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# Impacts of composite wastewater on a Pampean stream (Argentina) and phytoremediation alternative with *Spirodela intermedia* Koch (Lemnaceae) growing in batch reactors

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## ABSTRACT

Poultry slaughterhouses are producing increasing amounts of solid organic by-products and waste in Argentina. The objectives of this study are to assess the impacts of wastewater, mainly composed of poultry slaughterhouse effluent, on a Pampean stream and to evaluate the convenience of phytoremediation alternative with *Spirodela intermedia*. Impacts over La Chozza stream (Buenos Aires, Argentina) were evaluated by means of physical and chemical characterization of the water upstream and downstream input. Nutrients and total suspended solids increased downstream of the open channel outlet. A bioassay was performed in order to evaluate the role of *S. intermedia* in wastewater treatment. Two treatments were performed in batch reactors during 144 h (6 days), with (treatment A) and without (treatment B) the addition of *S. intermedia*. Water samples were taken at 8, 24, 48, 96 and 144 h after the start of the assay for the determination of ammonium, nitrite, nitrate, soluble reactive phosphorus and total phosphorus. Since removal rates of ammonium and total phosphorus were significantly higher in treatment A than in treatment B and the nitrification process was highly increased in treatment A, then the species could be used for the phytoremediation of composite wastewater.

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## 1. Introduction

Argentina has become the seventh largest producer of chicken meat (1625 million tons) and the fifth largest exporter (200,000 tons) in the world in 2010 (SIIA, 2012). As a result of this growing industry, poultry slaughterhouses are producing an increasing amount of solid organic by-products and waste. Slaughterhouse industries are ubiquitous in the Pampas region and usually discharge large volumes of wastewater. Poultry slaughterhouse wastewater is characterized by a high biological and chemical oxygen demand and large amounts of nitrogen and phosphorus (Debik and Coskun, 2009). Residual blood, fat from skin, grease exuded during scalding for feather removal and immersion chilling and feces are the main sources of organic matter in this wastewater. Residual blood, urine and feces are also significant sources of nitrogen. The phosphorus contained in this wastewater derives from blood, manure, and cleaning and sanitation compounds (USEPA, 2002). An oversupply of either nutrient can cause highly undesirable changes in the structure and function of aquatic

ecosystems that receive wastewater, which contributes to the eutrophication of water bodies.

Aquatic ecosystems are mostly threatened by small fluctuations in pH, temperature, dissolved oxygen and nutrient concentrations that lead to notable instability in the biological communities. Moreover, Pampean streams are characterized by low velocity currents (Feijoó and Lombardo, 2007) and therefore by high residence times. Hence, when elements with no gaseous form (e.g. phosphorus) reach streams, they are transported slowly downstream. For elements with a gaseous form at ambient temperature, substantial conversion can occur in streams and so they are permanently exported from the water into the atmosphere. On the other hand, between 16 and 50% of nitrate and more than 50% of the fixed carbon that enter streams and rivers are lost from the ecosystem as N<sub>2</sub> and CO<sub>2</sub> respectively (Doyle and Bernhardt, 2011). Dodds and Oakes (2008) attributed a much stronger effect of land use in first-order streams on the downstream water quality than of the whole-watershed land cover, because these streams provide the predominant hydrologic contributions to the watershed (Lowrance et al., 1997). Also, substantial in-stream nutrient processing and retention in upland streams and rivers can regulate the downstream water quality (Alexander et al., 2000; Peterson et al., 2001).

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Floating and submerged aquatic macrophytes have a high capacity to improve water quality by removing high loads of nutrients and heavy metals; furthermore, the high productivity of these aquatic plants has encouraged their use for wastewater treatment and resource recovery (Srivastava et al., 2008; Ranieri, 2012; Ranieri and Young, 2012). Floating aquatic macrophytes take up and retain nitrogen and phosphorus from the water in their tissues (Srivastava et al., 2008). Due to their small size, high growth rates, high nutrient requirements and high protein content (Vermaat and Hanif, 1998) many species of Lemnaceae represent an interesting alternative to other hydrophytes for the biological treatment of wastewater. Vermaat and Hanif (1998) concluded that *Lemna gibba* L. and *Spirodela polyrhiza* Schleid. play a substantial role in nutrient removal from wastewater, particularly phosphorus. For nitrogen removal, the authors attribute greater importance to the volatilization of  $\text{NH}_3$  and the action of denitrifying bacteria.

The objectives of this study are to assess the impacts of wastewater, mainly composed of poultry slaughterhouse effluent, on La Choza stream and to evaluate the convenience of phytoremediation alternative with duckweed, *Spirodela intermedia* W. Koch (Lemnaceae), a common species that has been reported in lentic water bodies in the Pampas (Argentina) (Feijoó and Lombardo, 2007; Miretzky et al., 2004) and other regions of Central and South America (DARWINION, 2011; Mazzeo, 1993).

## 2. Material and methods

### 2.1. Study area

The Pampean region is characterized by flat landscape (0.1–1 m/km); plains composed of Quaternary sediments and a drainage network with high densities of streams and rivers (Rodrigues Capítulo et al., 2010). Climate of the region is characterized as temperate humid, according to Köppen's classification, with an annual mean rainfall of 1055 mm and minimum and maximum temperatures of 11.1 °C (July) and 24.3 °C (January) respectively (Arreghini et al., 2005). Pampean streams that are relatively undisturbed can be described in relation to their physical and chemical characteristics, according to Feijoó and Lombardo (2007), as alkaline, with a high electrical conductivity (EC), a high concentration of dissolved oxygen (DO) and high irradiance levels due to the lack of a forested riparian zone (Feijoó et al., 2011).

The Reconquista River is a lowland river in the Pampean region which starts at the confluence of La Choza and Durazno streams (Fig. 1). La Choza sub basin covers 555 km<sup>2</sup>. The selected section of La Choza stream is a second-order stream according to the Strahler classification (Gordon et al., 1994). La Choza stream flows into the Roggero reservoir, which is part of a protected natural area (Fig. 1).

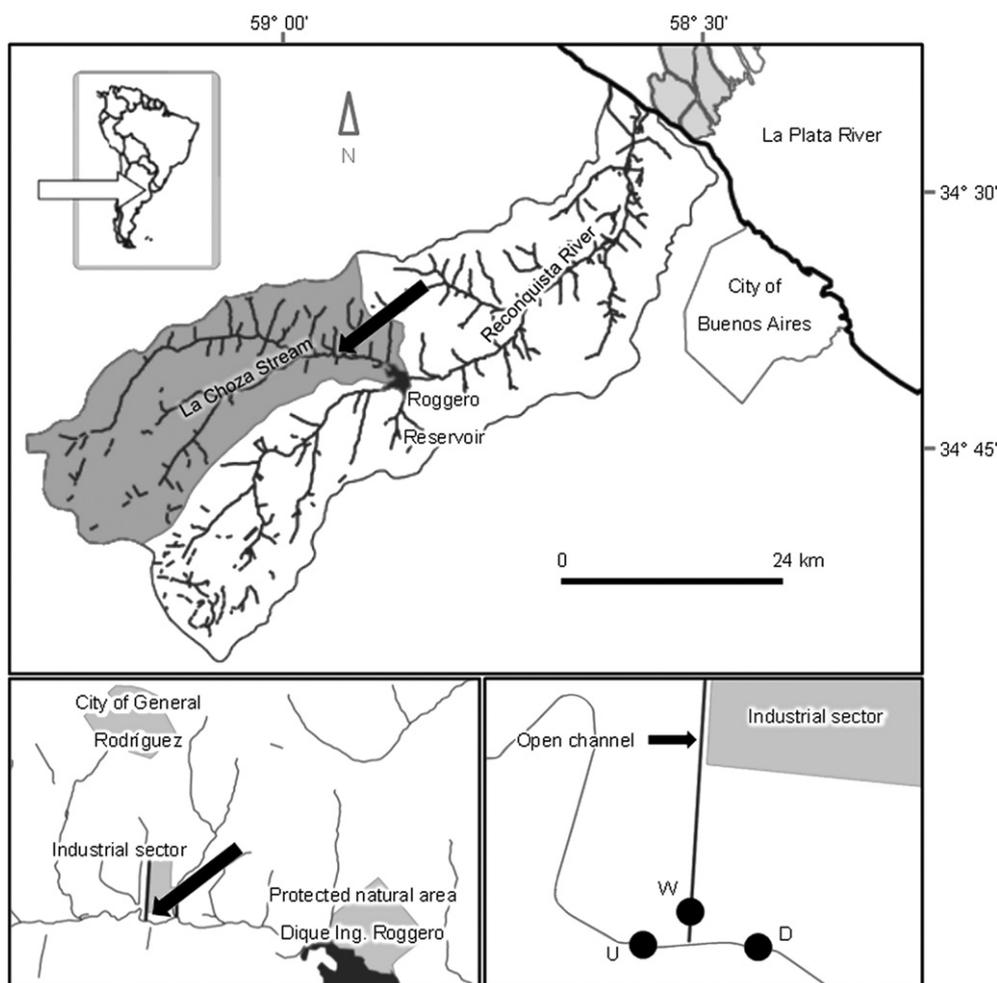


Fig. 1. Location of the open channel and sampling sites: wastewater (W), upstream (U) and downstream (D) of the channel outlet.

## 2.2. Sampling and water characterization

Water samples were taken in triplicate for chemical analysis from three sites: an open channel (site W) that flows into La Choza stream and receives wastewater from poultry slaughterhouse facilities; 50–100 m upstream of site W (U) and downstream (D) of the channel outlet (Fig. 1). Samples were extracted on four dates between May 2010 and February 2011 corresponding to each season and were taken at least 48 h after the last rain, to avoid any runoff effect from the agricultural and livestock activities in the area. Subsamples were preserved with H<sub>2</sub>SO<sub>4</sub> for total phosphorus (TP) and total organic carbon (TOC) determinations or filtered through Whatman GF/C filters and kept at 4 °C for other determinations. Dissolved organic carbon (DOC) was determined by digestion of filtered acid-preserved samples. Particulate organic carbon (POC) was estimated from TOC–DOC differences. Variables were determined according to the following methods: pH and EC with Hanna™ meters; total ammonium (N-NH<sub>4</sub><sup>+</sup>) by Blue Indophenol according to Mackereth et al. (1989); DO by iodometry, chlorides (Cl<sup>-</sup>) by volumetry with AgNO<sub>3</sub> and biological oxygen demand (BOD<sub>5</sub>) according to APHA-AWWA-WPCF (1992); nitrite (N-NO<sub>2</sub><sup>-</sup>) by diazotization, nitrate (N-NO<sub>3</sub><sup>-</sup>) by hydrazine sulfate reduction and diazotization, soluble reactive phosphorus (SRP) by molybdate ascorbic, and TP by molybdate ascorbic after digestion with H<sub>2</sub>SO<sub>4</sub> and K<sub>2</sub>S<sub>2</sub>O<sub>8</sub> according to Strickland and Parsons (1972); total suspended solids (TSS) by gravimetry, TOC and DOC by acid digestion with H<sub>2</sub>SO<sub>4</sub>/K<sub>2</sub>Cr<sub>2</sub>O<sub>7</sub> solution according to Golterman et al. (1978). The Mann–Whitney *U* test was used to compare concentrations of the main variables measured at sites U and D. Discharges for October 2010 were roughly estimated in the stream and the open channel from the section of the streambed considering depth profile and water velocity, by measuring the time taken by a floating object to cover a known distance downstream according to Gordon et al. (1994). Loads of TSS, ammonium, TP and TOC were calculated by multiplying stream discharge by concentration.

## 2.3. Bioassay

A bioassay was conducted with *S. intermedia* individuals grown in containers with Hoagland medium (Hoagland and Arnon, 1938) in greenhouse. Each of 30 containers, with 433 cm<sup>2</sup> of free surface area, was filled with 1 L of water from site W (Fig. 1) and they were randomly distributed in a controlled temperature (23 ± 1 °C) room.

At the beginning of the bioassay the plant material was rinsed several times with deionized water and then dried on tissue paper for 10 min to measure the fresh weight (FW). Biomass plant (20.1 g FW) was added to each of 15 randomly selected containers and most of the free surface (>90%) was covered to prevent extensive algal growth (Körner and Vermaat, 1998). Also, the sides of the containers were wrapped with an opaque plastic membrane to prevent algal growth. The containers were illuminated with fluorescent light in a 16:8 h (light:dark) photoperiod (treatment A). The remaining 15 containers without plants were kept in the dark to inhibit phytoplankton development (treatment B). Three containers of each treatment were removed after 8, 24, 48, 96 and 144 h from the start of the bioassay and water samples were taken for subsequent determination of nutrients. Nutrient removal rates were calculated using:

$$\%R = 100 \times \frac{V_i \times C_i - V_f \times C_f}{V_i \times C_i} \quad (1)$$

where %R is the removal rate of the variable considered, *V<sub>i</sub>* and *V<sub>f</sub>* are the initial and final volumes of wastewater in each container,

and *C<sub>i</sub>* and *C<sub>f</sub>* are the initial and final concentrations. A one-tail Student's *t*-test was performed to compare %R (at 144 h) between treatments A and B. Prior to analysis, the data were tested for normality and homogeneity of variance using Shapiro–Wilk and Levene tests, respectively (Zar, 1996).

Fresh weights (FW) of plant material were recorded at the beginning and corresponding final time in each container. Also, 20.1 gFW (initial FW) and total biomass at the corresponding final time for each container were dried at 60 °C for 48 h and weighed in order to obtain the dry weight (DW). Relative growth rate (RGR) was calculated by:

$$RGR = \frac{(\ln W_f - \ln W_i)}{\Delta t} \quad (2)$$

where *W<sub>f</sub>* and *W<sub>i</sub>* are the final and initial biomass and Δ*t* is the bioassay duration.

## 3. Results and discussion

### 3.1. Water characterization

High concentrations of ammonium, TSS, DIN, SRP, TP, BOD<sub>5</sub> and low dissolved oxygen values were determined in water from the open channel (W). Ammonium was the main form of DIN representing 98%, while nitrite and nitrate values were low. High ammonium levels could be toxic for fish and potential inhibitor for algal photosynthesis (Guillen-Jimenez et al., 2000).

Phosphorus was mainly found in a dissolved form (60%) (Table 1). Similar values (5.8–12.1 mg/L) for TP concentration were reported by Debik and Coskun (2009) for poultry slaughterhouse wastewater. TSS values were lower than the values shown by these authors (1850–3750 mg/L). Ammonium was similar to some values cited for raw abattoir wastewater (19–20 mg/L; Mittal, 2006) and sewage (25–96 mg/L; Al-Nozaily et al., 2000). EC and BOD<sub>5</sub> were lower than values shown by Chávez et al. (2005) for poultry slaughterhouse wastewater. Considering that the electrical conductivity may be mainly governed by the chloride concentration, and conservative parameters such as chloride are controlled by mineralization processes, the relative stability (CV) in the values of EC (CV = 0.03) and chloride (CV = 0.07) reflected stability in the hydrological conditions of the channel (W) on the sampling dates.

Table 1 shows the characteristics of water measured upstream (U) and downstream (D) of the open channel. Further, at site U

**Table 1**

Characterization of water in the industrial open channel (W) and at sites in La Choza stream, upstream (U) and downstream (D) of the discharge (mean ± SD).

Variable (unit)	Industrial open channel W	La Choza stream	
		U	D
pH	7.48 ± 0.40	8.10 ± 0.24	7.96 ± 0.39
EC (mS/cm)	1.33 ± 0.04	1.21 ± 0.10	1.22 ± 0.07
OD (mg/L)	0.4 ± 0.8	4.0 ± 2.9	2.4 ± 2.4
TSS (mg/L)	198 ± 39	17 ± 6	44 ± 16
N-NH <sub>4</sub> <sup>+</sup> (mg/L)	21.27 ± 4.36	6.64 ± 2.16	7.80 ± 2.06
N-NO <sub>2</sub> <sup>-</sup> (mg/L)	0.05 ± 0.02	0.10 ± 0.04	0.22 ± 0.15
N-NO <sub>3</sub> <sup>-</sup> (mg/L)	0.08 ± 0.05	0.73 ± 0.35	1.63 ± 2.29
DIN (mg/L)	21.67 ± 4.73	7.65 ± 2.68	9.55 ± 2.65
SRP (mg/L)	3.5 ± 1.0	0.9 ± 0.1	1.1 ± 0.3
TP (mg/L)	5.8 ± 0.9	1.3 ± 0.2	1.9 ± 0.5
Cl <sup>-</sup> (mg/L)	84 ± 6	64 ± 5	65 ± 5
BOD <sub>5</sub> (mg/L)	197 ± 19	23 ± 26	49 ± 23
TOC (mg/L)	121 ± 41	7 ± 1	12 ± 4
DOC (mg/L)	49 ± 24	4 ± 1	6 ± 1
POC (mg/L)	72 ± 19	3 ± 1	7 ± 3

mean concentrations of ammonium and SRP were higher than the mean values reported by Feijóo and Lombardo (2007) in a study of 41 Pampean streams and by Vilches et al. (2011) for upper sections of La Chozza stream subjected to diffuse pollution. Point sources located upstream of site U (i.e. municipal wastewater from the city of General Rodríguez; Schreiber, 2010) (Fig. 1) seems to greatly affect stream water quality. Variability of BOD<sub>5</sub> measurements was high in the site U due to the different water quality and magnitude of these upstream-located inputs. Arreghini et al. (2008) also found high levels of ammonium and SRP and low levels of OD, nitrites and nitrates at a sampling station in the proximity of site U.

Total suspended solids was significantly higher ( $p < 0.05$ ) downstream of the open channel outlet. Ammonium, nitrite, DIN, SRP, TP, BOD<sub>5</sub>, TOC, DOC and POC were increased while pH and OD decreased downstream of the open channel outlet in terms of mean values (Table 1). The EC, nitrate and chloride mean values did not follow a clear pattern downstream in respect to upstream of the open channel outlet. In the model proposed by Seeboonruang (2012), the authors found that total dissolved solids and EC are in agreement with the water quality when taking land use into account. However, in the present study EC did not reflect the land use. Nitrogen and phosphorous compounds, TSS, BOD<sub>5</sub>, and organic carbon compounds were the water quality parameters that were mainly affected by the wastewater input. Sabater et al. (1990) showed that nutrient content was influenced not only by climate, soil and vegetation but also by variables related to human activities.

A relevant feature is that the spatial dynamics of nutrients might be affected by point-sources inputs. Merseburger et al. (2011) found that uptake lengths ( $S_w$ ) of ammonium and phosphate were increased by four and five times respectively, downstream of a wastewater treatment plant in a Mediterranean stream. On the other hand, inputs of DOC have been reported as a factor that modifies N dynamics in a forested stream, decreasing  $S_w$  for ammonium and nitrates (Bernhardt and Likens, 2002).

### 3.2. Bioassay

In the culture, higher water losses occurred in the treatments with plants due to evapotranspiration and so these losses were taken into account in subsequent estimations. Fronds of *S. intermedia* had a uniform green color in all containers revealing no symptoms of toxicity. The pH increased along time in both treatments, from 7.63 at the beginning to  $8.30 \pm 0.02$  and  $8.60 \pm 0.02$  for treatments A and B, respectively, at the end of the bioassay. The increase in pH values created an optimal condition for the volatilization of ammonia, especially in treatment without plants. The average values of EC over time were in the range of 1.21–1.41 mS/cm for treatment A and 1.22–1.37 mS/cm for treatment B.

Fig. 2 shows the concentrations of N-NH<sub>4</sub><sup>+</sup>, N-NO<sub>2</sub><sup>-</sup>, N-NO<sub>3</sub><sup>-</sup>, DIN, SRP and TP determined in the bioassay. First-order kinetic equations were used to model ammonium concentrations over time in

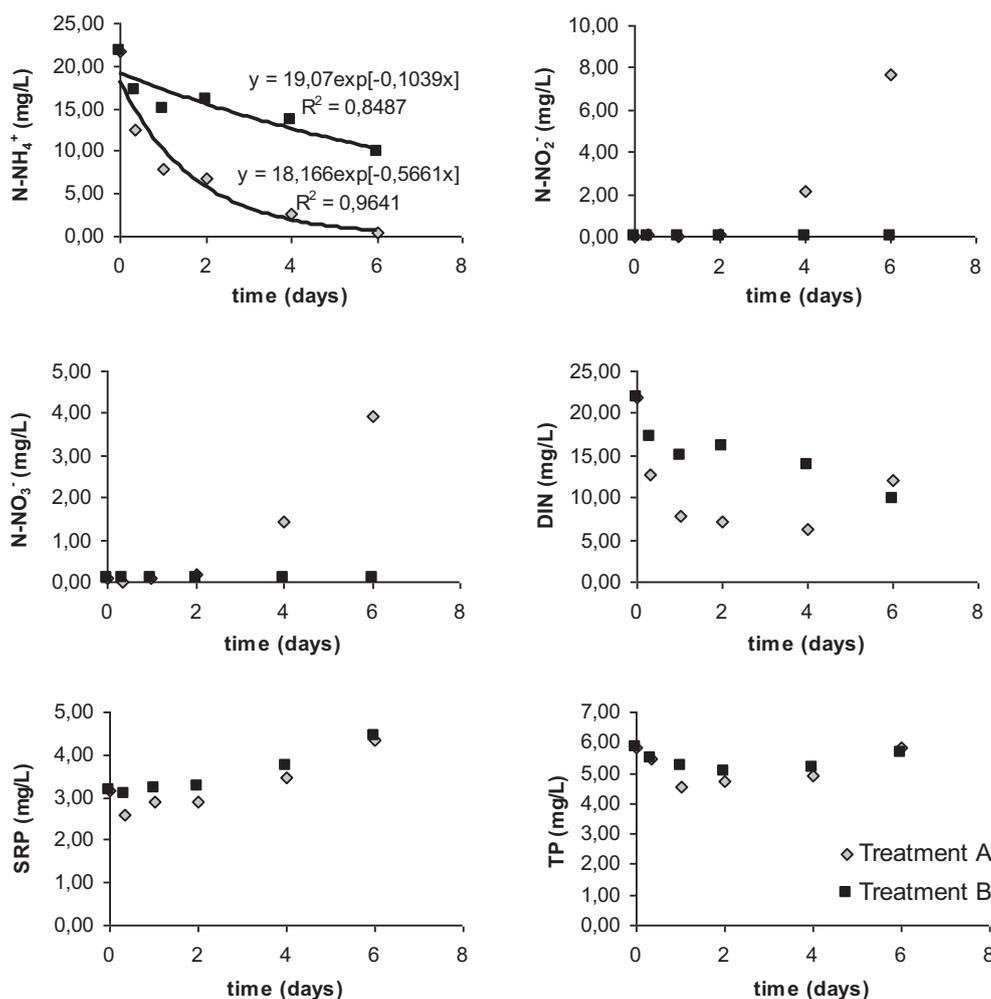


Fig. 2. Mean concentrations of N-NH<sub>4</sub><sup>+</sup>, N-NO<sub>2</sub><sup>-</sup>, N-NO<sub>3</sub><sup>-</sup>, DIN, SRP and TP in both treatments, A and B ( $n = 3$ ). First-order kinetics model were calculated for N-NH<sub>4</sub><sup>+</sup>.

both treatments. Table 2 shows the removal rates of some of the nutrients measured after 6 days (144 h). The final mean concentrations of ammonium, DIN, TP, TOC, DOC and POC decreased in respect to the initial values in both treatments. Removal rates of ammonium and TP were significantly ( $p < 0.05$ ) higher in the treatment with plants. No significant differences for DIN, TOC, DOC and POC removal rates were found between treatments (Table 2).

The removal rates of ammonium and DIN were higher in the first 24 h of the bioassay for both treatments but they were higher in treatment A due to uptake by the plants. DIN concentration in treatment A decreased up to 96 h, when the lowest concentration was observed. The ammonium concentration until fourth day of the assay was higher than 5 mg/l. After that, ammonium decreased and nitrates and nitrites increased due to nitrification (Fig. 2).

At the end of the assay, ammonium removal in treatment A was almost complete (Table 2) and very high increases in nitrite and nitrate were achieved (Fig. 2). Reddy et al. (1990) observed an increase in the oxygen level after the culture of aquatic plants in domestic wastewater due to the exchange of oxygen from aerial tissue into root zone. So DO levels might achieve higher values, in the treatment with plants than in the treatment without plants. The oxidized rhizosphere so formed and the presence of nitrogen-fixing bacteria in the attached biota favors nitrification. Moreover, ammonia volatilization does not seem to occur in the treatment with plants. El-Shafai et al. (2007) did not find any ammonium volatilization in duckweed ponds under similar conditions. The cover floating vegetation enhanced nitrogen cycling and reduced ammonia volatilization. In contrast, in the treatment without plants, the nitrate and nitrite concentrations were always very low (Fig. 2) and the high pH values found could enhance the process of ammonia volatilization, and this seems to be the main factor of ammonium elimination.

Removal efficiencies of ammonium and DOC by *S. intermedia* were similar to *L. gibba* (El-Shafai et al., 2007). Sooknah and Wilkie (2004) found 99.6% ammonium removal by *Eichhornia crassipes* in a 31-day batch growth. So, ammonium removal recorded in this study shows a higher efficiency of *S. intermedia*, as the ammonium removal levels were comparable to those reported in the literature but the exposure time was much lower.

A decrease of SRP was observed up to 8 h, mainly in treatment A, and then it increased until the end of assay in both treatments, resulting in higher final concentrations (Fig. 2) in connection with the rapid turnover of P. After the first few hours, the system was deficient in the removal of SRP. Minimum TP concentration was reached 24 h after the start of treatment A and after 48 h in treatment B and then the concentrations increased (Fig. 2).

The initial TP retention may be related to phosphorous precipitation, rhizofiltration and uptake by plants. At some threshold,

pollutant removal may become asymptotic (Nairn and Mitsch, 2000). The increase in the values of SRP and TP concentrations at the end of bioassay relates to the effect of decomposition of particulate matter and plant material in the case of the treatment with plants. In treatment B low nitrate levels were recorded. Nitrate may be important for the redox-dependent retention of phosphorus. For instance, Kozerski et al. (1999) found high phosphorus release rates at low nitrate input. In the present study, the absence of nitrate would create a redox potential sufficiently low to release P bound to clay particles. Clay is a major component of the soils and sediments of the region (Giorgi et al., 2005).

According to Gross et al. (1998), the major factor controlling phosphorus dynamics in ponds was adsorption of P by bottom soils and sediments. In the present study the water used was not filtered and suspended solids might play an important role. Resuspension of sediments could affect P dynamics especially in shallow reactors where the effect is more marked. Increased phosphorus release at high pH, as in the present study, has been observed in connection with resuspension events (Søndergaard, 2007).

The total mean concentration of organic carbon decreased in both treatments but was lower in treatment A at the end of bioassay (Fig. 3). Otherwise, POC was the main form of carbon in both treatments and its decrease in both treatments toward the end of the bioassay was related to coagulation and sedimentation processes (Wetzel, 1981) and filtration through the root systems in the treatments with plants (Sooknah and Wilkie, 2004). The greater decrease of DOC in both treatments may be associated with degradation of DOC by saprobic microorganisms in the wastewater. A higher DOC removal rate was found in the treatment with plants due to a higher percentage of microorganisms establishing a symbiotic relationship with the plant roots (Sooknah and Wilkie, 2004).

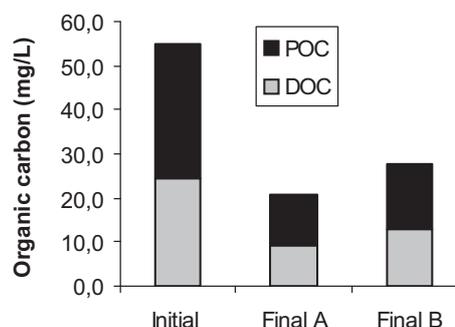
Table 3 shows the changes of the plant biomass (FW and DW), weight gain per day, productivity and RGR values. Estimated values were lower than reported for other Lemnaceae in different types of wastewater in terms of RGR (Landesman et al., 2005; Vermaat and Hanif, 1998). Low growth of plant biomass could be associated with suboptimal lighting conditions or lack of acclimation. Cheng et al. (2002) report low growth rates for *Lemna minor* corresponding to an initial (first 100 h) lag phase, followed by an exponential phase. On the other hand, plant growth may be limited to the first few days because duckweed absorbs ammonium preferentially as a nitrogen source (Caicedo et al., 2000) and the concentration of this nutrient reached low values on the fourth day when nitrogen probably becomes limiting for plant growth. Sooknah and Wilkie (2004) registered a productivity of 1 g/m<sup>2</sup> day, an average growth rate for water lettuce growing in anaerobically digested flushed dairy manure wastewater. These values were similar to those estimated in the present study.

**Table 2**

Removal rates (%R, mean  $\pm$  SD) measured in both treatments: with (A) and without (B) *Spirodela intermedia*.

Variable	Treatment	%R	P-value
N-NH <sub>4</sub> <sup>+</sup>	A	98.1 $\pm$ 0.8	0.002*
	B	57.4 $\pm$ 11.4	
DIN	A	53.0 $\pm$ 11.9	0.344
	B	57.2 $\pm$ 11.4	
TP	A	15.4 $\pm$ 2.3	0.016*
	B	9.7 $\pm$ 2.7	
TOC	A	67.9 $\pm$ 11.6	0.222
	B	60.5 $\pm$ 9.9	
DOC	A	68.5 $\pm$ 12.7	0.085
	B	50.0 $\pm$ 14.2	
POC	A	67.5 $\pm$ 11.7	0.208
	B	55.7 $\pm$ 17.2	

Asterisk (\*) indicates significant differences among %R of both treatments ( $p < 0.05$ , one-tail Student "t" test).



**Fig. 3.** Mean concentrations of organic carbon measured in wastewater at 0 and 6 days in treatments A and B.

**Table 3**  
Changes in biomass of *Spirodela intermedia* in treatment A (mean  $\pm$  SD).

Variable	Biomass				
	Initial biomass (g/m <sup>2</sup> )	Final biomass (g/m <sup>2</sup> )	Weight gain (g/m <sup>2</sup> )	Productivity (g/m <sup>2</sup> day)	RGR (g/g day)
Fresh weight (FW)	464 $\pm$ 0	514 $\pm$ 6	50 $\pm$ 6	8 $\pm$ 1	0.017
Dry weight (DW)	35.6 $\pm$ 0.4	37.9 $\pm$ 0.7	2.2 $\pm$ 0.7	0.4 $\pm$ 0.1	0.010

### 3.3. Integration of field and laboratory results

Bioassay results cannot be directly extrapolated to the *in situ* remediation of effluent given the inherent variability of factors such as climate, hydrology, solids and nutrient levels in the effluent or the eventual presence of phytotoxic contaminants (i.e. heavy metals or pesticides). However, since the presence of *S. intermedia* favors nitrification, and ammonium and TP removal, is justified to test the *in situ* treatment of the effluent. Moreover, a mesocosm study level in semi-field conditions using the same species in continuous flow reactors (Basílico and de Cabo, 2012) showed promising results and comparable to those of this study for the same effluent, with greater plant biomass produced. It should be noted that the demand of nutrients by macrophytes is also usually exponential during the exponential phase of growth of plant biomass (Oscarson et al., 1989). Thus, a greater removal of nutrients would be expected in the *in situ* remediation, where the plant growth is stimulated.

Water discharge estimated in October 2010 was 0.020 m<sup>3</sup>/s for site W and 0.879 and 0.686 m<sup>3</sup>/s for U and D, respectively. Loads of TSS, ammonium, TP and TOC on this date are shown in Table 4. Apparently ammonium and TOC are removed efficiently in the stretch between U and D. On the contrary, increased loads of TSS between U and D in this date cannot be explained solely by the effluent because the sum of the loads in U and W is less than load in D. TP load in site D was approximately the sum of U and W loads. Vilches et al. (2011) attributed the increase of particulate material and phosphorus to non-point sources related to farming activities, which predominate in the study area. According the bioassay results, the presence of floating plants at the mouth of the open channel could improve the efficiency in the reduction of ammonium and TP. The presence of floating plants also promotes the ammonium volatilization reduction that represents a major source of atmospheric pollution. Considering the results of Basílico and de Cabo (2012), TSS loads could be also decreased, beyond the source of solids.

A relevant aspect is the nutrients loading stored in plant tissues. If the plants are not harvested, nutrients can be rapidly released into the water column during decomposition of detrital tissue and this should be considered in phytoremediation experiences with this species. For instance, phosphorus assimilation in vegetation was found to be short-term because of the rapid turnover (Patel and Kanungo, 2010). Phosphorus release from decomposing tissues is rapid, with about 80% of P released in 15 days for *Pistia* sp. and *Hydrocotyle* sp. (Reddy et al., 1995).

**Table 4**  
Loads on sites W, U and D for the date of October 2010.

Site	Loads			
	TSS (kg/day)	Ammonium (kg/day)	TP (kg/day)	TOC (kg/day)
W	351	34	12	303
U	1671	380	91	638
D	3617	371	107	824

## 4. Conclusions

Concentrations of TSS, BOD<sub>5</sub> and nutrients increased in the La Choza stream segment under study by the discharge of effluent W. *S. intermedia* was tolerant to this wastewater mainly composed of poultry slaughterhouse effluent. The results suggest that the species could be used for the removal of ammonium, and secondarily TP from wastewater, although there are other potential non-point sources of the latter in the system. Bioassay showed that although there were no significant differences in DIN removal rates between treatments, plant uptake and nitrification seem to favor ammonium decrease in the presence of the species. Total phosphorus removal was significantly higher in treatments with plants, revealing the importance of *S. intermedia* in the process. However, SRP increased in both treatments. In the treatment without plants, P was released from sediments during resuspension events, favored by high pH. For the *in situ* treatment of the effluent with *S. intermedia* it should be considered a regular harvesting of plant biomass in order to avoid P release. Macrophyte presence may not enhance dissolved phosphorus retention by direct assimilation. So, additional research is necessary to study the role of the algal community in this system.

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