0, 34.78 and 69.57 %, respectively. Cu removal efficiency % in treatment T4 after 1, 3 and 6 days were 71.25, 88.75 and 83.75 %, respectively. These values in T5 were 80, 93.33 and 90.33 %, respectively.

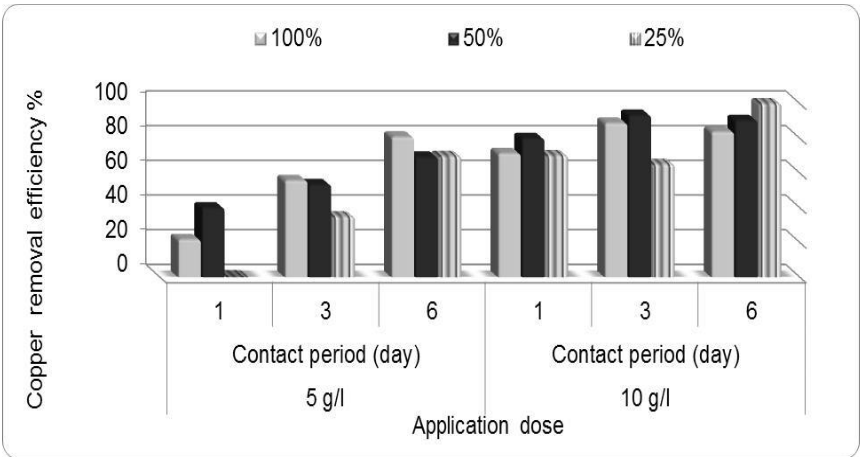

Figure 3. Copper removal efficiency after different contact periods between 5 or 10 g/l of *Lemna minor* and different dilutes (100, 50 and 25 %) of sewage water.

Abou *el*-kheir *et al*. (2007) reported that duckweed aquatic treatment system performed 100% copper after 8 days from primary treated sewage water.

Cd: Cadmium is a metal element which is highly toxic to marine and fresh water aquatic life. Elemental cadmium is insoluble in water though many of its organic and inorganic salts are highly soluble. Cadmium occurs primarily in fresh waters as divalent forms including free cadmium (II) ion, cadmium chloride and cadmium carbonate, as well as a variety of other inorganic and organic compounds. Cadmium is defined by the United States Environmental Protection Agency as potentially hazardous to most forms of life, and is considered to be toxic and relatively accessible to aquatic organisms (South African Water Quality Guidelines, 1996).

Table 4 indicating that initial cadmium concentrations in different dilutions of sewage water (100, 50 and 25 %) were 0.0035, 0.0012 and 0.0004 mg/l, respectively. Treating these waters with 5g *Lemna minor* per 1 liter of sewage water reduced its cadmium concentrations than initial. Initial cadmium concentrations in 100, 50 and 25 % sewage water before being treated with 10 g/l of *Lemna minor* were 0.0045, 0.003 and 0.002 mg/l, respectively. This practice significantly reduced sewage water Cd concentrations**.**

Table 4. Mean ± SE of cadmium concentrations (mg/l) in initial and treated different dilutes (100, 50 and 25 %) of sewage water.

Absorbent conc. →	**5 g/l**				**10 g/l**			
Sewage water conc. ↓	**Contact period (day)**				**Contact period (day)**			
	0	**1**	**3**	**6**	**0**	**1**	**3**	**6**
100 %	0.0035± 0.0009^{Aa}	0.0004± 0.0001^{Ab}	0.0035± 0.0003^{Aa}	0.0002± 0.00003^{Ba}	0.0045± 0.0009^{Aa}	0.0003± 0.0002^{A}	0.0007± 0.0003^{Aa}	0.0005± 0.0003^{Aa}
50 %	0.0012± 0.0002^{ab}	0.0005± 0.0001^{Bb}	0.001± 0.004^{Aa}	0.0008± 0.0004^{Ba}	0.003± 0.0012^{Ab}	ND	0.002± 0.002^{Aa}	0.00037± 0.0002^{Ba}
25 %	0.0004± 0.0001^{Ba}	0.00019± 0.0006^{Bb}	0.0003± 0.00003^{Bb}	0.0003± 0.0003^{Bb}	0.002± 0.0001^{Ac}	ND	0.00058±0.0042^{Ba}	0.0005±0.0003^{Ba}

Data shown with different capital letters in the same row for each *lemna* concentration separately or different small letters in the same column are statistically different at P < 0.05 level. ND = non detectable.

As shown in Figure 4 Cd removal efficiency percentages in T1 after 1, 3 and 6 days of treatment with *Lemna minor* were 88.57, 0 and 94.29 %, respectively. These values in T2 were 58.33, 16.67 and 33.33 %, respectively. Cd removal efficiency percentages in T3 after 1, 3 and 6 days of contact period were 52.5, 25 and 25 %, respectively. Cd removal efficiency percentages in T4 after 1, 3 and 6 days were 93.33, 84.44 and 88.89 %, respectively. These values in T5 and T6 were 100, 33.33, 87.67, 100, 71 and 75 %, respectively.

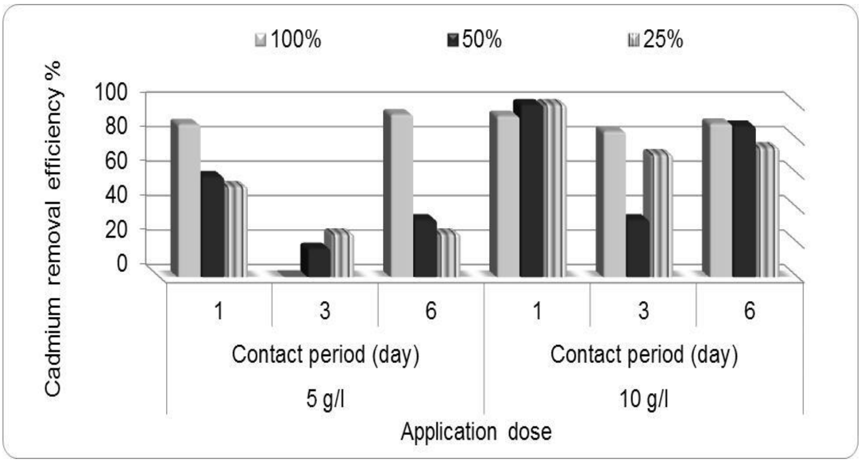

Figure 4. Cadmium removal efficiency after different contact periods between 5 or 10 g/l of *Lemna minor* and different dilutes (100, 50 and 25 %) of sewage water.

Abou *el*-kheir *et al*. (2007) reported that duckweed aquatic treatment system performed 66.7% of cadmium after treating period of 8 days from primary treated sewage water.

Zn: It's a metallic element, is an essential micronutrient for all organisms as it forms the active site in various metalloenzymes. Zinc occurs in two oxidation states in aquatic ecosystems, namely as the metal, and as zinc (II). In aquatic ecosystems the zinc (II) ion is toxic to fish and aquatic organisms at relatively low concentrations (South African Water Quality Guidelines, 1996).

Table 5 revealing that the initial zinc concentrations in 100, 50 and 25 % sewage water were 0.63, 0.547 and 0.498 mg/l, respectively. The application of 5 g/l *Lemna minor* into these waters significantly reduced these concentrations with a rate directly proportional to contact periods. Initial zinc concentrations before applying 10 g/l of *Lemna minor* were 0.593, 0.536 and 0.476 mg/l. The mentioned practice significantly reduced these concentrations with a rate directly proportional to contact periods.

Table 5. Mean ± SE of zinc concentrations (mg/l) in initial and treated different dilutes (100, 50 and 25 %) of sewage water.

Absorbent conc. →	**5 g/l**				**10 g/l**			
Sewage water conc. ↓	**Contact period (day)**				**Contact period (day)**			
	0	**1**	**3**	**6**	**0**	**1**	**3**	**6**
100 %	0.6297± 0.0107^{Aa}	0.615± 0.037^{Aa}	0.0168± 0.003^{Ba}	0.0148± 0.0009^{Ba}	0.593± 0.06^{Aa}	0.438± 0.007^{Ba}	0.3697± 0.012^{Ba}	0.335± 0.003^{Ba}
50 %	0.547± 0.0219^{Aab}	0.542± 0.028A^{ab}	0.0172±0.005^{Ba}	0.012± 0.0005^{Bb}	0.536± 0.0003^{Ab}	0.483± 0.017^{Aa}	0.293± 0.038^{Ba}	0.016± 0.003^{Cb}
25 %	0.498± 0.04^{Ab}	0.45± 0.0144^{Ab}	0.012± 0.0018^{Ba}	0.007± 0.0006^{Bc}	0.476± 0.0143^{Ab}	0.433± 0.035^{Aa}	0.313± 0.0318^{Ba}	0.012± 0.002^{Cb}

Data shown with different capital letters in the same row for each *lemna* concentration separately or different small letters in the same column are statistically different at $P < 0.05$ level.

As shown in Figure 5 removal efficiency of *Lemna minor* toward zinc in T1after 1, 3 and 6 days of contacting with sewage water were 2.33, 97.33 and 97.65 %, respectively. These values in T2 were 0.91, 96.86 and 97.81, respectively. Zn removal efficiency in T3 after 1, 3 and 6 days were 9.64, 97.59 and 98.59 %, respectively, while in T4 these values were 26.14, 37.66 and 43.51 %, respectively. In T5 and T6 these values were 9.88, 45.34, 97.02, 9.03, 34.24 and 97.48, respectively.

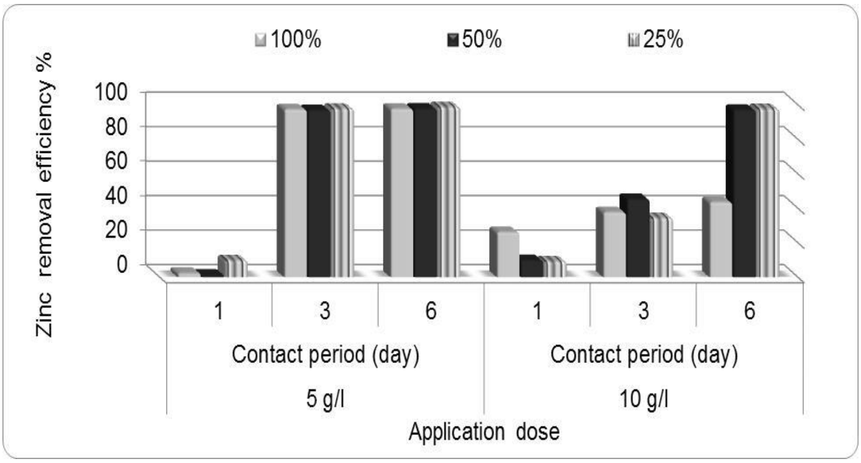

Figure 5. Zinc removal efficiency after different contact periods between 5 or 10 g/l of *Lemna minor* and different dilutes (100, 50 and 25 %) of sewage water.

Abou *el*-kheir *et al*. (2007) reported that duckweed aquatic treatment system performed 93.6 % of zinc after 8 days of treating from primary treated sewage water.

Fe: Iron is the fourth most abundant element in the earth's crust and may be present in natural waters in varying quantities depending on the geology of the area and other chemical properties of the water body. The two common states of iron in water are the reduced (ferrous, Fe) and the oxidized (ferric, Fe) states. Most iron in oxygenated waters occurs 2^{+} 3^{+} as ferric hydroxide in particulate and colloidal form and as complexes with organic, especially humic, compounds. Ferric salts are insoluble in oxygenated waters, and hence iron concentrations are usually low in the water column. In reducing waters, the ferrous form, which is more soluble, may persist and, in the absence of sulphide and carbonate anions, high concentrations of ferrous iron may be found (South African Water Quality Guidelines, 1996).

Table 6 revealing that the application of 5 g/l of *Lemna minor* into 100, 50 and 25 % of sewage water, significantly reduced its initial concentrations which were 1.816, 1,583 and 1,197 mg/l, respectively. Initial iron concentrations of 100, 50 and 25 % sewage water before applying 10 g/l *Lemna minor* were 1.903, 1.197 and 0.98 mg/l, respectively. The investigated practice significantly reduced iron concentrations. The rate of the decrease in Fe concentrations directly proportionated with the contacting period.

Table 6. Mean ± SE of iron concentrations (mg/l) in initial and treated different dilutes (100, 50 and 25 %) of sewage water.

Absorbent conc. →	5 g/l				10 g/l			
Sewage water conc. ↓	Contact period (day)				Contact period (day)			
	0	1	3	6	0	1	3	6
100 %	1.816± 0.0342^{Aa}	1.788± 0.007^{Aa}	0.0299± 0.009^{Ba}	0.0296± 0.027^{Ba}	1.903± 0.344^{Aa}	1.68± 0.00^{Aa}	1.55± 0.04^{Aa}	1.68± 0.015^{Aa}
50 %	1.583± 0.093^{Aa}	1.36± 0.153A^{ab}	0.016± 0.003^{Ba}	0.0058± 0.004^{Ba}	1.197± 0.102^{Ab}	0.667± 0.049^{Bb}	).44±0.075 ^{Cb}	0.072± 0.029^{Db}
25 %	1.197± 0.155^{Ab}	1.08± 0.162^{Ab}	0.013± 0.004^{Ba}	0.0062± 0.0026^{Ba}	0.98± 0.15^{Ab}	0.379± 0.0015^{Bc}	0.0104± 0.0004^{CDc}	0.045± 0.003^{Cb}

Data shown with different capital letters in the same row for each *lemna* concentration separately or different small letters in the same column are statistically different at $P < 0.05$ level.

Figure 6 showing that iron removal efficiencies percentages in T1 after 1, 3 and 6 days of contact between *Lemna minor* and sewage water were 1.542, 98.35 and 98.37 %, respectively. In T2 these values were 14.087, 98.99 and 98.37 %, respectively. Concerning *Lemna minor* removal efficiency toward iron, its percentages in T3 after 1, 3 and 6 days were 9.77, 98.91 and 99.,48, respectively. These values in T4 were 11.72, 18.55 and 11.72, respectively, while in T5 and T6 these values were 44.28, 63.24, 93.98, 48.58, 98.59 and 93.89 %, respectively.

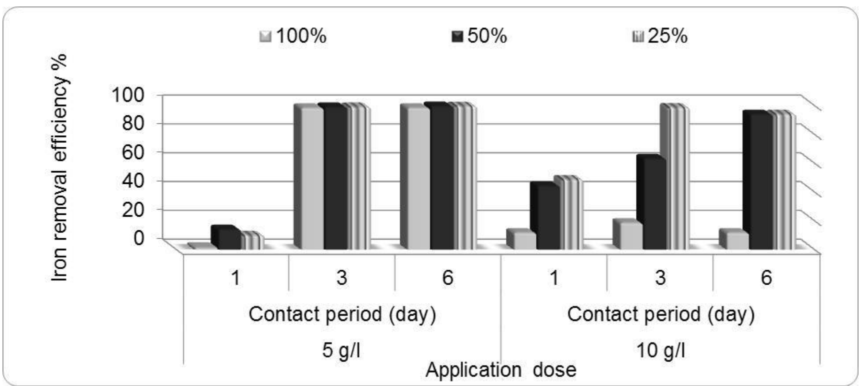

Figure 6. Iron removal efficiency after different contact periods between 5 or 10 g/l of *Lemna minor* and different dilutes (100, 50 and 25 %) of sewage water.

Abou *el*-kheir *et al*. (2007) reported that duckweed aquatic treatment system reduced 11.8 % of iron concentration after 8 days of treating from primary treated sewage water.

Obtained results during the present work are in agreement with those obtained by Žaltauskaitė *et al*. (2014) who revealed that *Lemna minor* has been shown to be a potential scavenger of nutrients and heavy metals from wastewater and may be used in wastewater treatment systems.

Bacteriological parameters:

Total bacterial count, total coliform, fecal coliform, *Aeromonas sp.*, *Pseudomonas sp.*, and total fungi were estimated after 0, 1, 3 and 6 days of the applying 5 or 10 g of *Lemna minor* to each liter of different dilutions (100, 50 or 25 %) of sewage water, to assess the plant's efficiency in purifying sewage water.

Table 7. Mean ± SE of some bacteriological parameters in different dilutions of sewage water as treated with 5g *Lemna minor* /l for different contacting periods.

Sewage water dil.	%100				%50				%25			
	Contact period (day)				Contact period (day)				Contact period (day)			
	0	1	3	6	0	1	3	6	0	1	3	6
T.C (10^4CFU/ml)	98± 12.03	47± 1.05	4.67± 0.67	9.4± 4.6	28± 5.69	17± 4.16	4± 1.00	10.1± 3.498	10± 3.287	9± 1.03	3.1± 0.3	22± 11.2
C.F (10^3CFU/ml)	900± 5.78	150± 5.78	44± 1.1	40± 1.5	139± 6.25	250± 5.78	28± 0.46	5± 0.5	117± 4.24	96± 11.58	17± 0.35	90± 0.33
E.C (10^2CFU/ml)	380± 13.3	143± 6.2	48.6± 4.6	14.3± 0.34	326.7± 6.57	100± 4.49	14± 0.55	63± 1.8	55± 11.09	162± 2.1	80± 2.00	2± 0.0
Aer. (10^2CFU/ml)	90± 11.3	35± 1.04	3.2± 0.12	7± 0.57	77.3± 3.71	31.3± 10.1	2± 0.0	2.1± 0.0	29.3± 2.3	13.67± 0.88	2± 0.0	0.1± 0.0
Ps. (CFU/ml)	700± 3.05	700± 2.08	10± 0.0	510± 0.69	14± 3.09	390± 4.9	10± 0.0	110± 0.06	10± 0.0	156± 0.5	10± 0.0	10± 0.0
Fungi (CFU/ml)	240± 7.89	70± 1.4	10± 0.0	60± 1.7	60± 2.6	70± 1.5	60± 2.3	10± 0.0	90± 0.5	30± 0.3	10± 0.0	63± 1.8

T.C = total bacterial counts, C.F = total coliform, E.C = Fecal coliform, Aer = *Aeromonas sp.* Ps. = *Pseudomonas sp.* Fungi = total fungal count.

Data in Table (7) showed that 5g duckweed/liter reduced the total bacterial counts gradually with increasing treatment period reaching the minimal values after 3 days of contacting periods in treatments; 100%, 50% and 25% with the values 4.67, 4 and 3.1(10^4 CFU/ml), respectively. The values returned to increase slightly in the 6^{th} day. The highest numbers which recorded at zero time were 98, 28 and 10 (CFU/ ml) in 100%, 50% and 25% respectively.

The lowest numbers of total coliform were recorded after 6 days of contacting periods in 100% and 50% treatments and after 3 days in 25% treatment (40, 5 and 17 [10^4 CFU/ ml]), respectively.

With respect to fecal coliform, the lowest values were recorded in the 6^{th} day of contacting period in 100% (14.3[10^2CFU/ml]) and 25% (2.0[10^2CFU/ml]) treatments while the lowest number in 50% treatment was recorded in after 3 days of contacting period (14[10^2CFU/ml]).

The present results are in agreement with those of Pandey (2001) who reported that bacteriological analysis in influent and treated effluent at Delhi duckweed pond indicated removal of fecal coliform in the range of 99.27% and 99.78% at hydraulic retention time of 6 to 14 days. Results of Ran *et al*. (2004) revealed that duckweed has a good efficiency in reducing fecal coliform by approximately 95% under average hydraulic residence time of about 4 days.

Initial *Aeromonas sp.* Counts were 9000, 7730 and 2930 (CFU/ml) for 100%, 50% and 25%, respectively. The lowest numbers in 100% and in 50% treatments, which recorded after 3 days of contacting period, were 320 and 200 (CFU/ml), respectively. With respect to 25% treatment, the lowest value (10 CFU/ml) was recorded after 6 days of contacting period.

Pseudomonas sp. numbers were decreased gradually to 100 CFU/ml 3 in both 100% and 50% at the 3^{rd} day, while in 25% treatment the investigated practice didn’t reduced the initial *Pseudomonas sp.* Count.

Total fungi counts affected with the investigated practice where its values reduced to a minimum value of 10 CFU/ml after 3 days of contacting period at both 100 and 25% treatments while the same lowest value recorded in 50% treatment was after 6 days of contacting period.

Table 8. Mean ± SE of some bacteriological parameters in different dilutions of sewage water as treated with 10 g *Lemna minor*/l for different contacting periods.

	%100				**%50**				**%25**			
	Contact period (day)				**Contact period (day)**				**Contact period (day)**			
	0	**1**	**3**	**6**	**0**	**1**	**3**	**6**	**0**	**1**	**3**	**6**
T.C	12.3	3.6	1.79	2.5	7.5	14	4.3	1.86	9.5	22	1.9	1.47
(10^4CFU/ml)	±1.9	±0.99	±0.1	±0.76	±1.8	±2.8	±0.88	±0.13	±3.3	±2.09	±0.55	±0.3
C.F	93	224	124	12.6	36	93	34	23	2.3	57	13	9
(10^3CFU/ml)	±0.57	±3.1	±1.4	±0.2	±0.88	±0.44	±0.73	±0.88	±0.33	±0.93	±0.34	±1.5
E.C	36.6	450	66.7	1.4	21.3	57.6	28	2.53	14.9	37	10	2.83
(10^3CFU/ml)	±1.8	±21.1	±1.2	±0.48	±2.3	±1.6	±0.46	±0.24	±2.9	±1.5	±1.1	±0.09
Aer.	123	40	22.6	1.2	64.6	30	6.3	2.5	53	9.7	2	4.1
(10^2CFU/ml)	±0.33	±0.57	±0.6	±0.023	±0.2	±0.5	±1.03	±0.75	±0.88	±1.2	±0.52	±0.60
Ps.	160	430	420	10	50	96	40	10	13	90	96	36
(CFU/ml)	±0.4	±0.88	±1.3	±0.0	±0.5	±33	±0.57	±0.0	±0.3	±0.57	±0.33	±0.033
Fungi	100	66	33	53	80	23	36	53	70	13	190	50
(CFU/ml)	±0.0	±2.4	±0.88	±2.4	±1.1	±0.33	±1.2	±2.40	±0.5	±0.33	±0.63	±1.2

T.C = total bacterial counts, C.F = total coliform, E.C = Fecal coliform, Aer = *Areomonas sp.* Ps. = *Pseudomonas sp.* Fungi = total fungal count.

Data in Table 8 showing that 10 g of duckweed; *lemna minor* reduced the total bacterial count and other pathogens with a rate directly proportionate with the contact period, except in few cases. The minimum values of total bacterial counts were recorded in the 3rd day (1.79×10^4 CFU/ml) in 100% and in the 6th day (1.86×10^4 and 1.47×10^4) in 50% and 25%, respectively.

Total coliform and fecal coliform counts increased than initial after 1 day of contacting period. This could be attributed to the fact that the increase in the amount of *lemna minor*, the oxygen level in the environment decreased to suit this type of bacteria, where it facultative anaerobic this conclusion mean mention previous in (MacIntyre *et al*., 2006. After that the numbers were gradually decreased and recorded the lowest numbers in the 6th day in all concentrations due to the effect of *Lemna minor*. (Haack and McFeters, 1992) revealed that *Lemna gibba* is expected to create a nutrient-rich environment through population increase and excretion of photo-assimilated organic compounds. The ability to remain within this environment enables large numbers of heterotrophic bacteria, including coliforms, to proliferate even in environments unsuitable for survival.

The investigated practice reduced *Aeromonas sp.* counts where the lowest values in 100% and 50% treatments (1.2 and 2.5 [10^2 CFU/ml]), respectively) were recorded in the 6^{th} day, while in 25% treatment the lowest count (2 [10^2 CFU/ml]) was recorded after 3 days of contacting period.

Table 8 revealing that the application of 10 g *Lemna minor* to each liter of 100% sewage water, increased its *Pseudomonas sp.* count than initial gradually through 1 and 3 days of contacting period, until decreased to the lowest value (10 CFU/ml) after 6 days of contacting period. The same manner obtained in 50% sewage water where *Pseudomonas sp.* count increased than initial after 1 day until reached the minimal count (10 CFU/ml) after 6 days of contacting period. With respect to 25% sewage water, the investigated practice increased *Pseudomonas sp.* count than initial until the end of the experiment.

The total fungal count in 100% sewage water was reduced gradually during the time period from the highest value recorded at initial (100 CFU/ml) until the 3^{rd} day of the experiment where the minimum count (33 CFU/ml) was recorded. Concerning each of 50% and 25% sewage water, the lowest total fungal counts were recorded after 1day of contacting period. The lowest counts recorded in 50% and 25% treatments were 23 and 13 CFU/ml, respectively.

CONCLUSION

In this study, phytoremediation using *L. minor* was chosen because it can enhance the properties of sewage water. The investigated practice was more efficient toward reducing nitrogen than phosphorous. The application of 5 g *lemna minor* gave better results toward removing the tested heavy metals from sewage water than the application of 10 g *lemna minor*. The investigated practice reduced the number of total bacterial count and various pathogens as total and fecal coliform, Aeromonas *spp.*, *Pseudomonas spp.* and total fungi. The use of 5g of *lemna* gave peter results than the 10g. This may be attributed to that 10 g is high density in liter and the *lemna* die-off encourages the bacterial regrowth.

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كفاءة نبات عدس الماء في معالجة مياه الصرف الخام

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الملخص العربي

نظرا لما تمر به البلاد الآن من الفقر المائي، فلقد أصبحت الحاجة ملحة إلي ضرورة إستخدام كل مصادر المياة المتاحة بعد تحسين كفاءتها. لذا كان من الضروري الإلتفات إلي معالجة مياه الصرف الصحي لتحديد مدي إمكانية إستخدامها في إستزراع الأشجار الخشبية أو الورود وخلافه.

في تجربة معملية تم إختبار كفاءة نبات عدس الماء في معالجة مياه الصرف الصحي الغير معالج حيث أضيف بتركيزان (٥ ، ١٠ جم/ لتر). تم إختبار تأثير فترات مختلفة للمعالجة (٠ ، ١ ، ٣ ، ٦ أيام) علي كفاءة عملية المعالجة. تم قياس تركيزات كل من النيتروجين الكلي والفوسفور الكلي بالإضافة إلي قياس تركيزات بعض العناصر الثقيلة (االرصاص ، النحاس ، الحديد ، الزنك والكادميوم) في مياه الصرف قبل وبعد المعالجة. تم كذلك قياس بعض العوامل البكتريولوجية (البكتيريا الكلية ، بكتيريا القولون الكلية ، بكتيريا القولون البرازية ، الإيرومونس ، البسيدومونس والفطريات.

أوضحت النتائج المتحصل عليها في نهاية الدراسة أن معالجة مياه الصرف الصحي الغير معالجة بإستخدام عدس الماء أدي لتقليل تركيزات كل العوامل المختبرة. كانت النسب المئوية لإزالة كل من النيتروجين الكلي والفوسفور الكلي والنحاس والحديد والزنك والكادميوم ٩٧.٤٥ ، ٣٥.٥١ ، ٩٣.٣٣ ، ٩٩.٦٣ ، ٩٨.٥٩ ، ١٠٠ % علي التوالي. وقد أوضحت الدراسات أيضا إنخفاض قيم كل العوامل البكتريولوجية التي تم قياسها بعد معالجة مياه الصرف بعدس الماء.

تخلص هذه الدراسة إلي كفاءة عدس الماء في معالجة مياه الصرف الصحي لتحسين خواصها الكيميائية والبكتريولوجية لتصبح أكثر ملائمة للعديد من الإستخدامات ومنها الإستزراع السمكي.