



Potentials of using duckweed (*Lemna gibba*) for treatment of drainage water for reuse in irrigation purposes

A. Allam^{a,*}, A. Tawfik^{a,*}, A. El-Saadi^b, A. Negm^a

^aEnvironmental Engineering Department, Egypt-Japan University of Science and Technology (E-JUST), P.O. Box 179, New Borg Al-Arab City, Postal Code 21934 Alexandria, Egypt, Tel./Fax: +20 03 4599520; emails: ayman.elkhelaly@ejust.edu.eg (A. Allam), ahmed.tawfik@ejust.edu.eg (A. Tawfik), negm@ejust.edu.eg (A. Negm)

^bDrainage Research Institute, National Water Research Center (NWRC), El-Kanater El-Khairia, P.O. 13621/5, Cairo, Egypt, Tel. +20 02 42189383; email: a.elsaadi@gmail.com (A. El-Saadi)

Received 11 March 2014; Accepted 12 September 2014

ABSTRACT

The potential use of duckweed (*Lemna gibba*) for the treatment of drainage water was investigated. Three continuous flow duckweed-based treatment systems (one-pond, two-pond, and three-pond) were used. Removal efficiencies of COD_{total} and ammonia in the two-pond system were significantly higher ($60.2 \pm 6.1\%$ and $80.2 \pm 1.4\%$) than that found for single-pond system ($30.6 \pm 7.9\%$ and $56.8 \pm 3.3\%$), respectively, at a total hydraulic retention time (HRT) of 14 d. Performance of three-pond system connected in series was evaluated at different HRTs of 21, 14, and 7 d. Results showed that increasing the HRT and area of duckweed pond to pond depth ($A_{\text{duckweed}}/d_{\text{pond}}$) ratio from 7 to 14 d and from 63.83 to 127.66 substantially increased the removal efficiency of COD_{total} from 59.7 ± 3.29 to $88.34 \pm 1.82\%$, respectively, resulting an effluent quality of 13.6 ± 2.3 mg COD/L in the treated effluent. However, the removal efficiency of COD_{total} remained almost constant when increasing the HRT from 14 to 21 d and $A_{\text{duckweed}}/d_{\text{pond}}$ from 127.66 to 191.49. This was not the case for nitrification efficiency, where ammonia removal increased from 32.6 ± 7.95 to $71.75 \pm 6.1\%$ and from 71.75 ± 6.1 to $85.6 \pm 4.6\%$ when increasing the HRT from 7 to 14 d and from 14 to 21 d, respectively.

Keywords: Duckweed-based treatment system; HRT; Nitrification; Protein; Arid regions

1. Introduction

Drainage water (DW) treatment and recycling facilities are becoming increasingly necessary in arid and semi-arid areas, especially in light of the shortage of conventional water resources [1]. Egypt has consistently been one of the most water-stressed countries in the world, with freshwater resources of approximately $800 \text{ m}^3/\text{capita}/\text{year}$. By 2025, it is expected to drop to

approximately $600 \text{ m}^3/\text{capita}/\text{year}$ [2]. Management of existing water resources is urgently needed, and alternative, non-conventional water resources are also essential. DW reuse is the most promising immediate and economically attractive option to make more water available for agricultural purposes [3]. However, spreading pollution in the networks of drainage canals threatens the application of these reuse activities [4,5]. Egyptian drainage networks receive large amounts of untreated domestic and industrial wastewater which

*Corresponding authors.

are unsuitable for irrigation purposes according to the Egyptian standards for direct reuse. Therefore, low-cost technology for the treatment of DW is urgently required.

Phytoremediation using submerged, floating, or emergent macrophytes is based on utilizing natural processes, and it represents an effective and low-cost technology for the treatment of contaminated water [6,7]. Duckweed are small-floating aquatic macrophytes belonging to the *Lemnaceae* family, which grow on the nutrient (N and P)-rich surface waters [8,9]. Phytoremediation of contaminated water using duckweed is promising due to its ability to grow at wide ranges of temperature, pH, and nutrient levels [10]. Moreover, duckweed has low-fiber (5%) and high-protein contents (10–40%) which represent a valuable fodder for fish and/or animals [11–13]. Duckweed-based treatment systems evaporate 20% less water compared with other open treatment systems such as waste stabilization ponds [14]. Recently, several studies have been reported for the treatment of various wastewaters using duckweed-based ponds, where 50 and 60% of nitrogen and phosphorus, respectively, were removed [15–20]. Removal efficiencies of 84, 88, and 87% for chemical oxygen demand (COD), 5-d biochemical oxygen demand (BOD₅), and total suspended solids (TSS) are registered, respectively, in duckweed-based wastewater treatment system [21–25].

Fortunately, duckweed (*Lemna gibba*) is naturally found in Egyptian drainage systems. In spite of the number of reported studies, the duckweed-based water treatment system is still largely unknown from an engineering perspective. The majority of studies carried out are laboratory-scale studies using a batch mode of operation. Therefore, the objectives of the study were to: (1) compare between the efficiency of a single- and two-duckweed pond system treating DW at a total HRT of 14 d and (2) assess the efficiency of three-duckweed pond system from the removal of COD, and nitrification efficiency at different HRTs of 7, 14, and 21 d.

2. Materials and methods

2.1. DW characteristics

DW was collected from the Gharbia drain in the middle of the Nile delta, Egypt. The drainage canal is located in a densely populated area where the domestic and food industry wastewater is directly discharged. Characteristics of the DW are presented in Table 1. Mean total dissolved solids (TDS), COD_{total}, and NH₃-N values exceed the standard limits (Law 48/1982) for reuse of DW in irrigation purposes [26].

2.2. Duckweed-based treatment system

Fig. 1 shows the schematic diagram of a continuous flow duckweed pond system [Duckweed-based treatment system (DBS)] treating DW. Three DBSs were designed and fabricated from Perspex. Each duckweed pond (DWP) had the following dimensions ($L = 0.50$ m, $W = 0.30$ m, and $D = 0.235$ m) with a capacity of 35.25 L/pond (Fig. 1). All sides of the units were covered by light impervious sheets in order to reduce unwanted algal growth. Reactors were continuously fed with the DW using a peristaltic pump (Masterflex[®] L/S). Two experiments were conducted: (1) comparison between the efficiency of a single- and two-pond system at a total HRT of 14 d and (2) assessment of the performance of three DWPs connected in series at different HRTs of 7, 14, and 21 d (Table 2). All experiments were conducted for a period of six months at a temperature of 19–25°C. Sixteen hours of photoperiod was applied to duckweed plants at a photosynthetic photon density of 101 $\mu\text{mol}/\text{m}^2 \text{ s}$ [10].

2.3. Tracer experiments

Tracer experiments were conducted to assess the hydraulic behavior in terms of the actual hydraulic retention time (HRT_{act}) and flow pattern of the three duckweed ponds (DWP-1, DWP-2, and DWP-3). Lithium chloride (LiCl) was used as a tracer pulse injection [27,28]. The flow rate was kept constant at a value of 0.211 L/h. The Li concentration was 14.56 mg/L. Three-grab samples were daily collected from the effluent of DWP-1, DWP-2, and DWP-3 for 50 consecutive days (more than two times of the theoretical HRT). Sampling frequencies were increased as days approached the end of the theoretical retention time for each DWP. Li content of the samples was analyzed using SHIMADZU, AA-7000 atomic absorption spectrophotometer.

2.4. Collection, culturing, and harvesting of duckweed

Duckweed species are found naturally in the agriculture drainage system and open water streams in Egypt. The duckweed (*L. gibba*) used in the present study was collected from the Gharbia drain in the middle of the Nile delta, Egypt. They were initially washed vigorously with tap water for 10 min to remove debris, and were acclimated for one week with DW. The cultured duckweed stocking density was 50 mg/cm² (wet weight), avoiding overcrowding, providing sufficient cover on the water surface, and overcoming algal growth. During the experimental

Table 1

Mean \pm SD characteristics of the DW used in the study and the Egyptian standards (Law 48/1982) for water reuse in agricultural purposes

Parameters	Unit	Drainage water (DW)	Law 48/1982 for reuse [26]
Total dissolved solids (TDS)	mg/L	553 \pm 82.45	<500
Turbidity	NTU	16.67 \pm 0.45	–
Total suspended solids (TSS)	mg/L	37.9 \pm 3.45	–
COD _{total}	mg/L	119 \pm 9.11	<60
COD _{soluble}	mg/L	43.42 \pm 3.17	–
COD _{particulate}	mg/L	75.67 \pm 9.14	–
Total Kjeldahl nitrogen (TKN)	mg/L	13.42 \pm 1.20	–
Nitrate (NO ₃ -N)	mg/L	0.65 \pm 0.02	<45
Ammonia nitrogen (NH ₃ -N)	mg/L	8.81 \pm 1.29	<0.5
Total phosphorous (TP)	mg/L	2.82 \pm 0.42	<1

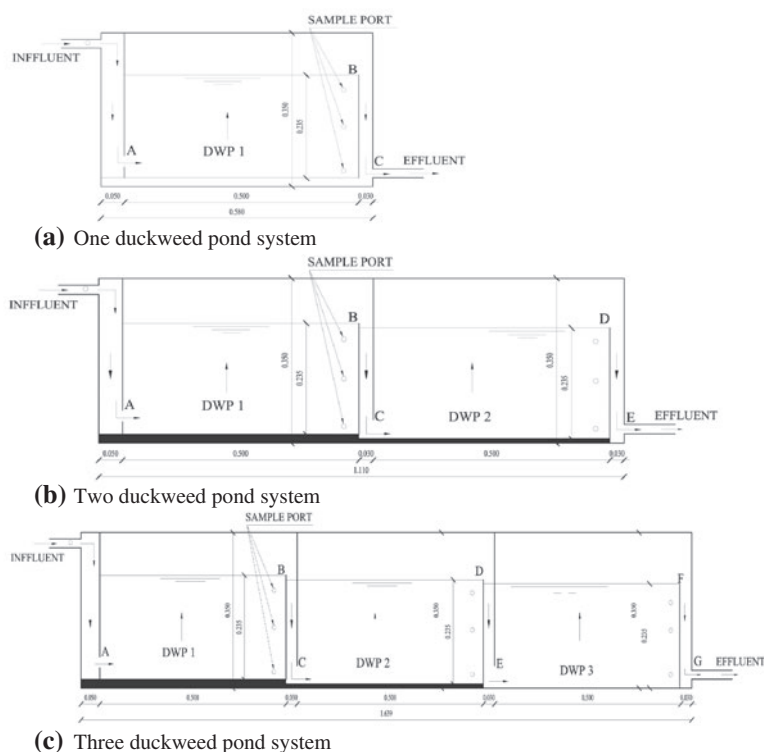


Fig. 1. Front view for continuous flow of duckweed-based treatment systems treating DW (width of all systems = 0.30 m, all dimensions in meters).

period, duckweed was harvested from each DWP every 4 d on 50% of the total surface area, which was based on the typical reported doubling time of duckweed of 2.3–7.3 d [29].

2.5. Samples collection and analytical methods

Composite samples of influent and treated effluents were collected weekly for analysis. Dissolved oxygen

(DO), temperature, pH, and TDS were measured using Thermo Scientific Orion Star™ A111 meters. Turbidity (NTU) was measured using a turbidity meter (WTW-Turb® 430 IR). TSS, COD_{total}, TKN, NH₃-N, NO₃-N, NO₂-N, and total phosphorous (TP) were measured according to APHA [30]. Filtered samples using 0.45 μ m membrane filter were used for the determination of COD_{soluble}. COD_{particulate} was calculated by the difference between COD_{total} and COD_{soluble}.

Table 2
Operational conditions of one-, two-, and three-duckweed pond systems treating DW

Exp. No.	System	V_{total} (L)	A_{duckweed} (cm ²)	$A_{\text{duckweed}}/d_{\text{pond}}$	HRT (d)	Q (L/h)
1	One-DWP	35.25	1,500	63.83	14	0.105
	Two-DWP	70.5	3,000	127.66	14	0.210
2	Three-DWP	105.8	4,500	191.49	7	0.629
					14	0.315
					21	0.210

Samples of 5 cm² were harvested periodically every 2 d for determination of the biomass production. Plants were rinsed thoroughly with deionized water, drained through a sieve, and then blotted on paper towels for 5 minutes. Afterwards, plants were weighed prior to drying at 105 °C for 24 h. The organic nitrogen content in the harvested duckweed tissue was spectrophotometrically determined using hydrogen peroxide digestion method [31]. Protein content was calculated based on the following equation protein (g/g) = organic N (g/g) × 6.25 [5]. Duckweeds production rate was calculated according to the following equation:

$$\text{Duckweeds production rate} = \frac{(D_f - D_i)}{t_0} \quad (1)$$

where D_f is the final fresh duckweed density (g/m²), D_i is the initial fresh duckweed density (g/m²), and t_0 is the harvesting cycle (d).

3. Results and discussion

3.1. Tracer study

Results for various parameters used to describe hydraulic behavior were calculated according to [28] and are presented in Table 3. The tracer experiments showed that actual retention times (HRT_{act}) were slightly higher than the theoretical one (HRT_{theo}) due to the spurious tracer curves resulting in negative dead spaces. The calculated dispersion number was 0.68 for DWP-1, 0.58 for DWP-2, and 0.31 for DWP-3. The hydraulic behavior of the three ponds (DWP-1, DWP-2, and DWP-3) was neither plug flow nor completely mixed, but rather showed a dispersed flow. These trends are similar to those obtained by Garcia et al. [28].

3.2. Comparison between the efficiency of one- and two-duckweed pond systems

Results in Table 4 show the efficiency of one- (DWP-1) and two-duckweed pond (DWP-2) systems

Table 3
Hydraulic characteristics of the three-DWP systems

Parameters	Duckweed-based pond system		
	DWP-1	DWP-2	DWP-3
HRT_{theo} (d)	7	14	21
HRT_{act} (d)	7.18	14.32	21.45
σ^2 (h ²)	40,676	135,857	164,500
R_n	0.73	0.87	1.61
Dispersion number (d)	0.68	0.58	0.31
Dead zone (%)	-2.57%	-2.29%	-2.14%
Recovery (%)	100%	100%	100%

treating DW at a total HRT of 14 d. The DWP-2 system achieved higher removal efficiency in terms of COD fractions and nitrification efficiency as compared with single DWP-1. The latter provided removal efficiencies of $56.8 \pm 3.3\%$ for COD_{total} , $42.5 \pm 3.6\%$ for COD_{soluble} , and $69.8 \pm 4.9\%$ for $COD_{\text{particulate}}$. This corresponds to $80.2 \pm 1.4\%$, $59.1 \pm 5.1\%$, and $84.3 \pm 4.69\%$ in a DWP-2, respectively. Apparently, the remaining portion of COD in the treated effluent ($COD_{\text{total}} = 23.6$ mg/L) was partly inert or slowly biodegradable [32]. The removal efficiency of ammonia in the DWP-1 system ($30.6 \pm 7.9\%$) was significantly lower than that found for the DWP-2 system ($60.17 \pm 6.1\%$). The ammonia and nitrate concentrations in the treated effluent of the DWP-2 system were 3.48 ± 0.6 and 1.01 ± 0.13 mg/L as compared with 6.1 ± 1.0 and 2.29 ± 0.2 mg/L for the DWP-1 system, respectively. A higher nitrification efficiency in the two-duckweed pond system is mainly due to a high biomass of duckweed, and $A_{\text{duckweed}}/d_{\text{pond}}$ which provides a relatively higher fraction of nitrifiers contained in its roots and induces turbulence near the interface, facilitating efficient mass transfer (oxygen, substrate, and nutrients, etc.). The biomass concentration was 1.45 g/m² in the DWP-1 and 1.87 g/m² in the DWP-2 system. Moreover, the measured DO was 4.25 ± 0.25 and 5.47 ± 0.22 mg/L in the DWP-1 and DWP-2 system, respectively. Results that are summarized in Table 4 revealed that the two-duckweed pond

Table 4

Comparison between the efficiency of a single- (DWP-1) and two-duckweed pond (DWP-2) system treating DW at an HRT of 14 d

Parameters	Unit	DW influent	DWP-1 effluent	R (%)	DWP-2 effluent	R (%)
pH		7.10 ± 0.30	7.40 ± 0.30		7.55 ± 0.20	
DO	mg/L	3.35 ± 0.25	4.25 ± 0.25		5.47 ± 0.22	
TDS	mg/L	553 ± 82.45	439.03 ± 18.25	20.60 ± 2.05	394.72 ± 14.91	28.622 ± 1.35
Turbidity	NTU	16.67 ± 0.45	8.05 ± 0.26	51.7 ± 2.81	3.67 ± 0.15	77.98 ± 3.19
TSS	mg/L	37.9 ± 3.45	15.5 ± 1.80	59.66 ± 3.84	9 ± 1.66	76.68 ± 3.18
COD _{total}	mg/L	119 ± 9.11	47.87 ± 5.14	56.79 ± 3.29	23.58 ± 2.69	80.23 ± 1.4
COD _{soluble}	mg/L	43.42 ± 3.17	24.89 ± 1.7	42.54 ± 3.61	17.68 ± 2.06	59.13 ± 5.06
COD _{particulate}	mg/L	75.67 ± 9.14	22.97 ± 5.17	69.75 ± 4.94	11.74 ± 3.23	84.29 ± 4.69
TKN	mg/L	13.42 ± 1.20	9.21 ± 1.39	35.39 ± 9.88	5.34 ± 1.07	58.08 ± 5.63
NO ₃ -N	mg/L	0.65 ± 0.02	2.29 ± 0.21	–	1.01 ± 0.13	–
NH ₃ -N	mg/L	8.81 ± 1.29	6.10 ± 1.03	30.57 ± 7.88	3.48 ± 0.58	60.17 ± 6.10
TP	mg/L	2.82 ± 0.42	1.38 ± 0.16	50.25 ± 7.7	0.71 ± 0.18	74.41 ± 7.73

system achieved higher removal efficiency than the single-DWP system. Accordingly, the performance of three-duckweed ponds connected in series was evaluated.

3.3. Performance of three-duckweed pond system

Results presented in Fig. 2(a)–(c) show the variations of COD fractions in the three-duckweed pond system at different HRTs of 7, 14, and 21 d. Results reveal a significantly improved COD_{total} removal when increasing the HRT from 7 to 14 d; however, the COD removal efficiency remained almost constant when increasing the HRT from 14 to 21 d. The system provided a mean effluent quality of 13.6 ± 2.3 mg/L for COD_{total} at a HRT of 14 d, which is significantly lower than that at an HRT of 7.0 d (47.9 ± 4.1 mg/L). Correspondingly, removal efficiencies of COD_{total} were 59.7 ± 3.29%, 88.34 ± 1.82%, and 90.97 ± 1.27%, at HRT of 7, 14, and 21 d, respectively. The improved removal efficiency of COD_{total} was mainly due to a higher removal efficiency of COD in a soluble and particulate form (Fig. 2(b) and (c)). This performance toward the removal of COD_{particulate} and COD_{soluble} can be attributed to settling of coarse suspended solids and biodegradation/uptake processes. Oron et al. [33] and Awuah et al. [22] found that the duckweed contribution for the removal of organic matter is due to their ability to directly use simple soluble organics, as well as the microbial degradation processes.

Low removal efficiency of COD_{total} at HRT of 7 d can be explained by the decreased degradation of organics, possibly due to lower contact time between the substrate and the cultivated duckweed, subsequently reducing the mass transfer capabilities. Moreover, DO concentrations dropped from 5.45 ± 0.16 to

4.0 ± 0.10 mg/L in the duckweed system as the HRT decreased from 14 to 7 d. Results in Fig. 2(b) and (c) show that the effluent quality of COD_{soluble} and COD_{particulate} was positively affected when increasing the HRT from 7 to 14 d. Nevertheless, at an HRT of 21 d, the COD_{soluble} removal was slightly improved. This indicates that the reactor is operated under soluble and particulate organic substrate limiting conditions. Based on these results, it is recommended to apply such a system with an HRT not to exceed 7 d, as it is clear that the COD effluent was below the required Egyptian standards (COD <60 mg/L) for reuse in agriculture purposes [26].

Substantial nitrification efficiency was achieved at different HRTs in the three-duckweed pond system treating DW. Fig. 3(a) shows that the ammonia concentration in the final effluent increased from 2.63 ± 0.6 to 6.25 ± 1.0 mg/L when decreasing the HRT from 14 to 7 d, respectively. However, the ammonia residual in the final effluent reduced to 1.34 ± 0.4 after increasing the HRT up to 21 d. At HRTs of 7, 14, and 21 d, ammonia was removed by 32.6 ± 7.9, 71.75 ± 6.1, and 85.6 ± 4.6% (Fig. 3(c)), while 1.15 ± 0.21, 1.04 ± 0.13, and 1.20 ± 0.15 mg/L nitrate were produced, respectively. Similar trends were observed by Zimmo et al. [34] and Awuah et al. [22]. The presence of higher ammonia concentrations in the final effluent at HRT of 7 d can be mainly due to the shorter reaction time. However, the removal efficiency for ammonia (85.6%) is substantially higher than that (60.15%) obtained by Awuah [22] in a three-duckweed pond system treating diluted sewage with HRT of 7 d.

The effluent of TKN concentration of the DWP system was substantially increased from 2.67 ± 0.22 to 12.05 ± 1.02 mg/L as the HRT decreased from 21 to

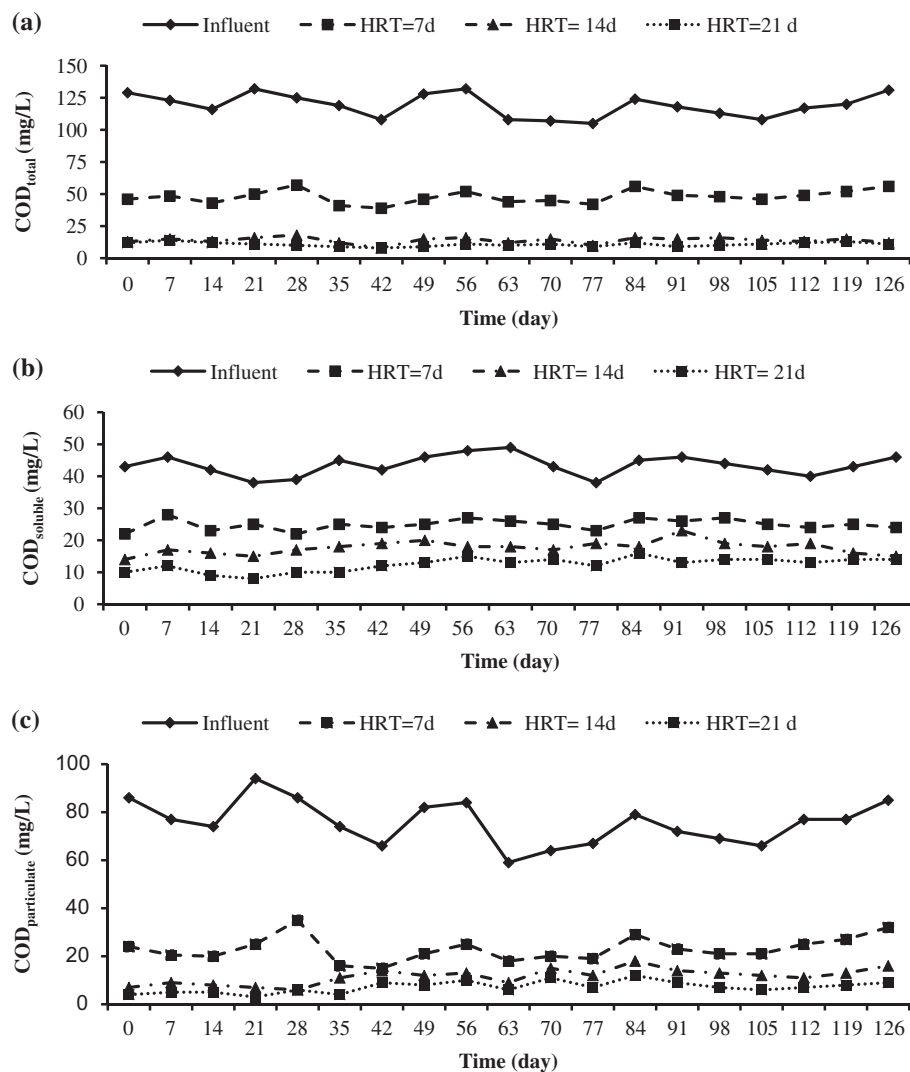


Fig. 2. Variation of COD_{total} (a), COD_{soluble} (b) and COD_{particulate} (c) in the influent and the effluent of the three-duckweed pond system at different HRTs.

7 d, respectively, (Fig. 3(b)). The nitrogen balance made across the DWP system indicates that 4.8 ± 0.67 and $9.87 \pm 1.05\%$ of nitrogen remained unaccountable at HRTs of 21 and 7 d, respectively. The observed nitrogen removal implies that anoxic conditions must have formed somewhere in the system. Duckweed DO gradients supply adequate growth conditions for nitrifying bacteria in the surface layers, and for denitrifying bacteria in the deeper layers.

3.4. Effect of DWP area-to-pond depth ratio on the removal efficiency of COD_{total}, TKN, NH₃-N, and TP

The area of the DWP-to-the pond depth ratio ($A_{\text{duckweed}}/d_{\text{pond}}$) affected the removal efficiency of

COD_{total}, TKN, NH₃-N, and TP in two ways. First, reducing the depth would decrease the distance required for diffusion of organics and nutrients (N and P) from the lower regions to the upper areas of the reactors in which the area is accessible to duckweed roots. Second, large surface areas support a greater number of plants, resulting in an increase in the uptake of COD_{total}, nitrification, and phosphorus. Reinhold [35] found that the uptake of organics and nutrients from DW by aquatic plants could be described by the pseudo-first-order reaction equation. The model of pseudo-first-order reaction is as follow:

$$C_t = C_0 \cdot e^{-K_r \cdot t} \quad (2)$$

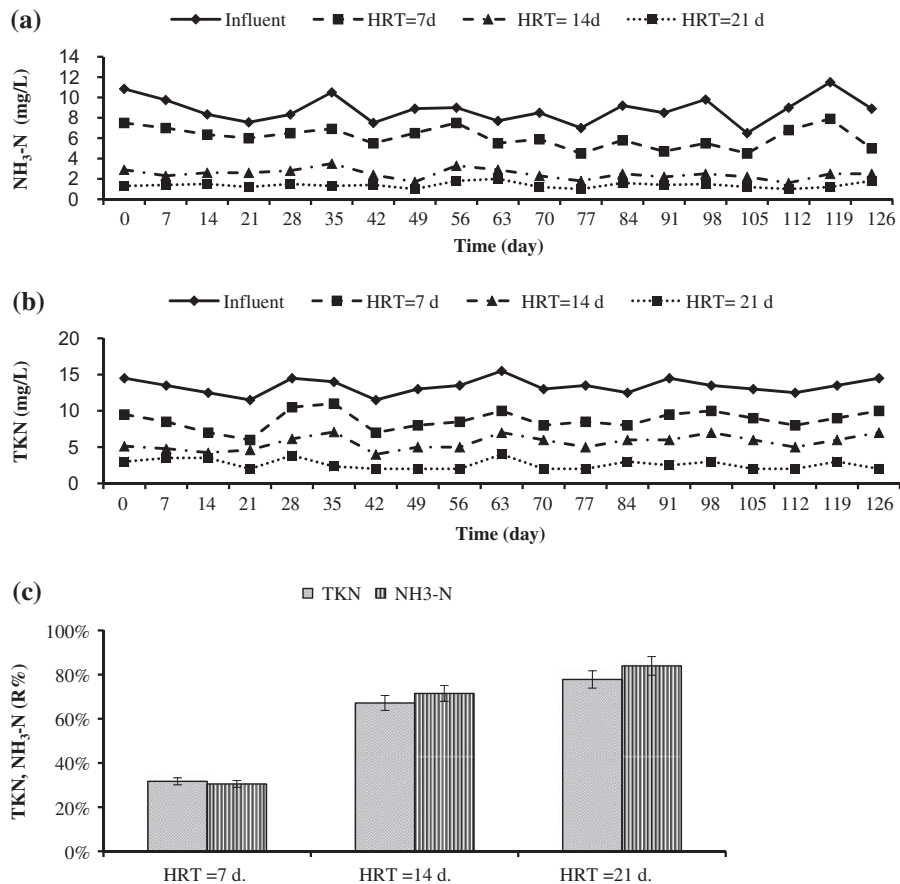


Fig. 3. Variation of NH₃-N (a) and TKN (b) in the influent and the effluent of three-duckweed pond system, and removal efficiency of NH₃-N and TKN (c) at different HRTs.

where C_t is the residual concentration (mg/L) of the pollutant at t time (d); C_0 is the initial concentration (mg/L); and K_r is the first-order removal rate constant (d^{-1}).

First-order uptake rate coefficients are dependent on duckweed mass, contact time, and initial concentration [35]. The first-order removal rate coefficients were established by fitting normalized concentration data through the DWP system using Excel 2013. Potential relationships between initial concentrations of COD_{total}, NH₃-N, TKN, and TP and removal efficiency using duckweed systems at different $A_{\text{duckweed}}/d_{\text{pond}}$ ratios are summarized in Table 5. Results revealed a significantly improved COD_{total}, TKN, NH₃-N, and TP removal rate when increasing $A_{\text{duckweed}}/d_{\text{pond}}$ ratio from 63.83 to 127.66. The COD_{total}, TKN, NH₃-N, and TP removal rate (K_r) were increased from 0.059 to 0.116 d^{-1} , from 0.034 to 0.073 d^{-1} , from 0.036 to 0.078 d^{-1} , and from 0.062 to 0.107 d^{-1} , respectively. Nevertheless, the removal rate (K_r) values were slightly improved by increasing the $A_{\text{duckweed}}/d_{\text{pond}}$ ratio from 127.66 to 191.49 (Table 5).

3.5. Duckweed biomass production

The yield of duckweed biomass and its characteristics is presented in Table 6. Dry weight yield was 80.820 ± 0.334 , 85.695 ± 0.325 , and 92.568 ± 0.436 kg/ha d in DWP system at HRTs of 7, 14, and 21 d, respectively. Similar results were reported by Ran et al. [7]: the duckweed yield was about 74–164 kg dry matter/ha d. Dry matter content of the duckweed ranged between 4.66 ± 0.32 and $5.20 \pm 0.21\%$. It was reported by Benjawan and Koottatep [29] that the dry duckweed yield ranged between 80 and 150 kg/ha d for *L. gibba* treating domestic wastewater. The average protein contents of duckweed dry matter were $21.35 \pm 0.76\%$, $19.2 \pm 0.64\%$, and $18.57 \pm 0.34\%$ in DBS at an HRT of 7, 14, and 21 d, respectively. Benjawan and Koottatep [28] reported a protein content of 15–48.1% in the dry matter of *L. gibba* treating domestic wastewater. Higher protein content of *L. gibba* (31.8–47.1%) grown on a mixture of the Nile water mixed with domestic wastewater was reported by Hammouda et al. [36].

Table 5
Removal rate coefficients of DW pollutants at different $A_{\text{duckweed}}/d_{\text{pond}}$

Parameters	$A_{\text{duckweed}}/d_{\text{pond}}$	Exponential eq.	Measured K_r (d^{-1})	Simulated K_r (d^{-1})	R^2
COD _{total}	63.83	$y = 126.99e^{-0.061t}$	0.059	0.061	0.997
	127.66	$y = 130.26e^{-0.105t}$	0.116	0.105	0.988
	191.49	$y = 127.63e^{-0.116t}$	0.093	0.116	0.999
TKN	63.83	$y = 15.03e^{-0.032t}$	0.034	0.032	0.970
	127.66	$y = 14.77e^{-0.071t}$	0.073	0.071	0.996
	191.49	$y = 14.594e^{-0.075t}$	0.085	0.075	0.951
NH ₃ -N	63.83	$y = 10.115e^{-0.036t}$	0.036	0.036	0.999
	127.66	$y = 10.182e^{-0.078t}$	0.078	0.078	0.992
	191.49	$y = 10.257e^{-0.084t}$	0.098	0.084	0.989
TP	63.83	$y = 3.34e^{-0.056t}$	0.062	0.056	0.972
	127.66	$y = 3.328e^{-0.092t}$	0.107	0.092	0.965
	191.49	$y = 3.305e^{-0.1t}$	0.111	0.100	0.962

Table 6
Biomass production rate and protein content in DWPs

HRT	HRT (7 d)	HRT (14 d)	HRT (21 d)
Fresh yield (t/ha d)	1.552 ± 0.435	1.765 ± 0.284	1.973 ± 0.375
Dry yield (kg/ha d)	80.82 ± 0.324	85.695 ± 0.325	92.568 ± 0.436
Dry matter (%/d)	5.20 ± 0.21	4.85 ± 0.25	4.66 ± 0.32
Protein content (%/d)	21.35 ± 0.76	19.2 ± 0.64	18.57 ± 0.34

4. Conclusions

The potentials of using duckweed-based treatment systems at different imposed HRTs for treatment of DW were investigated. Increasing the HRT from 7 to 14 d significantly improved the effluent quality of COD_{total} and ammonia in the effluent of DWP system. However, residual values of COD_{total} and ammonia remained unaffected when increasing the HRT from 14 to 21 d. Increasing the DWP surface area (A_{duckweed}) to the depth of the pond (d_{pond}) positively affected the removal efficiency of COD_{total}, TKN, NH₃-N, and TP. The average protein contents of duckweed dry matter were 21.35 ± 0.76%, 19.2 ± 0.64%, and 18.57 ± 0.34% in DBS at an HRT of 7, 14, and 21 d, respectively. Overall, the results recommended to use duckweed plants as an alternative cost-effective biological tool for the treatment of DW for reuse in agriculture purposes.

Acknowledgements

The first author would like to thank the Egyptian Ministry of Higher Education (MoHE) for providing him the financial support (PhD scholarship) for this research as well as the Egypt-Japan University of Science and Technology (E-JUST) for offering the facility and tools needed to conduct this work.

References

- [1] A. Fleifle, A. Tawfik, O. Saavedra, M. Elzeir, Treatment of agricultural drainage water via downflow hanging sponge system for reuse in agriculture, *Water Sci. Technol.* 13(2) (2013) 403–412.
- [2] A.E. Fleifle, O. Saavedra, H. Nagy, F. Elfetiany, A. Tawfik, M. Elzeir, Simulation-optimization model for intermediate reuse of agriculture drainage water in Egypt, *J. Environ. Eng.* 139(3) (2013) 391–401.
- [3] A. Fleifle, A. Tawfik, O. Saavedra, C. Yoshimura, M. Elzeir, Modeling and profile analysis of a down-flow hanging sponge system treating agricultural drainage water, *Sep. Purif. Technol.* 116 (2013) 87–94.
- [4] A. Fleifle, A. Tawfik, O. Saavedra, M. Elzeir, Assessment of the performance of a down-flow hanging sponge system for treatment of agricultural drainage water, *Desalin. Water Treat.* (2013) 1–8.
- [5] W. El-Kheir, G. Ismail, F. El-Nour, T. Tawfikand, D. Hammad, Assessment of Ganabiet-Tersa drain wastewater quality improvement of by in-stream *Lemna gibba* naturally occurring system in Egypt, *Int. J. Agric. Biol.* 9 (2007) 638–644.
- [6] D. Pateland, V. Kanungo, Phytoremediation potential of duckweed (*Lemnaminor*: a tiny aquatic plant) in the removal of pollutants from domestic wastewater with special reference to nutrients, *Bioscan* 5 (2010) 355–358.
- [7] N. Ran, M. Agamiand, G. Oron, A pilot study of constructed wetlands using duckweed (*Lemna gibba* L.) for treatment of domestic primary effluent in Israel, *Water Res.* 38 (2004) 2240–2247.

- [8] K.C. Bal Krishna, C. Polprasert, An integrated kinetic model for organic and nutrient removal by duckweed-based wastewater treatment (DUBWAT) system, *Ecol. Eng.* 34 (2008) 243–250.
- [9] J. Dalu, J. Ndamba, Duckweed based wastewater stabilization ponds for wastewater treatment (a low cost technology for small urban areas in Zimbabwe), *Phys. Chem. Earth* 28 (2003) 1147–1160.
- [10] N. Khellaf, M. Zerdaoui, Growth, photosynthesis and respiratory response to copper in *Lemna minor*: A potential use of duckweed in biomonitoring, *Iranian J. Environ. Health Sci. Eng.* 7 (2010) 299–306.
- [11] G. Oron, Duckweed culture for wastewater renovation and biomass production, *Agric. Water Manage.* 26 (1994) 27–40.
- [12] N. Ozengin, A. Elmaci, Performance of duckweed (*Lemna minor* L.) on different types of wastewater treatment, *J. Environ. Biol.* 28 (2007) 307–314.
- [13] M. Islam, M. Kabir, S. Khan, M. Ekramullah, G. Nair, R. Sack, D. Sack, Wastewater grown duckweed may be safely used as fish feed, *Can. J. Microbiol.* 50 (2004) 51–56.
- [14] G. Oron, D. Porath, L.R. Wildschut, Wastewater treatment and renovation by different duckweed species, *J. Environ. Eng.* 112 (1986) 247–263.
- [15] G. Oron, L. Wildschut, D. Porath, Waste water recycling by duckweed for protein production and effluent renovation, *Water Sci. Technol.* 17 (1984) 803–817.
- [16] N. Azeez, A. Sabbar, Efficiency of duckweed (*Lemna minor* L.) in phytotreatment of wastewater pollutants from Basrah oil refinery, *J. Appl. Phytotechnol. Environ. Sanit.* 1 (2012) 163–172.
- [17] J. Cheng, A. Stomp, Growing duckweed to recover nutrients from wastewaters and for production of fuel ethanol and animal feed: Review, *Clean* 37 (2009) 17–26.
- [18] J. Vermaat, M. Khalid Hanif, Performance of common duckweed species (*Lemnaceae*) and the water fern *Azolla filiculoides* on different types of waste water, *Water Res.* 32 (1998) 2569–2576.
- [19] S. Soda, Y. Kawahata, Y. Takai, D. Mishima, M. Fujita, M. Ike, Kinetics of nutrient removal and biomass production by duckweed *Wolffia arrhiza* in continuous-flow mesocosms, *Ecol. Eng.* 57 (2013) 210–215.
- [20] S. Lasfar, F. Monette, L. Millette, A. Azzouz, Intrinsic growth rate: A new approach to evaluate the effects of temperature, photoperiod and phosphorus–nitrogen concentrations on duckweed growth under controlled eutrophication, *Water Res.* 41 (2007) 2333–2340.
- [21] G. Alaerts, R. Mahbubar, P. Kelderman, Performance analysis of a full-scale duckweed-covered sewage lagoon, *Water Res.* 30 (1996) 843–852.
- [22] E. Awuah, M. Oppong-Peprah, H. Lubberding, H. Gijzen, comparative performance studies of water lettuce, duckweed, and algal-based stabilization ponds using low-strength sewage, *J. Toxicol. Environ. Health Part A* 67 (2004) 1727–1739.
- [23] R. Mohedano, R. Costa, F. Tavares, P. Belli Filho, High nutrient removal rate from swine wastes and protein biomass production by full-scale duckweed ponds, *Bioresour. Technol.* 112 (2012) 98–104.
- [24] C. House, S. Broome, M. Hoover, Treatment of nitrogen and phosphorus by a constructed upland-wetland wastewater treatment system, *Water Sci. Technol.* 29 (1994) 177–184.
- [25] S. El-Shafai, F. El-Gohary, J. Verreth, J. Schrama, H. Gijzen, Apparent digestibility coefficient of duckweed (*Lemna minor*) fresh and dry for Nile tilapia (*Oreochromis niloticus* L.), *Aquacult. Res.* 35 (2004) 574–586.
- [26] DRI (Drainage Research Institute), Atlas of Water Objective Uses. Report Submitted to: National Water Quality and Availability Management Project (NAW-QAM), DRI, National Water Research Center, Egypt, 2007.
- [27] O. Levenspiel, *Chemical Reaction Engineering*, Wiley, New York, NY, 1999.
- [28] J. García, J. Chiva, P. Aguirre, E. Álvarez, J. Sierra, R. Mujeriego, Hydraulic behaviour of horizontal subsurface flow constructed wetlands with different aspect ratio and granular medium size, *Ecol. Eng.* 23 (2004) 177–187.
- [29] L. Benjawan, T. Koottatep, Nitrogen removal in recirculated duckweed ponds system, *Water Sci. Technol.* 55 (2007) 103–110.
- [30] APHA, *Standard Methods for the Examination of Water and Wastewater*, nineteenth ed., American Public Health Association (APHA), New York, NY, 1995.
- [31] I. Novozamsky, V.J.G. Houba, R. van Eck, W. van Vark, A novel digestion technique for multi-element plant analysis, *Commun. Soil Sci. Plant Anal.* 14 (1983) 239–248.
- [32] A. Tawfikand, A. Klapwijk, Polyurethane rotating disc system for post-treatment of anaerobically pre-treated sewage, *J. Environ. Manage.* 91 (2010) 1183–1192.
- [33] G. Oron, A. Deveg, D. Porath, Nitrogen removal and conversion by duckweed grown on waste-water, *Water Res.* 22 (1988) 179–184.
- [34] O.R. Zimmo, N.P. Van Der Steen, H.J. Gijzen, Effect of organic surface load on process performance of pilot-scale algae and duckweed-based waste stabilization ponds, *J. Environ. Eng.* 131 (2005) 587–594.
- [35] D.M. Reinhold, Fate of fluorinated organic pollutants in aquatic plant systems: Studies with *Lemnaceae* and *Lemnaceae* tissue cultures, PhD Thesis, Georgia Institute of Technology, 2007.
- [36] O. Hammouda, A. Gaber, M. Abdel-Hameed, Assessment of the effectiveness of treatment of wastewater-contaminated aquatic systems with *Lemna gibba*, *Enzyme Microb. Technol.* 17 (1995) 317–323.