

ASSOCIATED FAUNA TO *EICHHORNIA CRASSIPES* IN A CONSTRUCTED WETLAND FOR AQUACULTURE EFFLUENT TREATMENT

Lúcia Helena SIPAÚBA-TAVARES *, Bruno SCARDOELLI TRUZZI *,
Ana MILSTEIN ** and Aline MARCARI MARQUES *

* São Paulo State University, Aquaculture Center, Laboratory of Limnology and Plankton Production, Via de Acesso Professor Paulo Donato Castellane s/n, CEP 14884-900, Jaboticabal SP, Brazil, sipauba@caunesp.unesp.br, brscardoeli@hotmail.com, alinemarcari@gmail.com

** Agricultural Research Organization, Fish and Aquaculture Research Station Dor, M. P. Hof HaCarmel, Israel, IL-3082000, anamilstein@agri.gov.il

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ABSTRACT

Water, sediment and associated fauna were studied in a water hyacinth (*Eichhornia crassipes*) stand of a constructed wetland, used for aquaculture effluent treatment in SE Brazil, in February-April (summer/rainy season) and July-September (winter/dry season). The hydrological regime and decomposition processes had strong impact on the wetland water quality and on the associated fauna composition. Protozoa and Rotifera were at high densities, mainly in the dry season. *Vorticella* sp. was the dominant species in both seasons. Zooplankton richness, evenness and diversity were high during both seasons, with higher levels during the rainy season. Protozoa diversity and evenness were higher in the dry season when the water volume was lower. Maximum plant residence time in this wetland should be about 60 days.

RESUMEN: Fauna asociada a *Eichhornia crassipes* en un humedal artificial para tratamiento de efluentes de acuicultura.

Agua, sedimento y fauna asociada fueron muestreados entre las *Eichhornia crassipes* de un humedal artificial en el SE de Brasil usado para tratar efluentes de acuicultura, durante febrero-abril (verano/estación lluviosa) y julio-septiembre (invierno/estación seca). El régimen hidrológico y los procesos de descomposición tuvieron un fuerte impacto sobre la calidad de agua dentro del humedal y sobre la composición de la fauna asociada. Protozoa y Rotífera presentaron alta densidad, especialmente durante la estación seca. *Vorticella* sp. fue la única especie dominante en ambas estaciones. En ambos períodos la riqueza, uniformidad y diversidad del zooplancton fueron altas, siendo mayores en la estación lluviosa. La diversidad y uniformidad de Protozoa fueron mayores en la estación seca, cuando el volumen de agua era menor. El tiempo máximo de residencia de las plantas debería ser de unos 60 días.

REZUMAT: Fauna asociată speciei *Eichhornia crassipes* într-o zonă umedă artificială pentru tratarea efluenților din acvacultură.

S-au analizat apa, sedimentul și fauna asociată dintr-o parcelă de *Eichhornia crassipes* dintr-o zonă umedă construită pentru tratarea efluenților din acvacultură în SE Braziliei, în perioadele februarie-aprilie (vară/sezon ploios) și iulie-septembrie (iarnă/sezon secetos). Regimul hidrologic și procesele de descompunere au avut un impact puternic asupra calității apei din zona umedă precum și asupra compoziției faunistice asociate. Protozoarele și rotiferele au prezentat densități mari, în special pe timpul sezonului secetos. *Vorticella* sp. a fost singura specie dominantă din ambele sezoane. Abundența, distribuția și diversitatea zooplanctonului au fost mari în ambele sezoane, dar cu valori superioare în sezonul ploilor. Diversitatea protozoarelor și distribuția lor au fost mai ridicate în sezonul secetos, când volumul de apă a fost mai scăzut. Timpul maxim de ședere al plantelor în această zonă umedă ar trebui să fie de circa 60 zile.

INTRODUCTION

Macrophytes and riverine vegetation perform different functions in the aquatic ecosystem, such as increasing habitat heterogeneity, providing food and shelter for many organisms and creating patches under specific physicochemical conditions (Curtean-Bănăduc et al., 2014; Gutierrez and Mayora, 2016). In turn they are affected by the growth rate, abundance and activity of those associated organisms (O'Hare et al., 2012). The organic matter retained among their roots consists of particles that can be utilized as food by the macrofauna associated with the plants, promoting a rapid circulation through the trophic web. Different species of macrophytes have been characterized by dissimilar physiological and chemical composition, which result in the release of different types of substrates that may affect the availability of ecological niches for different associated communities (Zeng et al., 2012).

Eichhornia crassipes, commonly known as water hyacinth, is an aquatic plant native to the Amazon Basin, with a high potential to retain inorganic and organic material in its biomass. Outside its native range it is often considered a problematic invasive species (Villamagna and Murphy, 2009), so that serious measures have been taken to impair its propagation, as well as to find a practical use for its large available biomass. In recent years it has received much attention due to its potential benefits for biogas production, fertilizer, water purification, as well as animal and aqua feed (Malik, 2007). *E. crassipes* has well-developed adventitious roots, lateral roots, and epidermal hairs, which together with its stems, leaves and stolons constitute a complex structure which offers suitable habitat or shelter for numerous organisms (Montiel-Martínez et al., 2015).

Zooplankton is a key component of freshwater ecosystems (Onciu et al., 1999) and frequently uses macrophytes as a refuge from vertebrate predators or as a substrate where to dwell and feed (Gutierrez and Mayora, 2016). Some species of the major secondary producers (Rotifera, Cladocera and Insecta larvae) are commonly found in pelagic and vegetation areas, whereas others are found in or around the vicinity of vegetation stands (Kouamé et al., 2011). Aquatic plants can be used not only as a place to grow in but also as a refuge, since they reduce mortality risk to zooplankton capable of detecting chemical cues from potential predators (Montiel-Martínez et al., 2015). In addition, macrophytes are usually covered by epiphytes that are grazed upon by several invertebrates whose diversity, abundance and distribution pattern are influenced by macrophytes (Kouamé et al., 2011).

Constructed wetlands are artificial ecosystems based on macrophytes that are widely used to treat effluents of different types. The community structure and hence the ecological functioning of these systems varies with climatic conditions. The current article investigates *Eichhornia crassipes* development, structure of its associated fauna and environmental conditions during the rainy and dry seasons, in the *E. crassipes* section of a constructed wetland used to treat effluents of an aquaculture farm.

MATERIAL AND METHODS

Study area and sampling site

The study was carried out in the constructed wetland for aquaculture effluents treatment in a southeastern Brazil farm (21°14'S and 48°18'W) (Fig. 1). Monthly means of meteorological conditions during the summer/rainy season (February to April) were – air temperature: $24 \pm 1.2^{\circ}\text{C}$, water temperature: $24 \pm 3.3^{\circ}\text{C}$ and 85 ± 2 mm rainfall; during the winter/dry season (July to September) the monthly means were $22 \pm 2.3^{\circ}\text{C}$, $23 \pm 3^{\circ}\text{C}$ and 25 ± 23 mm. The effluent entering the wetland originated from six earthen fishponds with areas varying between 1,822 and 8,067 m². Some fishponds received water from other smaller earthen ponds further up the watershed. The wetland also receives effluent (swine manure)

from an anaerobic sludge blanket reactors (UASB) installed in series, with volumes ranging between 50 and 908 l, which discharged the material about once a week when reactors are switched on. During the rainy period the wetland also receives surface runoff through a channel. The wetland was 71 m long, with a total area of 96.6 m², and lined with a shallow clay bottom. It was planted with three native macrophyte species, *Cyperus giganteus* and *Typha domingensis* (34 m²) close to the inlet water to restrain the strong water flow, mainly in the rainy season, followed by a *Eichhornia crassipes* (32 m²) stand where the associated fauna and ecological conditions were evaluated. Immediately after plant transplantation, the wetland was filled to a depth of approximately 0.30 m. The plants were installed twice, once for the summer/rainy season (February to April) sampling and once for the winter/dry season (July to September) sampling, and were completely removed between both periods.

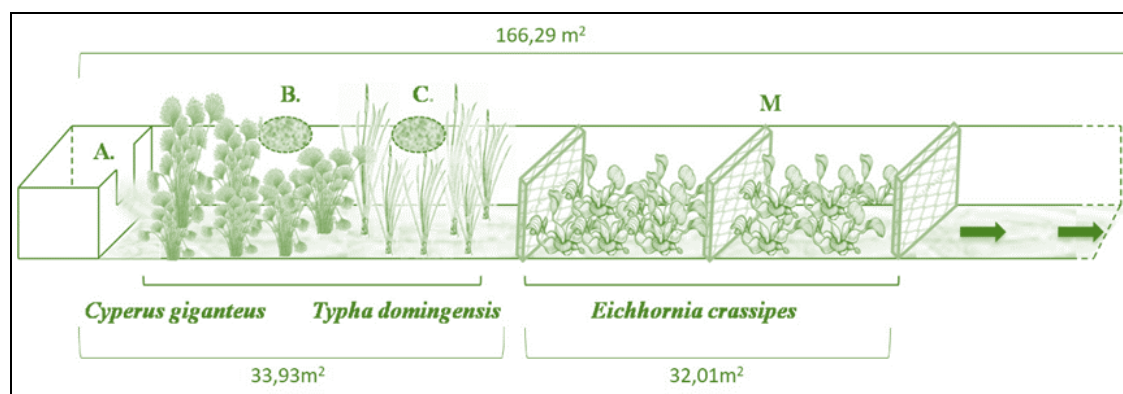


Figure 1: Diagram of the wetland system studied; A = effluents from the aquaculture farm, B = effluents from the biodigester, C = rainwater input, M = sampling site, arrows = direction of water flow.

E. crassipes biological data

The growth of *E. crassipes* biomass was determined every fortnight during each sampling period, measuring their foliar length and width and rhizome length. Measurements were taken on the same 12 marked plants randomly chosen at the beginning of the experiment. Dry and wet masses were recorded at the end of each growth period. The plants were collected using a 0.18 m² quadrant, dried at 60°C until constant weight, and weighted. Plant nutrients composition was analysed according to Bataglia et al. (1983).

Associated fauna

Samples of associated fauna were retrieved every fortnight during both periods of aquatic plant growth. All plants collected with the 0.18 m² floating quadrant, were washed in the laboratory with distilled water using 200 µm, 58 µm and 25 µm sieves. The collected fauna was preserved in 4% formalin, allowed to settle and the total collected volume measured, and stored in amber glass jars. Rotifera and Protozoa samples were analyzed with a Sedgewick-Rafter counting cell and examined under 100X magnification. Cladocera, Copepoda, Insecta, Mollusca, Platyhelminthe, Nematoda, Ostracoda and Harpacticoida were counted in a reticulated chamber. Protozoa were grouped into two taxonomic categories: Testate amoebae (Amoebozoa and Rhizaria) and Ciliates (Ciliophora) (Adl et al., 2005). Insecta were identified at order level and Nematoda, Mollusca and Platyhelminthes at phylum level.

Water and sediment

Surface water and sediment samples were collected every fortnight inside the *E. crassipes* stand. Water was sampled with a one l Van Dorn bottle and transported in refrigerated polyethylene bottles to the laboratory. Water temperature, turbidity, pH, dissolved oxygen (DO) and conductivity were measured in situ with a Horiba U-10 multi-sensor. Total phosphorus and nitrogen compounds were quantified spectrophotometrically, following Golterman et al. (1978) and Koroleff (1976). Chlorophyll-*a* was extracted with alcohol 90% and quantified at 663 nm and 750 nm (Nusch, 1980). Total suspended solids (TSS) and total dissolved solids (TDS) were determined according to Boyd and Tucker (1992). Water samples for microbiological analysis, using the multiple-tube methods, were collected in sterilized 500-ml flasks and taken to the laboratory in an isothermal container. The material used in the microbiological analysis (thermotolerant coliforms) was sterilized prior to use (APHA, 1995). Vertically mixed sediment samples were retrieved with a four cm diameter PVC core up to approximately 10 cm deep. Sediments were air dried, gently disaggregated and dried in a convection oven at 70°C until completely dry. Determinations of organic matter (OM), Ca, Mg, P, K, N, Fe, Zn and pH were performed according to methods described by Raij et al. (2001). Analyses were performed immediately after sampling or when necessary samples were duly stored under refrigeration.

Data analysis

All data underwent one-way analysis of variance (ANOVA) with the Statistica 10 package, to test differences between seasons (Statsoft, 2011). Differences were considered significant at $p < 0.05$. All results were expressed as means \pm SD (Standard Deviation). Pearson correlation coefficients were calculated among environmental variables. Associated fauna diversity was calculated with the Shannon-Wiener (H') index (Pielou, 1975). Richness (S) was calculated as the total number of species present and evenness or equitability (E) was determined as H/H_{max} , where H is the Shannon-Wiener index and $H_{max} = \ln S$. Species dominance (D) and abundance (A) were analyzed for organisms associated to macrophyte. Species were considered dominant when the density was higher than 50% of the total number of specimens present in the sample; they were abundant when the number of specimens was higher than the mean density of all occurring species (Lobo and Leighton, 1986).

Table 1: Water quality in the constructed wetland: Mean \pm SD in the rainy and dry seasons * and list of variables significantly correlated with each variable **.

Variables	Rainy season	Dry season	Variable positively correlated with	Variable negatively correlated with
temperature (°C)	25.4 \pm 1.6 ^a	20.8 \pm 1.8 ^b	TSS	cond, DO, NO ₃ , TDS
pH	6.7 \pm 0.1 ^a	6.8 \pm 0.5 ^a	NO ₃	PO ₄ , Tot P
Conductivity (μ S. cm ⁻¹)	93 \pm 19 ^a	105 \pm 15 ^a		tem
turbidity (NTU)	21.6 \pm 15.1 ^a	20.2 \pm 5.6 ^a	colif, chlor	
alkalinity (mg. l ⁻¹)	54 \pm 6.0 ^a	50 \pm 3.7 ^a		
DO (mg. l ⁻¹)	2.9 \pm 1.3 ^b	5.3 \pm 1.9 ^a	NO ₃	tem
TAN (μ g. l ⁻¹)	550 \pm 167 ^a	818 \pm 615 ^a	NO ₂ , PO ₄ , Tot P	

Table 1 (continued): Water quality in the constructed wetland: Mean \pm SD in the rainy and dry seasons* and list of variables significantly correlated with each variable**.

Variables	Rainy season	Dry season	Variable positively correlated with	Variable negatively correlated with
NO ₂ ($\mu\text{g. l}^{-1}$)	11.5 \pm 6.7 ^a	13.9 \pm 6.5 ^a	TAN, Tot P	
NO ₃ ($\mu\text{g. l}^{-1}$)	37.8 \pm 31 ^b	150 \pm 95 ^a	pH, DO, TDS	tem
PO ₄ ($\mu\text{g. l}^{-1}$)	19.8 \pm 9.0 ^a	48 \pm 68 ^a	TAN, Tot P	pH
Total P ($\mu\text{g. l}^{-1}$)	38.6 \pm 19 ^a	86 \pm 125 ^a	TAN, NO ₂ , PO ₄	pH
BOD (mg. l ⁻¹)	57 \pm 54 ^a	83 \pm 47 ^a	TDS	
TSS (mg. l ⁻¹)	12.6 \pm 4.1 ^a	8.4 \pm 2.9 ^a	tem, colif, chlor	TDS
TDS (mg. l ⁻¹)	180 \pm 57 ^a	364 \pm 188 ^a	NO ₃ , BOD	tem, TSS
coliforms (MPN)	19.512 \pm 41.644 ^a	542 \pm 462 ^a	turb, TSS, chlor	
chlorophyll- <i>a</i> ($\mu\text{g. l}^{-1}$)	80 \pm 76 ^a	39 \pm 22 ^a	turb, TSS, colif	

* In each row, means followed by the same letter do not significantly differ ($p < 0.05$).

** Pearson Correlation at least $p < 0.05$.

RESULTS

The water quality parameters (Tab. 1) showed significant differences between seasons only for temperature, dissolved oxygen and nitrate. Water temperature was higher in the rainy summer, while oxygen and nitrate were higher in the dry winter. There were significant positive correlations among TSS, coliforms, chlorophyll-*a* and turbidity, which indicates that phytoplankton and bacteria formed an important portion of the particles flowing in the wetland water. There were significant positive correlations among TAN, nitrite, phosphate and total phosphorus, which indicates that decomposition processes strongly affected water quality in the wetland. The lack of correlation among oxygen, pH and chlorophyll-*a* indicates that photosynthesis by phytoplankton had little effect on the water quality flowing through the wetland, at least as compared with the strong effect of decomposition.

In the sediment, significant differences between seasons were observed for organic matter, Ca and N, which were higher during in the dry season, and for K, which was higher during the rainy season (Tab. 2). The pH of the sediment was acidic and the highest nutrient concentration was observed for Ca in the dry season (Tab. 2).

E. crassipes reached 50-55 cm total length (rizoma + aerial plant) in 60 days of growth, in either the rainy or in the dry season (Fig. 2). Nutrients in the aquatic plants biomass were much higher than those observed in the sediment, indicating incorporation of these nutrients in their growth. Nutrients in the plant biomass were higher during the dry season, except for Mg, Fe and Zn. The highest nutrient concentrations in the aquatic plants were of K. The opposite occurred with K in the sediment, which was below 0.1 g.m⁻² (Tab. 2). Plants wet biomass was higher during the dry season, whereas plants dry biomass was similar in both seasons (Tab. 2).

Table 2: Mean and SD concentrations * of variables measured in sediment and *E. crassipes* plants during the rainy and dry seasons.

Variables	Rainy season	Dry season
Sediment (g. dm⁻³)		
N	0.64 ± 0.19 ^b	1.08 ± 0.33 ^a
P	0.08 ± 0.03 ^a	0.09 ± 0.01 ^a
K	0.06 ± 0.001 ^a	0.05 ± 0.002 ^b
Ca	1.81 ± 1.19 ^b	2.20 ± 0.51 ^a
Mg	0.24 ± 0.10 ^a	0.24 ± 0.04 ^a
Fe	0.11 ± 0.01 ^a	0.12 ± 0.02 ^a
Zn	0.003 ± 0.002 ^a	0.006 ± 0.003 ^a
Organic Matter	15.0 ± 3.9 ^b	29.8 ± 9.4 ^a
pH	5.2 ± 0.5 ^a	5.4 ± 0.2 ^a
Plant (g. m⁻²)		
N	8.3 ± 4.9 ^b	10.4 ± 6.2 ^a
P	1.0 ± 0.7 ^b	1.5 ± 1.1 ^a
K	24.0 ± 18.8 ^b	30.6 ± 18.8 ^a
Ca	4.5 ± 2.6 ^b	5.0 ± 2.7 ^a
Mg	1.5 ± 0.8 ^a	1.7 ± 0.7 ^a
Fe	3.9 ± 2.4 ^a	1.6 ± 0.9 ^b
Zn	0.05 ± 0.03 ^a	0.05 ± 0.03 ^a
Wet mass	7,698 ± 4,880 ^b	9,875 ± 5,709 ^a
Dry mass	339 ± 185 ^a	368 ± 184 ^a

* In each row, means followed by the same letter do not significantly differ ($p < 0.05$)

The *E. crassipes* associated zooplankton community comprised 51 taxa, 43 Rotifera, three Cladocera, four Copepoda, and one Ostracoda. Insecta consisted of six orders and Protozoa comprised seven genera. The phyla Nematoda, Mollusca and Platyhelminthes had low contribution. Diversity and species abundance were higher in the rainy season (Tab. 3) due to the contributions of allochthonous material from the fish ponds. Total density was 33% lower in the dry compared with the rainy season. The most abundant organisms were Protozoa, among which Ciliophora comprised over two thirds of the total organisms collected, followed by Rotifera with 22% of the total organisms in the rainy season and 14% in the dry season (Fig. 3). *Vorticella* sp. was the only dominant species in both seasons. *Arcella* sp. (Protozoa), *Proales doliare*s and *Lecane bulla* (Rotifera) were abundant in both seasons (Tab. 4). During the dry season only Rotifera and Protozoa occurred in the *E. crassipes* associated fauna, with dominance of *Vorticella* sp. Species of Cladocera were abundant or present during the rainy season, albeit absent in the dry season. The opposite was observed for the adult and nauplii Copepoda *Argyrodiaptomus furcatus*. The associated fauna tended to have high zooplankton species richness (above 46 species), evenness (above 0.66) and diversity (Shannon-Wiener index with 3.45 and 2.44 nats ind⁻¹) during the rainy and the dry seasons (Tab. 3).

Table 3: Volume of associated fauna washed out from the *E. crassipes* growing in a 0.18 m² area (range), and average density *, species richness, evenness and Shanon-Wiener diversity (H') index in each season.

	Rainy season	Dry season
Associated fauna volume (ml)	1,440-1,820	1,160-1,600
Zooplankton total	1,107,500	413,500
Cladocera (ind. m ⁻²)	109,800	0
Copepoda (ind. m ⁻²)	24,000	800
Ostracoda (ind. m ⁻²)	9,700	1,500
Rotifera (ind. m ⁻²)	964,000	411,200
Richness	51	46
Evenness	0.88	0.66
Diversity (H')	3.45	2.44
Protozoa total (ind. m⁻²)	3,287,000	2,558,000
Amoebozoa (ind. m ⁻²)	152,000	120,000
Rhizaria (ind. m ⁻²)	72,000	53,000
Ciliophora (ind. m ⁻²)	3,063,000	2,385,000
Richness	8	7
Evenness	0.12	0.16
Diversity (H')	0.46	0.60
Insecta total	4,654	90
Collembola (ind. m ⁻²)	3	0
Coleoptera (ind. m ⁻²)	57	8
Diptera (ind. m ⁻²)	4,500	25
Hemiptera (ind. m ⁻²)	6	30
Neuroptera (ind. m ⁻²)	0	1
Odonata (ind. m ⁻²)	88	26
Richness	6	5
Evenness	0.37	0.25
Diversity (H')	1.42	0.92
Others total	3,936	3,550
Nematoda (ind. m ⁻²)	3,600	2,300
Mollusca (ind. m ⁻²)	36	850
Platyhelminthes (ind. m ⁻²)	300	400
Richness	3	3
Evenness	0.09	0.24
Diversity (H')	0.36	0.89

* ind. m⁻² of *E. crassipes*.

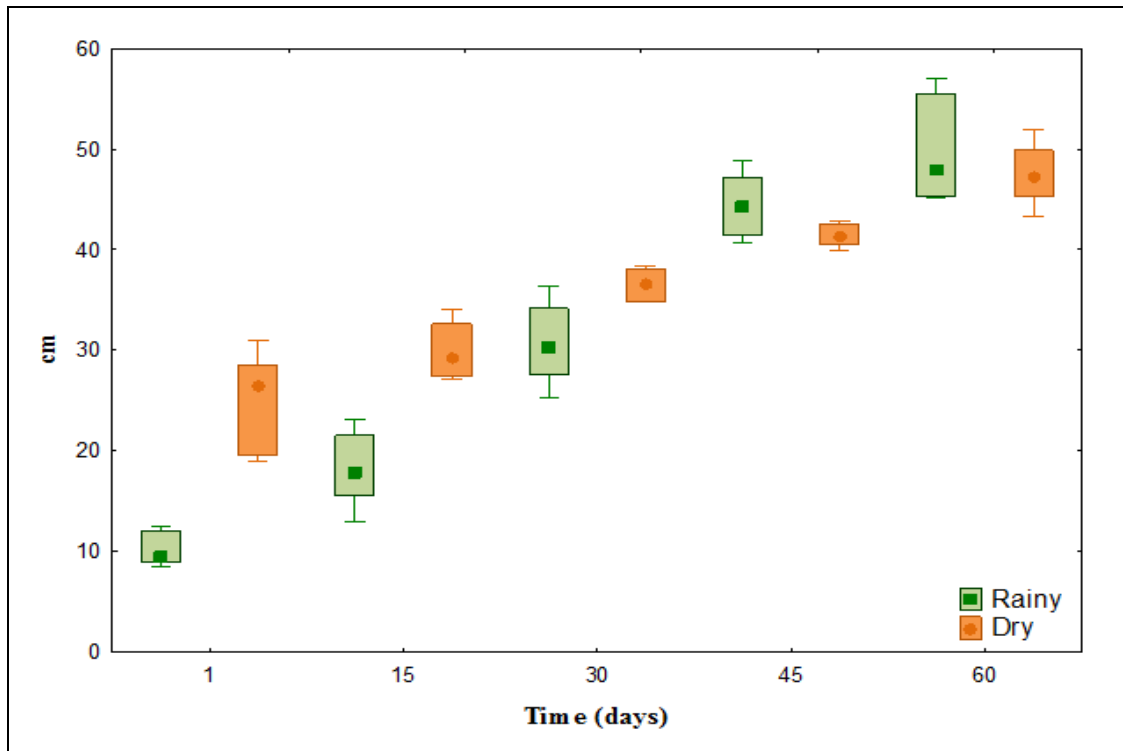


Figure 2: Total length (rizoma + aerial plant) growth of *Eichhornia crassipes* during the rainy and dry seasons. Measurements are mean \pm SD of 12 plants.

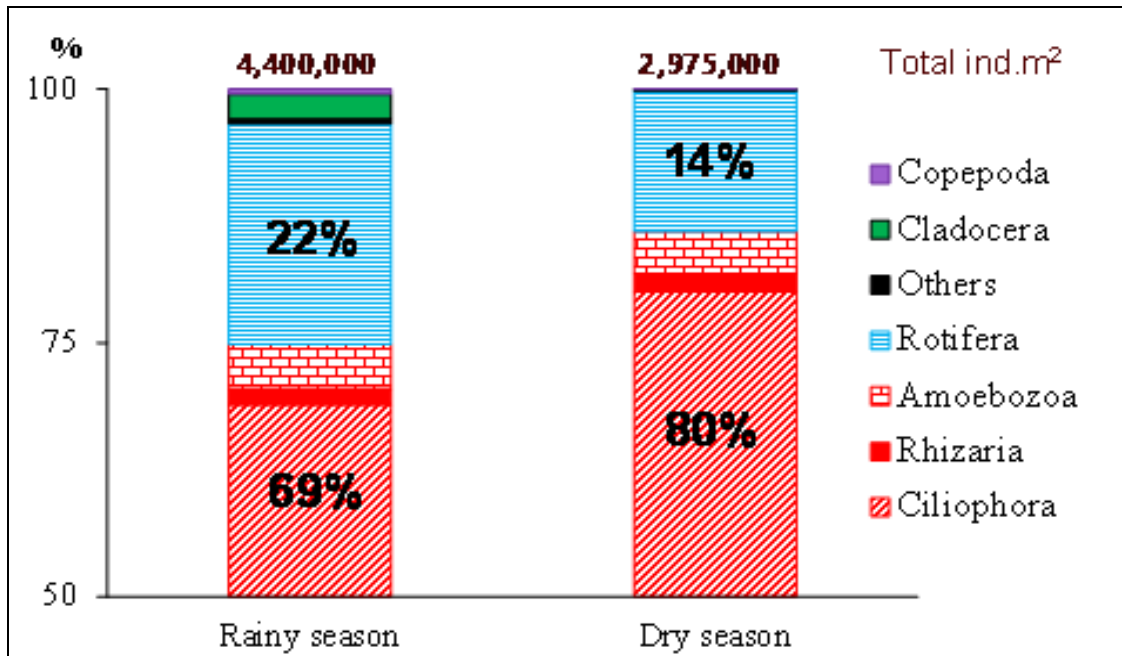


Figure 3: Relative abundance of *E. crassipes* associated fauna and average total density recorded in the rainy and dry seasons. Note that the x-axis starts at 50%.

Others = Insecta, Ostracoda, Nematoda, Mollusca and Platyhelminthes.

Table 4: Frequency of occurrence * of associated fauna to *E. crassipes* in the rainy and dry seasons.

Taxa	Rainy season	Dry season
Cladocera		
<i>Alona guttata</i> (Sars, 1862)	A	–
<i>Ilyocryptus spinifer</i> (Herrick, 1882)	A	–
<i>Diaphanosoma birgei</i> (Korinek, 1981)	+	–
Copepoda		
<i>Thermocyclops decipiens</i> (Kiefer, 1929)	+	+
<i>Thermocyclops minutus</i> (Lowndes, 1934)	+	+
<i>nauplii</i>	A	+
<i>Argyrodiaptomus furcatus</i> (Sars, 1901)	–	+
<i>nauplii</i>	–	+
Harpacticoida	+	+
Ostracoda	+	+
Rotifera		
<i>Ascomorpha ovalis</i> (Bergendal, 1892)	–	A
<i>Ascomorpha saltans</i> (Bartsch, 1870)	+	+
<i>Asplanchnopus</i> sp. (Guerne, 1888)	+	A
<i>Brachionus falcatus</i> (Zacharias, 1898)	+	–
<i>Brachionus calyciflorus</i> (Pallas, 1766)	–	+
<i>Cephalodella misgurnus</i> (Wulfert, 1937)	+	+
<i>Cephalodella gibba</i> (Ehrenberg, 1830)	+	A
<i>Collotheca mutabilis</i> (Hudson, 1885)	+	+
<i>Collotheca ornata</i> (Ehrenberg, 1832)	+	+
<i>Colurella obtusa</i> (Gosse, 1886)	A	+
<i>Colurella uncinata</i> (Müller, 1773)	A	+
<i>Conochilus</i> sp. (Ehrenberg, 1834)	–	+
<i>Dipleuchlanis propatula</i> (Gosse, 1886)	–	+
<i>Euchlanis dilatata</i> (Hauer, 1930)	+	A
<i>Epiphanes macrourus</i> (Barrois and Daday, 1894)	+	+
<i>Filinia opoliensis</i> (Zacharias, 1898)	+	–
<i>Heterolepadella heterostyla</i> (Murray, 1914)	+	+
<i>Keratella cochlearis</i> (Gosse, 1851)	–	+
<i>Keratella tropica</i> (Apstein, 1907)	–	+
<i>Lecane aculeata</i> (Jakubski, 1912)	+	+
<i>Lecane bulla</i> (Grosse, 1851)	A	A
<i>Lecane calcaria</i> (Harring and Myers, 1926)	–	+
<i>Lecane cornuta</i> (Müller, 1786)	+	+
<i>Lecane curvicornes</i> (Murray, 1913)	+	+
<i>Lecane furcata</i> (Murray, 1913)	+	+
<i>Lecane hornemanni</i> (Ehrenberg, 1834)	+	+
<i>Lecane inermis</i> (Bryce, 1892)	–	+
<i>Lecane ludwigii</i> (Eckstein, 1883)	+	+
<i>Lecane lunaris</i> (Ehrenberg, 1832)	A	+

Table 4 (continued): Frequency of occurrence * of associated fauna to *E. crassipes* in the rainy and dry seasons.

Taxa	Rainy season	Dry season
<i>Lecane monostyla</i> (Daday, 1897)	+	+
<i>Lecane proiecta</i> (Hauer, 1956)	+	+
<i>Lecane signifera</i> (Jennings, 1896)	+	+
<i>Lepadella</i> sp. (Bory de St. Vincent, 1826)	+	+
<i>Philodina</i> sp. (Ehrenberg, 1830)	+	+
<i>Proalinopsis caudatus</i> (Collins, 1873)	+	A
<i>Proales doliaris</i> (Rousselet, 1895)	A	A
<i>Proales globulifera</i> (Hauer, 1921)	+	+
<i>Ptygura fucillata</i> (Kellicott, 1889)	–	+
<i>Synchaeta stylata</i> (Wierzejski, 1893)	–	+
<i>Testudinella patina</i> (Hermann, 1783)	+	+
<i>Testudinella</i> sp.	+	+
<i>Trichocerca cavia</i> (Gosse, 1886)	+	+
<i>Trichocerca similis</i> (Wierzejski, 1893)	+	+
Protozoa		
Amoebozoa		
<i>Arcella</i> sp.	A	A
<i>Centropyxis</i> sp.	+	+
<i>Diffflugia</i> sp.	+	–
<i>Euplotes</i> sp.	–	+
Rhizaria		
<i>Euglypha</i> sp.	A	+
Ciliophora		
<i>Paramecium</i> sp.	A	+
<i>Vorticella</i> sp.	D	D
Insecta		
Collembola	+	–
Coleoptera	+	+
Diptera	+	+
Hemiptera	+	+
Neuroptera	–	+
Odonata	+	+
Others		
Nematoda	+	+
Mollusca	+	+
Platyhelminthes	+	+

* + = present; – = absent; A = abundant; D = dominant.

DISCUSSION

The water quality in the constructed wetland was very variable during each studied period, so that most of the environmental parameters measured did not show significant differences between the rainy and the dry seasons. The rainy season is during summer and the dry one during winter, which accounts for the water temperature differences between seasons. The significantly higher oxygen and nitrate concentrations in the dry than in the rainy seasons are at least partially accounted for by differences in the hydrological regime during both seasons. The rainy season is also the main fish culture period in the aquaculture farm, so that the constructed wetland received, together with rain and surface runoff, increased amounts of effluents (with organic matter that consumed oxygen). During this period, the water flow and turbulence inside the wetland were strong and the water outflow contained large amounts of particles. Under these conditions nitrification was reduced due to loss of nitrifying bacteria attached to the washed out particles, resulting in the observed reduced nitrate concentration in the water. Reduced nitrification related to washout were already observed in flow-through fish culture systems with high water exchange rates (Diab et al., 1992; Milstein et al., 2001) and even in stagnant fishponds flooded and washed out by heavy monsoon rains (Milstein et al., 2002). Together with this, numerous insects and rather large aquatic organisms that can withstand water turbulence and washout (crustaceans, mollusks, and helminths) were abundantly found in the *E. crassipes* associated fauna. On the contrary, during the dry season less effluents and few surface runoff entered the wetland (less oxygen consumption) so that water flow and turbulence in the constructed wetland were low allowing sedimentation of most particles and reducing particles washout from the system. Under these conditions nitrifying bacteria remained in the wetland, increasing nitrate levels in the water, as well as the small organisms (protozoa and rotifers) that dominated in the *E. crassipes* associated fauna.

In this flow-through system, phytoplankton entering with the effluents encountered an unfavorable environment within the wetland due to macrophytes shadowing and competition for nutrients, and also were rather quickly washed out. Thus, phytoplankton photosynthesis had little effect on the wetland water quality, as indicated by the lack of correlation among oxygen, pH and chlorophyll-*a*. The high loading of organic matter entering with the aquaculture ponds and UASB effluents and particle trapping by the macrophytes should have favored decomposition processes, as indicated by the significant positive correlations among the decomposition products TAN, nitrite and phosphate recorded in the water sampled among the macrophytes. The thermotolerant coliforms get it these effluents should have fostered a detrital food net where tiny in size rotifers, protozoa, and cyclopoid copepod nauplii have a successful competing favored position in this circumstance.

Protozoa, the most abundant organisms in this study, are widespread in wetland rhizospheres where feeding on roots and microorganisms is enhanced (Neori and Agami, 2016). Arcellidae is one of the predominant families, coupled with Diffugiidae and Centropxyidae in the community structures of wetland environments (Lansac-Tôha et al., 2014). High dominance of *Vorticella* sp. during the sampled period shows that the free-living ciliophorans are commonly encountered in fish ponds. However, when fish are stressed by adverse environmental conditions, the ciliophorans become a facultative ectoparasite (Abdel-Baki et al., 2014). According to Mieczan (2007), populations of Protozoa in aquatic environments are directly related to the presence of suspended solids produced by fast life cycle macrophytes, such as *E. crassipes*.

Rhizosphere invertebrates function ecologically as Protozoa in the consumption of exudates, biota, flora and root tissue, by modifying nutrients and influencing plant growth (Neori and Agami, 2016). The continuous growth of the plants until the end of each sampled period provided a large abundance of associated organisms, mainly during the rainy season.

E. crassipes provides an extra habitat for macro- and micro-invertebrates due to its large radicular system available in the water column, which traps particles and is used as both food and substrate (Chowdhary and Sharma, 2013). The observed large diversity, richness and evenness of zooplankton in the *E. crassipes* associated fauna were mainly related to rotifer species. Rotifers predominate in environments with human activities since they reproduce and rapidly adapt themselves to environmental changes (Dahms et al., 2011). The large abundance of rotifer species recorded is related to the large water volumes containing them, which the wetland received from several aquaculture ponds (Sipaùba-Tavares and Dias, 2014).

E. crassipes submerged and areal parts provide appropriate habitat and trophic resources for insects. At least for Diptera and Odonata assemblages, it was reported that the aquatic plants support assemblages of these insects, abundance and richness of which depend on the abundance and distribution of the vegetation (Capello et al., 2013).

The ecological indexes were higher in the rainy season for zooplankton and Insecta as a result of high water flow from the different fish ponds during that period. This occurred because the aquaculture farm has a continuous water flow of which about 60% of the water volume goes to the wetland under analysis. However, the diversity and evenness of Protozoa species were higher during the dry season when the water volume was lower and species stabilized at the *E. crassipes* roots. After 60 days, plants in the wetland had to be removed due to lack of space for their development. Consequently, maximum residence time of the plant in this wetland should be approximately 60 days, with complete regular harvest to maintain the retention and incorporation of water nutrients into the plants biomass.

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