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Use of Domestic effluent through duckweeds and red tilapia farming in integrated system

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Abstract. The present study aimed to evaluate duckweeds for wastewater treatment and as an alternative dietary feed source for tilapia in a reuse pilot system. Duckweeds presented a satisfactory pollutants removal, mainly for chlorophyll "a" (99.9±2.7%), suspended solids (67.2±13.6) and turbidity (90.3±20.0%), reaching adequate criteria for aquaculture (nitrite, nitrate and COD concentrations under acceptable levels). Duckweed average production registered could reach up to 38.17 t/ha/year (dry matter), with a crude protein and fiber content of 38.03% and 16.17%, respectively. Fish fed on commercial feed presented higher growth performance compared to fish fed on dried duckweed. Final weight, feed conversion rate and specific growth rate of tilapia fed on dried duckweed were 52.5g, 4.3 and 1.17%, respectively. For commercial feed, the values found were 85.7g, 2.6 and 2.24%. Microbiological analysis confirmed that fish reared in treated wastewater were free of pathogenic organisms. Therefore, production of duckweed in wastewater can be an alternative which contributes to the sustainability of sewage treatment plants, mitigating environmental impacts. Also, it can be an alternative dietary source for tilapia, reducing feeding expenses as a mixture of dried duckweed and commercial feed (1:1) could reduce its production costs.

Key words: tilapia, wastewater reuse, duckweeds

Resumo. Uso de efluentes domésticos através do sistema integrado das lemnáceas e cultivo de tilápias. O presente estudo objetivou avaliar o potencial das lemnáceas no tratamento do esgoto doméstico e como fonte alternativa de alimento para tilápias. As lemnáceas mostraram uma capacidade de tratamento satisfatória principalmente para clorofila "a" (99.9±2.7%), Sólidos Suspensos (67.2±13.6) e Turbidez (90.3±20.0%), alcançando os padrões necessários para a aqüicultura (níveis aceitáveis de nitrito, nitrato e DQO). A produção de lemnáceas registrada pode atingir um total de 38.17 t/ha/ano na matéria seca, com um nível de proteína bruta e fibras de 38.03% e 16.17%, respectivamente. Peixes alimentados com ração comercial apresentaram melhor desempenho zootécnico que os alimentados com lemnas peletizadas. O peso médio final, conversão alimentar, e taxa de crescimento específico de tilápias alimentadas com lemnas peletizadas foram de 52.5g, 4.3 and 1.17%, respectivamente. Para os alimentados com ração comercial, esses valores foram 85.7g, 2.6 e 2.24%. Análises microbiológicas mostraram que os peixes cultivados em esgoto doméstico tratado estavam livres de organismos patogênicos. Dessa forma, a produção de lemnáceas em esgoto doméstico pode ser uma alternativa que contribui tanto para a sustentabilidade de Estações de Tratamento, como para a alimentação de tilápias, na redução dos custos de produção.

Palavras-chave: tilapia, reúso de esgoto doméstico, lemnáceas

Introduction

Water quality deterioration is related to uncontrolled urbanization processes, as well as rural activities. Waste stabilization pond systems are inexpensive and known for their ability to achieve good removal of pathogens and organic pollutants (Zimmo *et al.* 2004). However, they may not achieve secondary effluent standards in terms of suspended solids and nutrients reduction due to algal growth in the ponds (Smith and Moelyowati 2001).

Duckweed based wastewater systems are promising to be used in effluent treatment considering organic matter, pathogen and nutrient removal (Smith and Moelyowati 2001). Besides, duckweeds are floater plants, which reduce suspended solids by blocking light penetration. Thus, light availability causes algae die off, which settle or disintegrate. Ran *et al.* (2004) points the advantages of using duckweeds due to its high production rate, easy manual harvest from the surface, high protein and low fiber content.

Considering that wastewater treatment should be geared towards the effective reuse of nutrients, all these factors make duckweed systems cost-effective for recycling as fertilizer and animal feed (Ran *et al.* 2004).

Recycling of domestic sewage in fish farming and agriculture is an effective form of pollution control, which can contribute to cost recovery and provides cheap protein food production (El–Shafai *et al.* 2007). Reuse of treated sewage in fish farming has been applied in experimental systems as well as in full scale. The main objective of this practice is to fertilize the ponds and enhance natural primary and secondary productivity (Mara and Cairncross 1989).

In this context, several attempts have been made to use alternative dietary protein sources in tilapia culture. Studies have been conducted on the use of aquatic plants in tilapia feeds (El-Sayed 1999). Duckweeds are good food source for tilapia, as it contains about 35-45% crude protein, good amino acid balance and mineral profile (Mbagwu *et al.* 1988). Mohedano *et al.* (2005) reported the reduction of feeding costs when replacing fish meal by duckweed meal in tilapia diet. Tavares *et al.* (2008) showed that dried duckweed can replace up to 50% of tilapia diet without affecting weight gain in tilapia fingerlings. Therefore, the goal of this study was to evaluate the efficiency and sustainability of duckweed-based-ponds as a tertiary treatment considering pollutants removal and its use as a feed source in tilapia production.

Materials and Methods

<u>Experimental layout.</u> Outdoor experiments were conducted at the wastewater treatment plant from CASAN (SC, Brazil) located in Potecas neighborhood, metropolitan region of Florianópolis, SC, Brazil.

The wastewater treatment adopted was based on waste stabilization pond system which treats the wastewater from a population of around 300,000 habitants, presents a total retention time of 15.3 days and has a BOD (Biochemical Oxygen Demand) removal efficiency around 92.4%. The waste stabilization pond system is composed by an anaerobic (7.3 ha) and three facultative ponds (10.5; 6.7 and 3.2 ha). The anaerobic pond depth was 2.8 m and the facultative ponds were 1.0 m deep each. Water flow average was 200L/, which was measured, registered and controlled during the trial.

Treated domestic wastewater through the stabilization ponds system was pumped to a duckweed pilot scale system, which was used for the present study. The effluent used for the trial was originated from the last facultative pond.

<u>Duckweed-based ponds – pilot scale.</u> The duckweed-based (*Lemna valdiviana* Phil.) ponds were used as a tertiary treatment for effluent polishing. It was composed by four fiberglass tanks with depth of 0.90m, surface area of 2.57 m^2 and volume of 2.3 m^3 each. All tanks were connected and operated under a continuous water flow of 2 L/min. The retention time adopted was 1 day/tank, totalizing 4 days.

<u>Biomass yield and plants nutritional value.</u> Weekly, the duckweed growth rate was measured through manual harvesting and plants weighing. The biomass yield was estimated with a 20 cm side PVC square allocated into the last duckweed tank. An initial duckweed amount (30 g) was weighed and stocked into the square. Therefore, it was possible to estimate plant growth by subtracting the initial weight (30 g) from the total amount produced at the end of each week. After estimating the week production, the same initial amount was weighed again and returned (30 g) to the square. This procedure was adopted and repeated weekly during the experimental period, allowing the calculation and estimate the duckweed biomass yield in kg/ha.

Duckweeds nutritional value was determined monthly through sampling and physical-chemical analysis. The material was dried in the sun for 24 hours and later in an oven at 50°C for another 24 hours to reach constant weight. Thereafter, it was grinded and submitted to nutritional analysis following AOAC (1999).

Reuse pilot system - Red tilapia production tanks. A fish production pilot system was installed and operated during 120 days (from 1st November 2006 to 5th March 2007). The system was set in open field exposed to weather conditions with mean temperatures of 23.5°C, average humidity of 77% and precipitation of 129 mm.

The reuse pilot system received domestic wastewater treated by CASAN waste stabilization ponds and the duckweed system, used as tertiary treatment (effluent polishing). The experimental units were composed by six rectangular fiberglass tanks with volume of 500 L each. All tanks were independent from each other and presented a 100%/day water renovation, with an average influent flow rate of 0.33 L/min (Figure I). Tanks were aerated constantly (Air compressor Schulz Jet Master MS 2,3).

All six tanks were stocked with red tilapia *Oreochromis mossambicus* (Peters, 1852) × *Oreochromis niloticus* (Linnaeus, 1758) fingerlings with an average weight of 23 g at a density of ten fish/tank with three replicates per treatment, which were: *i) dried duckweed*: tanks received dried duckweed as feed source; *ii) commercial ration:* tanks received commercial feed (Guabi, 40% crude protein). Feed amount offered was based on 10% body weight in the first week and 5% until the end of the trial, divided into two feeding periods (09:00 and 16:00h). Left over feed was collected with a plastic screen in order to clean the surface and the tanks, but it was not measured. Effluent sampling and physical-chemical analysis from duckweeds and fish tanks were conducted weekly. The following parameters were measured: Total ammonia nitrogen (NH_3 + NH_4^+), nitrite (NO_2^-), nitrate (NO_3^-), total nitrogen (TKN), chemical oxygen demand (COD), biochemical oxygen demand (BOD), suspended, total and fixed solids, orthophosphate, turbidity, total and fecal coliforms. Also, pH, conductivity, temperature, dissolved oxygen, and transparency were recorded weekly. All analyses followed APHA (1998) recommendations. Total and fecal coliforms were estimated through Colilert Quantitray method and effluent samples were collected in sterile test tubes, and transferred to the laboratory within minutes. The equipments and solutions were sterilized in an autoclave before each use and all analyses were carried out in triplicate.

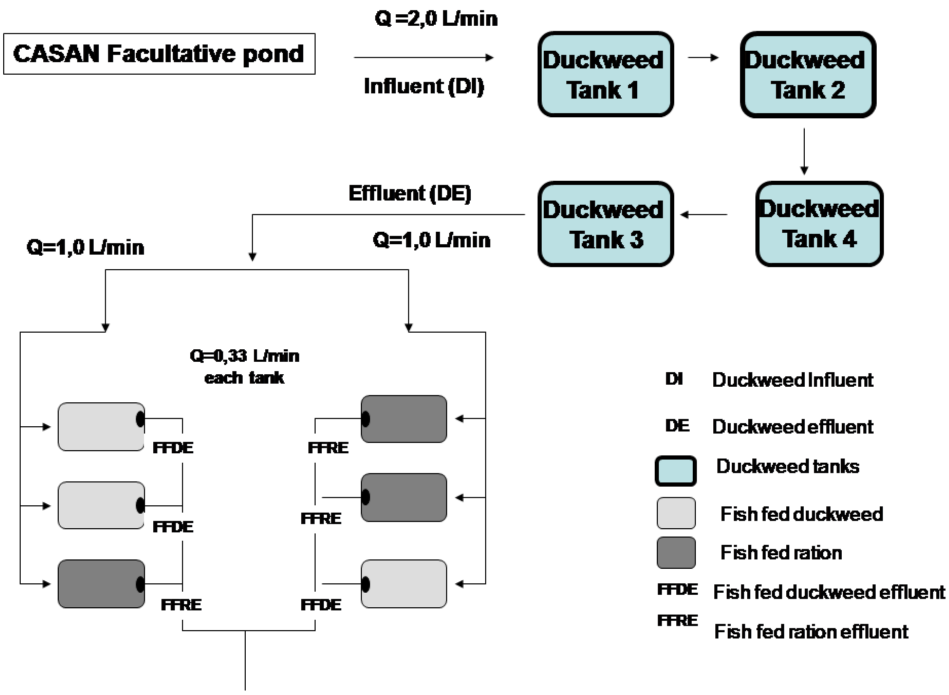

Figure 1. Duckweed-fish reuse pilot system.

Fish biometry and biomass yield. Fingerlings in each experimental unit were weighted every two weeks during experimental period for weight gain (final weight – initial weight), feed conversion (total dry feed fed /total wet weight gain), and specific growth rate (100 x Ln final average weight – Ln initial average weight/days) estimation.

Diets description. Diets compositions for the commercial feed and dried duckweed are shown in Table I.

Statistical and data analyses. Regarding the duckweed pilot system removal efficiency, all variables analyzed were considered and its polishing ability was estimated through the removal efficiency. In addition, the polishing performance of fish tanks fed ration and dried duckweed were compared by one way ANOVA, at 5 % significance level. All analyses were realized with the Statistica 7.0 software. Fish growth parameters (final weight gain, feed conversion and specific growth rate) from both treatments and effluent quality were also analyzed by one way ANOVA (Statistica 7.0).

Table I. Composition of experimental diets (dry matter).

Component, %	Commercial Feed	Dried Duckweed
Moisture	10.00	14.27
Crude protein	44.47	38.03
Crude fat	9.29	3.02
Crude fiber	7.35	16.17
Ash	9.24	14.56
Gross energy (kcal/100 g)	360	325.7

[1]Commercial feed contained a vitamin and mineral premix with the following composition (kg/product): folic acid 250mg, pantothenic acid 5000mg, biotin 125mg, copper 2000mg, coline 25000mg, iron 13820mg, iodine 100mg, magnesium 3750mg, niacin 5000mg, selenium 75mg, vitamin 1000000 IU, vitamin B_1 1250mg, vitamin B_{12} 3750mg, vitamin B_2 2500mg, vitamin B_6 1875mg, vitamin C 42000mg, vitamin D_3 500000 IU, vitamin E 20000 IU, vitamin K_3 500mg, zinc 17500mg.

Results

Duckweed and fish treatment performance. Variables analyzed and removal efficiencies found in the duckweed pilot system and fish tanks fed dried duckweed and ration are shown in Tables II, III, IV and V.

Table II. Average influent and effluent quality (with removal efficiencies) of the different parameters analyzed within the duckweed pilot system during the experimental period (n=26).

Variable	Duckweed Influent (DI)	Duckweed Effluent (DE)	Duckweeds removal efficiencies (%)
Turbidity (NTU)	96.9±53.2	9.37±5.1	90.3
Total COD (mg/L)	157.45±27.1	75.09±16.1	52.3
Soluble COD (mg/L)	101.78±26.1	59.06±14.7	42.0
NH_4^+-N (mg/L)	32.55±5.1	17.23±7.0	47.0
NO_2^- -N (mg/L)	0.06±0.05	0.47±0.25	-
NO_3^- -N (mg/L)	0.48±0.1	2.06±0.9	-
TKN (mg/L)	32.8±3.4	21.0±4.7	36.1
P-PO_4^{3+}(mg/L)	11.55±2.3	5.56±1.3	51.8
Total solids(mg/L)	1632.07±293.6	1522.0±266.7	6.2
Total Fixed Solids (mg/L)	1350.6±282.5	1328.8±303.8	1.8
Total Volatile Solids (mg/L)	295.6±105.2	200.15±49.9	26.1
Suspended Solids (mg/L)	56.14±18.1	17.10±10.6	67.2
Total Colifroms (#/100ml)	1.97E+06	3.37E+05	83.7
E. coli (#/100ml)	2.48E+03	1.22E+03	50.4
Chlorophyll “a” (mg/L)	0.07±0.47	0.01±0.03	85,7

Table III. Average influent and effluent quality (with removal efficiencies) of the different parameters analyzed within the Fish tanks fed duckweed during the experimental period (n=26).

Variables	Duckweed Influent (DI)	Duckweed Effluent (DE)	Fish tanks Effluent fed Duckweed (FTE D)	FTE D Removal Efficiency (%)
Turbidity (NTU)	96.9±53.2	9.37±5.1	17.4±13.6	82.0
Total COD (mg/L)	157.45±27.1	75.09±16.1	97.7±31.7	37.9
Soluble COD (mg/L)	101.78±26.1	59.06±14.7	66.4±22.7	34.7
NH_4^+-N (mg/L)	32.55±5.1	17.23±7.0	8.18±7.2	74.8
NO_2^- -N (mg/L)	0.06±0.05	0.47±0.25	1.6±1.42	-
NO_3^- -N (mg/L)	0.48±0.1	2.06±0.9	3.72±1.3	-
TKN (mg/L)	32.8±3.4	21.0±4.7	5.88±7.54	71.5
P-PO_4^{3+}(mg/L)	11.55±2.3	5.56±1.3	5.40±1.01	51.6
Total solids(mg/L)	1632.07±293.6	1522.0±266.7	1550.6±412.8	6.0
Total Fixed Solids	1350.6±282.5	1328.8±303.8	1286.9±328.5	5.0
Total Volatile	295.6±105.2	200.15±49.9	245.6±88.4	8.4
Suspended Solids	56.14±18.1	17.10±10.6	34.8±19.2	41.3
Total Colifroms	1.97E+06	3.37E+05	2.75E+04	82.8
E. coli (#/100ml)	2.48E+03	1.22E+03	8.20E+02	66.9
Log (10)	3.39	3.08	2.91	
Chlorophyll “a”	0.07±0.47	0.01±0.03	0.04±0.07	40,8

Table IV. Average influent and effluent quality (with removal efficiencies) of the different parameters analyzed within the fish tanks fed ration during the experimental period (n=26).

Variables	Duckweed Influent (DI)	Duckweed Effluent (DE)	Fish tanks Effluent fed Ration (FTE R)	FTER Removal Efficiency (%)
Turbidity (NTU)	96.9±53.2	9.37±5.1	20.6±16.3	66.6
Total COD (mg/L)	157.45±27.1	75.09±16.1	103.8±34.4	34.0
Soluble COD (mg/L)	101.78±26.1	59.06±14.7	69.0±35.0	32.2
NH_4^+-N (mg/L)	32.55±5.1	17.23±7.0	4.27±5.4	86.8
NO_2^- -N (mg/L)	0.06±0.05	0.47±0.25	1.28±1.02	-1216.0
NO_3^- -N (mg/L)	0.48±0.1	2.06±0.9	4.04±1.4	-831.4
TKN (mg/L)	32.8±3.4	21.0±4.7	9.29±3.79	71.6
P-PO_4^{3+}(mg/L)	11.55±2.3	5.56±1.3	4.56±1.3	60.5
Total solids(mg/L)	1632.07±293.6	1522.0±266.7	1468.00±471.0	10.9
Total Fixed Solids (mg/L)	1350.6±282.5	1328.8±303.8	1221.3±395.7	9.7
Total Volatile Solids(mg/L)	295.6±105.2	200.15±49.9	247.1±77.2	7.1
Suspended Solids (mg/L)	56.14±18.1	17.10±10.6	61.03±47.7	-3.8
Total Colifroms (#/100ml)	1.97E+06	3.37E+05	5.07E+04	97.4
E. coli (#/100ml)	2.48E+03	1.22E+03	8.20E+02	66.9
Log(10)	3.39	3.08	2.91	
Chlorophyll “a” (mg/L)	0.07±0.47	0.01±0.03	0,06±0,08	3,21

Table V. Fish tanks fed duckweed (FTED) and ration (FTER) removal efficiencies comparison through Analysis of Variance (n=26).
* Significance level of 5%

Variables	FTER Removal Efficiency (%)	FTED Removal Efficiency (%)	p value*
Turbidity (NTU)	66.6	82.0	0.9455
Total COD (mg/L)	34.0	37.9	0.8168
Soluble COD (mg/L)	32.2	34.7	0.7472
NH_4^+-N (mg/L)	86.8	74.8	0.2128
NO_2^- -N (mg/L)	-1216.0	-1542.9	0.3190
NO_3^- -N (mg/L)	-831.4	-767.9	0.7518
TKN (mg/L)	71.6	71.5	0.9288
P-PO_4^{3+}(mg/L)	60.5	51.6	0.3369
Total solids(mg/L)	10.9	6.0	0.5429
Total Fixed Solids (mg/L)	9.7	5.0	0.5197
Total Volatile Solids(mg/L)	7.1	8.4	0.9280
Suspended Solids (mg/L)	-3.8	41.3	0.0501
Total Coliforms (#/100ml)	97.4	82.8	0.2895
E. coli (#/100ml)	66.9	66.9	1.0000
Chlorophyll “a” (mg/L)	3,21	40,8	0.0481

According to the results, the duckweed pilot system presented a high removal efficiency, mainly for chlorophyll “a” (85.7%). Suspended solids removal was around 67.2±13.6. Chlorophyll “a” and suspended solids removal is related to the duckweed mat formation, which avoids sunlight penetration, reducing and limiting algae growth. Turbidity removal was also high, around 90.3±20.0%.

Organic matter measured as total and soluble COD have shown removal of 52.3% and 42.0%, respectively.

Considering the reuse pilot system, fish tanks polishing capacity was satisfactory. The highest removal efficiencies were observed for turbidity (66 to 82%), ammonia (from 74.8 to 86.8%), NTK (around 71%), total coliforms (82.8 to 97.4%) and E. Coli (66.9%). Average removal efficiency occurred for total COD (34.0 to 37.9%), soluble COD (32.2 to 34.7%) and phosphate (51.6 to 60.5%). Low efficiencies were registered for total solids (6.0 to 10.9%), total fixed solids (5.0 to 9.7%) and total volatile solids (7.1 to 8.4%). Considering suspended solids, it was noticed an increase in fish tanks fed commercial feed, probably due to a higher nutrient amount found in the commercial feed, which was excreted by fish, causing an excess algae growth.

Biomass yield and plants protein content. The average biomass yield obtained during the experimental period was 49.6 t/ha. Data from previous studies showed an annual production under these condition could reach up 381.7 t/ha, with a montly average of 31.8 t/ha. Considering the fact that duckweed present a water content of 90%, the dry matter annual production would be 38.17 t/ha. Plants protein content average found was 38.08% during the experimental period. Nitrogen and phosphorus levels were 6.08% and 0.4%, respectively (dry matter).

Fish nutritional performance. Tilapia weight gain, feed conversion rate and specific growth rate fed dried duckweed and commercial feed are shown in Table 6.

Final weight, feed conversion rate and specific growth rate of tilapia fed on dried duckweed were 52.5 g, 4.3 and 1.17%, respectively. For commercial feed, the values found were 85.7 g, 2.6 and 2.24%, respectively.

Table VI. Feed conversion rate and specific growth rate comparison between fish fed duckweed and commercial ration.

Variable	Diet		P value
	Dried duckweed	Commercial ration	
Feed conversion rate	4.3[b]	2.6[a]	0.0034
Specific growth rate	1.17[b]	2.24[a]	0.0001

* Significance level of 5%

Discussion

Duckweed pilot system performance. Rao (1986) suggests that aquatic weeds act as a biofilter by providing opportunities for aerobic heterotrophic bacteria, contributing to organic matter degradation. According to Al-Nozaily *et al.* (2000), Duckweed-covered sewage lagoons remove organic matter primarily through aerobic heterotrophic oxidation. For this, it needs the active diffusion or transportation of oxygen into the liquid phase.

Oron *et al.* (1987) obtained a removal efficiency of 66.5% in 5 days and 73.4% within 10 days, using *Lemna gibba* (Linaeus, 1978) in domestic wastewater treatment (COD from 500 to 750 $mg.L^{-1}$). In the present study, total and soluble COD removal efficiencies were 52.3±11.5 and 42.0±12.6, respectively. Therefore, it can be assumed that the retention time adopted is strongly related to duckweed systems efficiency on COD removal.

Al-Nozaily *et al.* (2000) reported that duckweed can readily absorb NH_4^+ and NO_3^- from the wastewater. The organic nitrogen in colloidal or particulate form in the sewage needs to be decomposed first by bacteria into NH_4^+. The ammonia can be removed in duckweed systems by plants uptake, nitrification-denitrification, ammonia stripping (at pH higher than eight) and algal and microbial assimilation. However, under anaerobic conditions or in the presence of a nitrification inhibitor, no nitrification-denitrification is likely to take place. Under such conditions, only duckweed uptake and ammonia stripping are the possible removal mechanisms. Besides, duckweed harvesting may also involuntarily remove attached bacteria and particulate matter that contain nutrients.

Oron *et al.* (1987) reported a number of biological, chemical and physical processes have been developed for nitrogen removal from wastewater. Although biological nitrification-denitrification processes have several advantages with respect to reliability and feasibility, removal of nitrogen by duckweeds is less energy consuming and easier for commercial implementation. Duckweeds are capable of purifying wastewater in joint performance with bacteria. Bacterial decomposition causes anaerobiosis, which is maintained by the duckweed mat that prevents aeration. The inorganic compounds which are converted into protein by duckweeds are HCO_2^{1-} or CO_3^{2}, NH_4^+ and PO_4^{3}, the main source for duckweed growth. In the present study, probably due to the occurrence of nitrification processes, it was observed an increase in NO_2^- and NO_3^- concentrations in the system. According to Zimmo *et al.* (2004), nutrient uptake is the main mechanism of nitrogen and phosphorus removal in duckweeds systems. The same authors affirm that denitrification process may occur in the duckweeds roots.

Coliforms removal, total coliforms and fecal coliforms remained around 83.7 and 50.4%, respectively, which can be explained by El-Shafai *et al.* (2007) who points that coliform removal in duckweeds systems occur due to competition of duckweeds and bacteria for nutrients and biomass harvesting, which contributes to coliforms removal from the system. These concentrations were above the acceptable limits for aquaculture.

Thus, the duckweeds performance in wastewater polishing was satisfactory considering most water quality variables, reaching acceptable conditions for fish production (nitrite, nitrate and COD concentrations).

Wastewater reuse in fish farming. Ghangrekar *et al.* (2007) affirm that maximum nitrate, nitrite and phosphate concentrations for aquaculture practice are 3.0, 0.3 and 3 mg/L, respectively. In the present study, these levels were around 2.0, 0.47 and 5.5 mg/L, showing that domestic effluent treated through stabilization ponds and duckweeds as tertiary treatment are within the permissible values. According to the same authors, maximum COD concentration is 50 mg/L and the values found were around 75 mg/L, becoming necessary a higher retention time of the effluent in duckweed ponds to increase the removal efficiency of organic matter.

Aeration was indispensable for fish production in domestic effluent, once duckweeds were not able to reduce ammonia concentration to acceptable levels for fish. In adition, the higher algae quantity present in the tanks would cause a oxygen depletion, which is desirable to remain around 4 mg/L for tilapia production.

Fish production modified effluent characteristcs, as an increase in suspended solids, chlorophyll "a" , turbidity, soluble/total COD and pH. However, ammonia, phosphate and coliforms presented lower values. It was observed that the different diets adopted in the present study did not influenced the effluent profile. Considering fish were fed with ration and peletized duckweed, it can be assumed that it increased nutrient input into the system, causing notable changes in the water quality, as higher algae production, turbidity and organic matter. Consequently, chlorophyll "a" and suspended solids analysis were also increased in fish tanks.

Duckweed production and nutritional value. The crude protein and fiber content of the dried duckweed used in the present study (38.03% and 16.17%, respectively) were similar to duckweed composition data determined by Gijzen and Khondker (1997). According to these authors, the protein content ranged from 30 to 40%, and the fiber content from 5 to 15%, when the duckweed was cultivated in a nutrient rich media. Once dried, duckweed can be stored in plastic bags, and no other treatment or process is necessary to use it directly as feed for tilapia. However, it is desirable to keep it stored in a cool environment (4°C) before using it for feeding. Furthermore, dried duckweed presents adequate stability in water, floating for up to 3h.

Duckweed protein production is higher than that observed for most vegetable crops with an annual productivity of 17.6 ton dry matter/ha/year and 37% protein concentration (Iqbal 1999). Duckweed productivity can vary from 10 to 30 ton dry matter/ha/year and its production in wastewater can be an alternative which contributes to the sustainability of sewage treatment plants, mitigating environmental impacts. According to Decamp and Warren (2000), duckweed can be used as a secondary treatment of effluents from activated sludge systems or stabilization ponds since it contributes with nutrient uptake from the wastewater, improving effluent quality. The results obtained in this study confirms these informations, as duckweed was produced in treated wastewater, contributing to environmental impacts mitigation and nutrient was converted into a high protein feed.

It has already been demonstrated that duckweed can serve as an alternative dietary source for tilapia (El Sayed 1999; Fasakin et al. 1999; Mohedano et al. 2005). However, most growth studies evaluating fresh duckweed as a dietary item have shown low weigh gain results (Gaigher et al. 1984; Hasan and Edwards 1992). Gaigher et al. (1984) evaluated the use of fresh duckweeds as feed for tilapia in a water recirculating unit and observed that fish fed only on duckweed presented lower weight gain. According to these authors, fresh duckweed intake was limited due to its high moisture content (96%) and the presence of air pockets in its leaves, which are used for flotation. The present study, however, is the first to evaluate dried duckweed fed directly to tilapia.

Tilapia growth parameters. Considering tilapia growth performance, it was observed a relevant difference between tilapia fed on dried duckweed and commercial feed. Some factors as the higher protein content and digestibility of commercial feed compared to dried duckweed could contribute to this result. Another duckweed disadvantage is related to its consumption, considering that fish need to come to the surface several times to reach satiation as duckweed floats in the water surface.

Tavares *et al.* (2008) cultivated tilapia in net-cages allocated in clean water earthen ponds, and tested three diets (commercial feed, dried duckweed and 50% commercial feed / 50% dried duckweed) for 60 days. The results showed that fish fed on commercial feed and 50% commercial feed / 50% dried duckweed presented weight gain statistically similar, which could reduce feeding expenses up to 50%. Conversion rates obtained with the three diets tested were 1.4, 2.9 and 2.0, respectively, which were better than fish produced in the treated wastewater (4,3 for fish fed on duckweed and 2,6 for fish fed on ration). In the present study, water quality, as higher ammonia concentration found in domestic effluent, could possibly have contributed to this result.

Considering growth parameters, fish fed on dried duckweed presented low perfomance compared to commercial feed. Thus, peletized duckweed is an alternative for future studies. When fish is fed on dried duckweed, it remains on the surface and does not sink, as happen with peletized ration, so if temperature is lower than 20ºC, as registered in some periods during the experiment, fish does not come to surface to eat, as its metabolism gets lower. Regarding this fact, peletized duckweed would minimize this difference, promoting more feeding oportunities.

Most of the studies conducted with tilapia production in domestic effluent show that fish productivity can reach up to 4 t/ha/year, without any suplemental feed. In the present study, fish fed on dried duckweed presented an average productivity of 11.2 t/ha/year. The higher production obtained in this study is related to the suplemental feed offered to tilapia, without any operational costs, increasing fish productivity.

The production of duckweed in wastewater can be an alternative which contributes to the sustainability of sewage treatment plants, mitigating environmental impacts.

Considering that feed can represent up to 70% of total production costs in tilapia culture in semi-intensive systems, feeding tilapia on a 1:1 mixture of dried duckweed and commercial feed could reduce production costs. However, the practical feasibility of such a feeding regime depends on duckweed production costs, which can vary depending on the resources available on each farm.

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References

AOAC (Association of the Official Analytical Chemists International). 1999. AOAC – **Association of Official Analytical Chemists. 1999. Official methods of analysis of AOAC**. 16th ed. AOAC, Washington, USA, 1141pp.

APHA (American Public Health Association, American Water Works Association and Water Pollution Control Federation). 1998. 19. **Standard methods for the examination of water and wastewater, 20th edition**. American Public Health Association, Washington, DC, USA.

Al-Nozaily, F., G. Alaerts, S. Veenstra. 2000. Performance of duckweed-covered sewage lagoons - II nitrogen and phosphorus balance and plant productivity. **Water Research** 34:2734-2741.

Decamp, O., A. Warren. 2000. Investigation of *Escherichia coli* removal in various designs of subsurface flow wetlands used for wastewater treatment. **Ecological Engineering** 14: 293-299.

El-Sayed, A. F. M. 1999. Alternative dietary protein sources for farmed tilapia, *Oreochromis* spp. **Aquaculture** 179:149-168.

El-Shafai, S., F. A. El-Gohary, F. A Nasr, N. P. Van der Steen, H. J. Gijzen. 2007. Nutrient Recovery from domestic wastewater using a UASB-duckweed ponds system. **Bioresource Technology** 98: 798-807.

Fasakin, E. A., Balogun, B. E. Fasaru. 1999. Use of duckweed, *Spirodela polyrriza*, L. Schleiden, as a protein feedstuff in practical diets for tilapia, *Oreochromis niloticus* L. **Aquaculture Research** 30: 313-318.

Gaigher, I. G., Porath, D., G. Granoth. 1984. Evaluation of duckweed *Lemna gibba* as feed for tilapia *O. niloticus* x *O. aureus* in a recirculating unit. **Aquaculture** 41:235-244.

Ghangrekar, M. M., N. Kishor, A. Mitra. 2007. Advancements in waste stabilization ponds technology - Sewage reuse for aquaculture after treatment in oxidation and duckweed pond. **Water Science and Technology** 55:173-182.

Gijzen, H., M. Khondker. 1997. **An overview of the ecology, physiology, cultivation and application of duckweed, literature review**. Report of duckweed research project, Dhaka, Bangladesh.

Hasan, M. S., P. Edwards. 1992. Evaluation of duckweed *Lemna Perpusilla* and *Spirodela polyrriza* as feed for Nile tilapia *Oreochromis niloticus*. **Aquaculture** 104: 315-326.

Iqbal, S. 1999. **Duckweed aquaculture**. Potentials, possibilities and limitations, for combined wastewater treatment and animal feed production in developing countries. Switzerland. SANDEC report n.6 91pp.

Mara, D. D., S. Cairncross. 1989. **Guidelines for the safe use of wastewater and excreta in Agriculture and Aquaculture: Measures for public health protection.** World Health Organization, Geneva.

Mbagwu, I. J., H. A. Adeniji. 1988. Nutritional content of duckweed *Lemna paucicostata* in the Kainji lake area, Nigeria. **Aquatic Botany** 29:357-366.

Mohedano, R. A., J. B. R. Rodrigues, D. M. Fracalossi. 2005. *Lemna valdiviana*, uma planta que além de tratar efluentes alimenta os peixes cultivados. **Panorama da Aqüicultura** 15:33 – 40.

Oron, G., D. Porath, H. Jansen. 1987. Performance of duckweed species *Lemna gibba* on municipal waste water for effluent renovation and protein production. **Biotechnology & Bioengineering** 29: 258-268.

Ran, N., M. Agami, G. Oron. 2004. A pilot study of constructed wetlands using duckweed *Lemna gibba* L. for treatment of domestic primary effluent in Israel. **Water Research** 38:2240-2247.

Rao, S. V. R., 1986. A review of the technological feasibility of aquacultures for municipal wastewater treatment. International Journal of Environmental Studies 27: 219-223.

Smith, M. D., I. Moelyowati. 2001. Duckweed based wastewater treatment (DWWT): design guidelines for hot climates. **Water Science and Technology** 43: 291-299.

Tavares, F. A.; J. B. R. Rodrigues, D. M. Fracalossi, J. Esquivel, R. Roubach. 2008. Dried duckweed and commercial feed promote adequate growth performance of tilapia fingerlings. **Biotemas** 21: 91-97.

Zimmo, O. R., N. P. Van der Steen, H. J. Gijzen. 2004. Nitrogen mass balance across pilot-scale algae and duckweed-based wastewater stabilization ponds. **Water Research**, 38: 913-920.

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