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Aquatic plant composition and environmental relationships in grazed Northwest Patagonian wetlands, Argentina

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ABSTRACT

Aquatic macrophyte assemblages and environmental features were assessed in 30 wetlands from Northwest Patagonia subjected to different intensity of grazing pressure. Species richness was surveyed in wetlands from three biozones of the Patagonia Ecoregion: Andean-Humid, Sub-Andean Sub-Humid and Extra-Andean Occidental. A total of 50 species of macrophytes were recorded, with the Cyperaceae, Juncaceae, Poaceae and Ranunculaceae the best represented groups. Canonical Correspondence Analysis (CCA) reflected the distribution of species along a gradient of disturbance (CCA1: salinity and ammonia). There was also a correspondence among community composition and intensity of grazing pressure at isolated wetlands, with total species richness, richness of natives and aquatic plant coverage significantly decreasing towards most disturbed sites. Only the species *Distichlis spicata*, *Xanthium spicatum* and *Eleocharis melomphala* appeared as indicators of wetland deterioration associated with degraded sites, subjected to strong erosion processes which increase the natural soil salinity. The submersed *Lilaeopsis macloviana* was the most tolerant species being recorded over a wide range of environments and intensity of land uses. Variables related to wetland size and connectivity (CCA2: depth, length, elevation and dissolved oxygen) displayed higher explanatory power on community assemblages, and subsequently macrophyte life-forms were clearly distinguished across this gradient. Most of species recorded in this work were native, however the proportion of exotics reached 25% in areas with intermediate and high grazing intensity. Although we were not able to clearly separate natural from grazing effects this study provides a first look at natural and anthropogenic controls of macrophytes in Patagonian wetlands.

1. Introduction

It has been suggested that wetlands are very resilient ecosystems because they exhibit characteristics of both aquatic and terrestrial systems that provides them a high level of robustness across a range of physical and chemical conditions (Ripken, 2009). However, recent studies on grasslands demonstrate that species losses and subsequent changes in diversity can decrease the resilience (recovery ability) of ecosystems after a disturbance and can alter ecosystem functioning. The responses of ecosystems to environmental perturbation or species loss ultimately depend on individual species traits and species interactions within a community (Engelhardt and Kadlec, 2001). According to Karr and Chu (1999) the presence of a wetland’s natural biological community means that the wetland is resilient to the normal variation in that environment. In this context, certain human activities can alter the interactions between wetland biota and their chemical and physical environment. When human activities within a wetland or its watershed are minimal, the biological communities are resilient and continue to resemble those that were shaped by the interaction of biogeographic and evolutionary processes (US EPA, 2002). On the contrary, a high level of disturbance such as that resulting from eutrophication, acidification, or alkalinization processes, or those driven by human land use practices can cause a shift, a decrease, or the complete loss of resident species of aquatic macrophytes in a wetland (Sass et al., 2010).

Aquatic macrophytes are well-suited as indicators of ecological integrity in wetlands (Cronk and Fennessy, 2001; DeKeyser et al., 2003). The measurement of aquatic plant attributes (e.g. percentage of exotic, sensitive, and tolerant species, total number of species, maximum depth of plant growth, species richness

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and composition, species tolerance, guild structure, vegetative abundance, conservation status, etc.) can reflect the effects of successive physical, chemical, and biological changes in the surrounding aquatic environment (Nichols et al., 2000; Miller et al., 2006). Moreover, the taxonomy of aquatic plants is well known and the sampling can be accomplished with minimal costs (Fore et al., 2007). At present there are different indices that combine some of the mentioned attributes of a floristic community that are employed to assess the biotic integrity of wetlands. Many countries have developed appropriate measurements for aquatic environments taking in to account the regional composition of aquatic plants as, for example those developed in North America (Fennessy et al., 1998; Nichols et al., 2000; Alix, 2006; Rothrock et al., 2008; Seilheimer et al., 2009), Europe (Maggioni et al., 2009; Pätzig et al., 2012), Australia and New Zealand (Smith et al., 2009; Lingl and Jacobs, 2011). These measures have also been successfully employed in rapid biological protocols and have been shown to be effective tools to assess the ecological condition and the effects of different levels of disturbance on wetlands (López and Fennessy, 2002). Nevertheless an obligate step in the implementation of these measures involves an adequate knowledge of floristic composition, species distribution, and of the specific responses of aquatic plants to environmental variables and disturbances (Mack, 2009; Sass et al., 2010).

Wetlands of the continental sector of southern South America are very diverse and include meadows, marshes, bogs, forests and shrubs (Clausen et al., 2006; Mazzoni and Vázquez, 2004). In Patagonia, due to the influence of topography on water redistribution, most wetlands occur in the drainage lines between the cordillera (mountain range) and the Plateau (Marcolin et al., 1978). The primary ecosystem functions of Patagonian wetlands includes: the cycling of nutrients and organic matter, and the provision of water, food and habitat (oviposition, nurseries, refuge, etc.) for a highly endemic fauna (Perotti et al., 2005; Ciari, 2009). Additionally, wetlands play a key role in providing forage and water supply for cattle that result in high herbivore preferences for this type of environment (Gaitán et al., 2011). This often leads to processes of soil and plant community degradation. As occurs in other regions of the world, accelerated desertification (a degradation process in arid, semiarid and subhumid areas resulting from anthropogenic activities) is one of the main environmental problems affecting the region (Del Valle et al., 1998). Other consequences of overgrazing in wetlands are increases in organic pollution, nutrient loads and sedimentation by stocking and trampling (Bronmark and Hansson, 2002). The increase of soil salinity is another symptom of deterioration after overgrazing, because the loss of plant cover induces the rate of evaporation on exposed soils increasing salt concentration (Del Valle, 1993).

Because animals consume mostly terrestrial plants the pathways in which livestock activity can affect aquatic plant communities are mostly indirect. Trampling is one of the most common effects of livestock stocking that often releases fine sediments from soils. These sediments are usually mobilized into the water by rain and wind, and also during the expansion and retraction of water bodies within wetlands, that changes bottom conditions and for instance altering some mechanisms of seeds propagation of aquatic plants (Gleason et al., 2003a,b). Increasing turbidity in the water column after these episodes in turn, diminishes light penetration, affecting some macrophyte life forms (Declerck et al., 2006). Grazing can also produce eutrophication symptoms in wetlands as a consequence of the nutrients from animal deposition (urine and faeces), that elevate nutrients and modify macrophytes assemblages and coverage (Markwell and Fellows, 2008; Xiao et al., 2010).

Compared with the extensive literature on macrophytes as indicators of wetland ecological status in several regions of the world (Fennessy et al., 1998; Seilheimer et al., 2009; Melzer, 1999; Smith et al., 2009), considerably less work has been done in South America, and in particular in southern Argentina. Whilst several studies of floristic composition and aquatic vegetation life forms were conducted in Chile (Ramírez and Stegmaier, 1982; Ramírez et al., 1991; Hauenstein, 2006; Clausen et al., 2006; Saldivia Pérez, 2006; Hauenstein et al., 2008), the knowledge of wetlands on the Argentinean side is still incipient with most works providing general descriptions of communities associated to ponds and wet meadows (Collantes and Faggi, 1999; Raffaele, 1999; Perotti et al., 2005; San Martín et al., 2009). Recently, Gaitán et al. (2011) performed a phytosociological study of terrestrial plant communities associated with wetland areas in northern Patagonia and analyzed the influence of natural environmental variables. However, the potential use of aquatic plants as indicators of wetland condition, and the assessment of species responses to anthropogenic disturbances, has not been yet investigated. This paper is also important because it documents Patagonian wetlands that have been relatively understudied.

In the present study we hypothesize that different level of grazing pressure on wetlands will lead to changes in environmental features and in aquatic plant communities (species richness and composition) reflecting the land degradation process. We also will attempt to determine which macrophytes species can be considered indicators of wetland conditions, taking into account the level of grazing pressure and of the environmental stress in which they are present. In order to achieve these objectives we studied the composition of macrophyte communities at 30 Patagonian wetlands subjected to different intensities of cattle grazing and assessed the main environmental constraints operating in their distribution. Given the economical and ecological importance of these key environments, the results of the present paper will be relevant for the management, conservation and restoration actions of Patagonian wetlands.

2. Methodology

2.1. Landscape setting and site selection

The study area belongs to the Andean-Humid, Sub-Andean Sub-Humid and the Extra-Andean Occidental biozones (Del Valle et al., 1998). It is located in the transition between the Precordillera and the Plateau in the Northwest of Chubut Province, Argentina (Fig. 1). Wetlands are associated with the four main river basins having differences in climate, geomorphology and phytogeography.

Due to the Pacific Ocean influence, the Andean-Humid and Sub-Andean biozone, exhibits a strong rainfall gradient decreasing from the west to east (3000–600 mm y^{-1}). Evergreen (*Austrocedrus chilensis*, *Nothofagus dombeyi* and *Maytenus boaria*) and deciduous species (*Nothofagus pumilio* and *Nothofagus antarctica*) constitutes the dominant trees in the area. The shrub and herbaceous strata are characterized mainly by *Chusquea culeou*, *Berberis microphylla*, *Lomatia hirsuta*, *Schinus patagonicus*, *Diostea juncea*, *Fuchsia magellanica*, *Alstroemeria aurea*, *Mutisia spinosa* and *Mutisia decurrens*. Futaleufú and Carrenleufú basins are located in these biozones (Del Valle et al., 1998).

The Extra-Andean biozone is characterized by a west-east rainfall gradient (500–100 mm y^{-1}). Mean annual air temperature is 8.5 °C. The landforms correspond to the Patagonian steppe, where the low precipitation has resulted in xerophytic and cushion plants. The shrub and herbaceous strata are well represented, being *Mulinum spinosum*, *Pseudostipa speciosa*, *Pseudostipa humilis*,

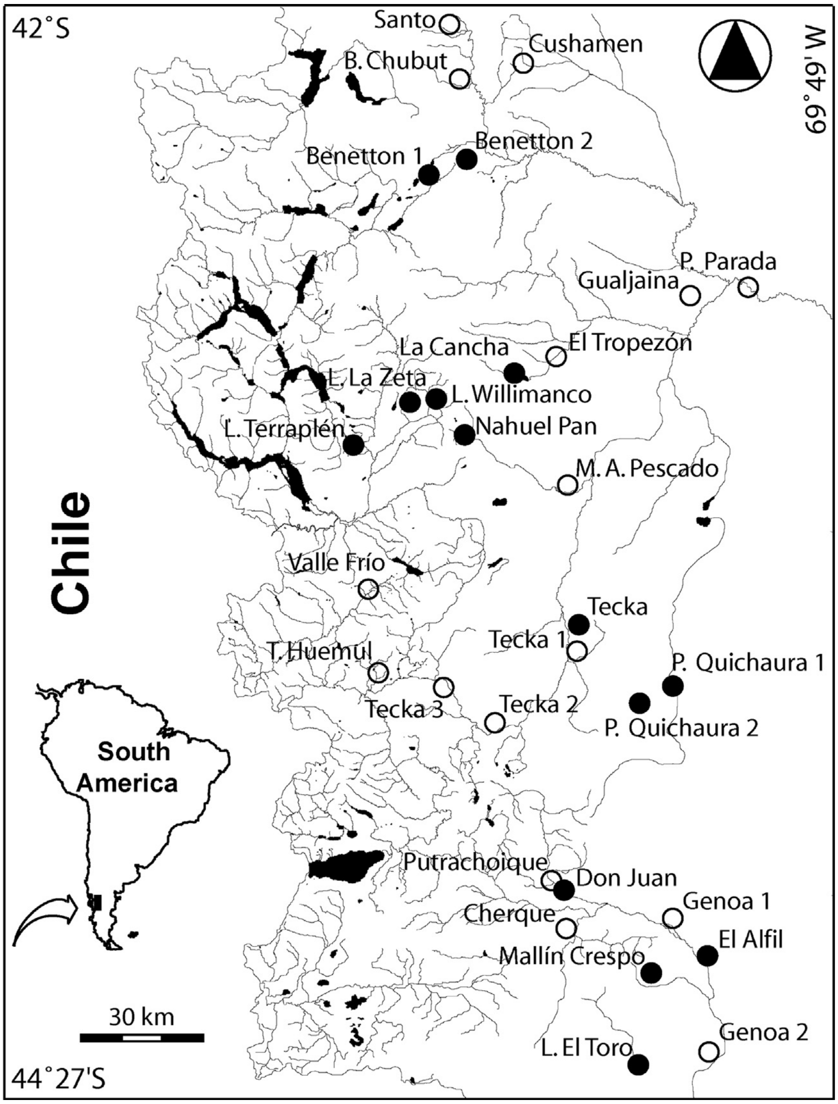


Fig. 1. Location of the 30 studied wetland, Chubut Province, Patagonia, Argentina. Dark circles: isolated wetlands, open circles: connected wetlands.

Poa ligularis, *Nassauvia glomerulosa*, *Chuquiraga avellanedae* and *Azorella monantha* the most common species. Tecka-Gualjaina, Languiñeo-Pampa de Agnia-Paso de Indios, Apeleg-Genoa and Chubut sub basins belong to this biozone. Bajo de las salinas is a small endorheic basin (396 km^2), characterized by salt accumulation and a marked physical and biological degradation as consequence of inadequate land uses practices with vegetation dominated by halophytic plants (Coronato and Del Valle, 1988; Del Valle, 1993).

In the last decades anthropogenic influences induced by the intensification of cattle raising has resulted in deterioration of wetland conditions in the Patagonian landscape. In particular in the Chubut Province, wetlands are important as water suppliers but also sustain large populations of livestock in semiarid areas (Gaitán et al., 2011). Another important factor is that the increase in the desertification process had resulted in low productive lands, forcing the landowners to change their extensive livestock practices for intensive use on more productive areas such as wetlands that have been converted to pastures. This greater pressure on these environments has reduced vegetation cover, diminished water retention, and exposed soils to erosion (Del Valle et al., 1998). In addition, these phenomena have led the increase of salinization and the loss of habitat and biodiversity (Steinman et al., 2003). Some specifics wetlands areas (e.g. Laguna El Toro, El Alfil) are being intervened by the INTA in a land recovery programme in which tall wheatgrass *Thinopyrum ponticum* (Poaceae) is being planted. The objectives are to stabilized soils and supply an alternative form of forage for livestock.

2.2. Field methods

The objective of our site selection was to have a broad range of environmental conditions that included representative wetlands from three different biozones in the Chubut Province. As a first step a LandSat 7 image (2003) in an area between 42° 02′ and 44° 22′ S and 70° 13′ and 71° 31′ was used. The study was also intended to incorporate different intensities of anthropogenic activities primarily defined by the pressure of grazing (cattle and sheep). For that purpose information about land use, cattle raising type and stocking densities was obtained at different governmental administration offices (DPByP-Dirección Provincial de Bosques y Parques, INTA-Instituto Nacional de Tecnología Agropecuaria). Although a set of 40 potential sites were initially established, a group of 30 sites were chosen based on accessibility and similarity in the prevailing land use.

Table 1
Physicochemical and geomorphologic features measured at 30 northwest Patagonian wetlands (Argentina).

Sites names and codes	Connectivity and permanency	Elevation m.a.s.l.	Water temperature (°C)	Depth (cm)	Width (m)	Large (m)	pH	Conductivityty ($\mu S\,cm^{-1}$)	TDS ($mg\,l^{-1}$)	Diss. oxygen ($mg\,l^{-1}$)	Salinity 0/00	TN ($\mu g\,l^{-1}$)	NO_3 ($\mu g\,l^{-1}$)	NH_4 ($\mu g\,l^{-1}$)	TP ($\mu g\,l^{-1}$)	SRP ($\mu g\,l^{-1}$)	Land-use class and intensity	Aq. plant cov.
Santo (SAN)*	C/In	718	26.5	14.2	5	5	7.01	118.8	54.7	10.7	0.0	251	2.5	19	31	10	P M	2
Cushamen (CUS)*	C/In	848	21.6	41	3	5	8.98	239	123.4	18.27	0.1	244	2.5	11	17	1	P M	5
B. Chubut (BCH)*	C/In	671	10.9	38	4	10	6.98	30.2	19.4	12.86	0.0	153	2.5	4	29	5	P L	4
Benetton 2 (MB2)*	I/In	727	14.3	20	50	150	8.08	170.8	103.7	10.5	0.1	419	140	4	36	3	P M	4
Benetton 1 (MB1)*	I/Pe	636	14.3	60	50	300	6.79	111.5	na	5.6	0.1	629	2.5	14	45	1	P L	5
P. parada (PiP)	C/In	448	20.6	13.2	10	50	8.20	4002	133	12.8	2.4	4761	2.5	315	445	22	P H	3
Gualjaina (GUJ)	C/In	473	22.2	26.5	5	10	7.99	185	168.3	9.96	0.2	1616	2.5	128	982	810	P H	4
La Cancha (LaC)	I/In	808	15.5	30	150	480	9.45	6610	4430	9.15	4.6	5181	14	260	3922	3062	P H	2
El tropezón (ElT)	C/In	781	17.9	3.8	2	2	8.27	1124	648	10.57	0.6	504	2.5	4	23	3	P H	4
L. La Zeta (LaZ)*	I/In	586	15	488	560	1420	7.67	229	138.8	8.97	0.1	662	2.5	9	28	1	P/R L	4
L. Willimanco (LaW)*	I/In	704	11.5	640	854	870	7.67	433	286	9.5	0.3	176	2.5	4	71	41	P M	4
Nahuel Pan (NhP)*	I/In	752	15	150	200	1585	8.1	534	321	9.65	0.3	809	2.5	29	26	1	P/R M	4
L. Terraplén (LaT)**	I/Pe	603	12.9	239	1850	2300	6.86	61.4	37.9	8.83	0.1	768	2.5	108	61	1	P M	4
M. A. Pescado (MAP)	C/Pe	594	12.5	60	5	5	8.49	257	161.8	12.5	0.2	537	316	4	71	41	P M	5
Valle frío (VFr)**	C/Pe	696	17.4	13.7	10	10	6.79	33.3	18.1	9.27	0.0	302	2.5	4	56	1	P M	6
Tecka 1 (TK1)	C/In	658	15	47.2	3	10	7.23	64.7	39	12.18	0.0	261	2.5	5	41	8	P M	5
Tecka 3 (TK3)*	C/Pe	800	14.1	28	20	30	7.16	40.6	24.2	12.32	0.1	208	2.5	4	37	3	P M	4
Tecka 2 (TK2)*	C/Pe	722	12.6	34.6	10	20	7.04	28	17.8	13.3	0.0	163	2.5	4	22	2	P L	3
Tecka (MTe)	I/In	656	17.8	20	80	400	7.7	153.6	86.1	10.21	0.1	1063	4	35	129	16	P M	6
T. Huemul (TrH)**	C/In	652	18.7	35	3	10	6.93	72.3	39.2	11.54	0.0	514	31	4	32	2	P L	6
P Quichaura (PQ1)	I/In	730	10.5	25	150	300	8.37	242	163.9	10.45	0.2	691	2.5	21	55	3	P M	3
P Quichaura (PQ2)	I/Pe	918	11.3	31.7	8	20	8.65	575	377	9.6	0.4	878	4	na	49	3	P M	3
Putrachoique (PCh)*	C/Pe	791	15.7	26.8	3	5	7.18	242	142.1	9.40	0.1	408	2.5	8	59	9	P M	7
Don Juan (DoJ)*	I/In	772	19	25	15	30	9.25	332	185.2	12.8	0.2	10,514	6	2269	1951	593	P H	3
Genoa 1 (G1)	C/Pe	683	18	17	2	10	8.90	124	67.6	12.48	0.0	555	2.5	55	84	16	P M	4
Cherque (CHE)*	C/In	772	18	43	2.5	50	9.11	135.2	74.8	7.72	0.0	347	2.5	4	29	2	P M	5
El Alfil (ElA)	I/In	675	24.7	25	15	30	8.76	608	307	9.2	0.3	613	2.5	57	84	3	P H	4
Mallín Crespo (MCr)	I/In	808	21	34.1	5	8	8.01	394	200	7.25	0.2	427	2.5	8	100	2	P L	5
Genoa 2 (Ge2)	C/In	632	18.6	21.7	5	10	8.98	216	118.4	12.04	0.1	496	2.5	6	46	1	P M	6
L. El Toro (LET)	I/P	605	14.1	150	400	800	8.93	1846	1197	9.45	1.2	874	2.5	12	65	10	P H	3

TDS: total dissolved solids, TN: total nitrogen, NO_3: nitrates, NH_4: ammonia, TP: total phosphorus, SRP: soluble reactive phosphate, C: connected, I: isolated, na: non available, Pe: permanent, In: intermittent.
Land-use codes: P: pasture, R: rural. Intensity: L: low, M: medium, H: high.

** Andean biozone.

* Sub-andean biozone.

Without * Extra-andean biozone.

The dominant land use and the intensity of disturbance at studied wetlands are presented in Table 1. Most wetlands were dedicated pastures in which the livestock was sustained by the herbaceous stratum. Selected sites present a mix of cattle types mostly consisting in sheep, cows and horses. Sites that had cows but not sheep were: L. La Zeta, T. Huemul and Genoa 1, conversely those sites that sustained sheep but not cows were: Mallín Crespo, El tropezón, Cushamen, P. Parada and Gualjaina. All sites have horses for domestic use moreover it has cultural value for some aboriginal communities. The level of grazing pressure was assessed as low, medium and high. This determination was based on the cattle stocking density which was provided by the INTA administration and the land carrying capacity score assigned to different biozones in the Patagonia region as recommended in Siffredi et al. (2005, 2007). Additionally we assessed photographic evidence of soil erosion and compaction (by stocking) in the adjacent land, signs of trampling in shorelines and within water bodies, faeces amount, and signs of foraging on vegetation. At all locations the access for livestock to the water in the wetlands was unrestricted, thus most ponds or water bodies were used for water supply. Nevertheless as cattle and horses usually get into the water bodies those places having this type of animal were ranked with higher levels of disturbances. In the assignation of the impact ranks, we also considered the suggested rotational practices for livestock (Nakamatsu, 2006). Selected sites were located distant from urbanizations however some wetlands were near to rural settlements.

According to US EPA, 2002 it is necessary to establish a standard period of time within the year to collect samples that (1) minimizes variation caused by natural, seasonal changes in community composition; (2) provides sufficient differentiation of communities across a disturbance gradient; and (3) is logistically practical. Summer represents the typical period of maturity and the best time for determining macrophytes community composition in Patagonian wetlands. It is also the time of the year in which wetlands usually displays sufficient water availability and due to the management of the livestock, exhibits a higher grazing pressure. Normal ranching practices in the region must include a seasonal rotation of livestock, in the summer livestock are generally stocked on wetlands areas having a better forage offer, and by the end of the season (dry period) livestock is moved to other parcels. These parcels are usually located in lowlands areas, or are some specifics areas in properties where grazing is temporarily restricted in order to improve and recover the pastures (Lloyd, 2002). Subsequently, the sampling period established to conduct the study is assumed as coincidently with livestock presence in wetlands at most sites.

The selected wetlands were visited once in summer: 15 were sampled in December 2006 and the rest in December of 2007. Survey was carried out under stable environmental conditions avoiding rainstorms or extremely high discharge events. An examination of weather conditions in a series of climatic data proceeding from different meteorological station (Norwest Chubut) from the INTA (rainfall, air temperature and wind) revealed that both sampling periods were comparable (INTA, 2007).

2.3. Environmental variables

Size, permanency of inundation and hydrologic connectivity of each studied wetland were determined through a LandSat 7 images RGB 123 (2003, mapping resolution 30 m × 30 m) and field data. Wetlands were classified in connected and hydrologically isolated following Mitsch and Gosselink (2007). Connected wetlands were those located between dry terrestrial systems and permanently flooded deepwater aquatic systems (e.g. rivers, lakes), whereas isolated wetlands were those at basins with little outflow and no adjacent deepwater systems, where the only nearby aquatic system is presumed the groundwater aquifer. Although we use the term "isolated" it is understood that they are usually hydrologically connected to groundwater (Mazzoni and Vázquez, 2004). Each site was also classified based on its wetland water dynamics: permanent or intermittent (Williams, 2006). Permanent wetlands were those associated with lakes and deep ponds, whereas wetlands that are dried at least once in a period of 10 years were classified as intermittent.

To detect possible effect of cattle stocking on wetland condition, water quality and nutrient cycling, several environmental variables including physical and chemical parameters were measured once at each site, this was carried out in order to establish among sites comparison. In shallow ponds depth was measured at five locations in each site, whereas mean depth for lacustrine fringes areas were obtained from available references (Quirós, 1988). At each sampling site water temperature, specific conductance ($\mu S_{20}\ cm^{-1}$), pH, and dissolved oxygen ($mg\ O_2\ l^{-1}$) were measured with a sensION 156 multiparameter probe. For better comparisons measurements were made approximately at same time each day. Water samples were collected below the water surface and kept at 4 °C prior to analysis. At the laboratory main nutrients ($\pm 0.01\ \mu g\ l^{-1}$) were assessed as follows: Total nitrogen (TN): alkaline oxidation with potassium persulphate and boric acid, cadmium column reduction and subsequent diazotization (Grasshoff et al., 1983). Total oxidized nitrogen (NO_3 plus NO_2) was analyzed following cadmium column reduction and subsequent diazotization. Ammonia (NH_4) was determined colourmetrically by the indophenol blue method. Total phosphorus (TP) was obtained by acid oxidation with Potassium persulphate and subsequent determination as soluble reactive phosphate (SRP), by the molybdate/ascorbic acid method (APHA, 1994).

2.4. Plant collection

Qualitative macrophytes samples were gathered from all the studied wetlands. At each site a stratified random sampling was performed in order to include all life forms present in a site: emergent (plants rooted, morphologically adapted to growing in a water-logged or submersed substrate), floating-leaves (plants rooted in the substrate with floating leaves), submersed (plants with photosynthetic tissue entirely submersed) and free-floating (surface-floating plants with few or no roots) (Cronk and Fennessy, 2001). In permanent environments (lakes) the lacustrine fringe areas was examined. Voucher specimens for all species were collected and put in collection bags then pressed for later laboratory identification. The species were observed in a LEICA MZ6 stereomicroscope and identified using regional bibliography (Correa, 1978–1999). Species were also classified as native, endemic and exotic following the Catalogue of the Vascular Plants from the Southern Cone (Zuloaga et al., 2008). Additionally, vegetation cover was assessed visually (Galatowitsch et al., 2000). The percent cover of aquatic plants was estimated and divided in seven categories (<1%, 1–5%, 6–25%, 26–50%, 51–75%, 75–99%, and 100%).

2.5. Data analysis

A Principal Component Analysis (PCA) on $\log(x+1)$ transformed data was performed to examine variation in physical and chemical parameters across the studied sites. This method used within its intended limits is a valuable procedure to detect structure in the relationships between variables (Ludwing and Reynolds, 1988). The non-parametric tests Mann–Whitney (pair comparisons) and Kruskal–Wallis (multiple comparisons) were used to examine differences in species richness, among different types and conditions of wetlands: permanency, connectivity and location (biozone). Same analysis was employed to test trends of species richness, life forms and plant origin with intensity of land use at isolated and connected wetlands separately (Sokal and Rohlf, 1995). Only those measures with certain variability and enough number of cases at disturbance types were tested.

Canonical Correspondence Analysis (CCA) was run using CANOCO (Ter Braak and Smilauer, 1999) to assess relationships between aquatic plant assemblages and environmental variables. CCA is a powerful tool for simplifying complex data sets and, being a direct gradient analysis, it allows integrated analysis of both taxa and environmental data (Ter Braak and Smilauer, 1998). The technique identifies an environmental basis for community ordination by detecting the patterns of variation in community composition that can be explained best by the environmental variables. Environmental variables included in Table 1 (expect land use and intensity) were used initially to evaluate the response of species and sites to environmental gradients. Variables (except pH) were $\log(x+1)$ transformed prior to analysis. Variables that were strongly inter-correlated with others (those with an inflation factor >20) in the initial analysis, were removed (conductivity, TDS) and a further analysis was carried out with the remaining environmental variables. The forward selection option provided by CANOCO was applied and those variables with $p < 0.1$ (Monte Carlo permutation test) were kept for the analysis (wetland width, TP, TN, and NO_3 were omitted). The final CCA was run using a set of independent and significant environmental variables (Ter Braak and Smilauer, 1998).

3. Results

3.1. Environmental characterization

Wetland assessed varied from pools to small lakes and this was reflected in their environmental features (Table 1). Water temperature ranged between 10.5 (Putrachoique 1) and 26.5 °C (Santo) sites whereas dissolved oxygen contents ranged between 5.6 mg l^{-1} (Benetton 1) and 18.27 mg l^{-1} (Cushamen). According to pH values sites varied between neutrals (Mallín Benetton 1, Valle frío, Laguna Terraplén) to extremely alkaline waters (La Cancha, Don Juan; Cherque). Even though most wetland showed intermediate water conductivity values (~100 μs cm^{-1}), sites in the Extra Andean biozone showed significantly higher conductivity values than Andean Humids ones, displaying those isolated wetlands significantly higher salinity than connected ones (Kruskal Wallis, $p < 0.001$).

Regarding nutrient contents maximum values of total nitrogen and ammonia were recorded at Don Juan, La Cancha and Piedra Parada. However, most studied sites (23 of 30) showed very low nitrate values (~2.5 μg l^{-1}) and with total phosphorous values lower than 130 μg l^{-1}. High values were recorded at La Cancha (3922 μg l^{-1}), Don Juan (1951 μg l^{-1}) and Gualjaina (982 μg l^{-1}).

As shown in the PCA ordination graph physical and chemical data provided a clear distinction among the studied wetlands (Fig. 2). Sites showing higher nutrient concentrations, conductivity and alkalinity were placed on the upper and lower right quadrants, those having diluted waters were positioned to the negative end of PC1. This axis highlighted also an eutrophication gradient that increases from left to right on PC1 where sites were distributed accordingly. The second axis highlighted a gradient in oxygen content, water temperature and depth of the water body. Moreover there is a clear segregation between connected (open circles) and isolated wetlands (black circles). The first three principle components axes accounted for most of the variation in the data set (62% of total variation). PC1 (35.3% of total) consisted in water chemical variables and physical attributes of the sites (i.e. high positive loadings by conductivity, TN, TP, NH4, PRS and pH) and PC2 (14.2% of total) consisted mostly of physical variables (i.e. high positive loadings large of the water body and high negative loadings by oxygen contents and water temperature). PC3 captured 12.5% of data variation and included most of variables comprised in PC1 and PC2.

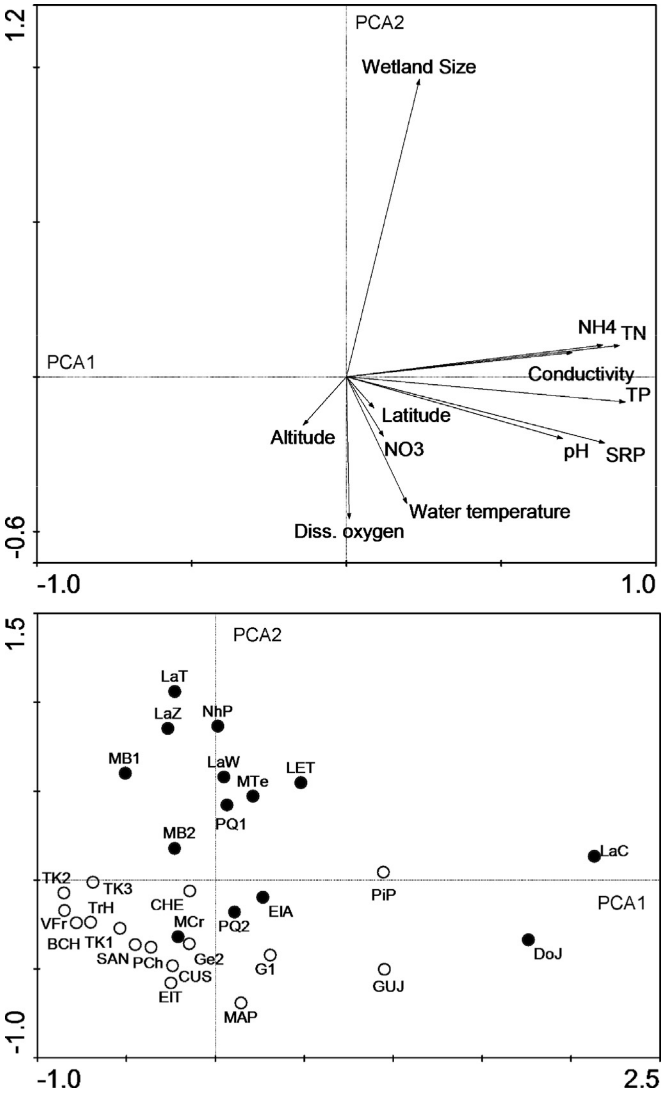


Fig. 2. Ordination of sites according to Principal Component Analysis (PCA) on the environmental variables measured at 30 wetlands, Chubut Province, Patagonia Argentina. (December 2006 and December 2007). Open circles: connected, black circles: isolated wetlands. Names of the sites are in Table 1.

3.2. Aquatic plant assemblages

We recorded a total of 52 species of aquatic plants, 47 determined to species level, four to genus level and the remaining to division level (Table 2). The taxonomic groups best represented were Dicotyledons (Magnoliopsida, 45.2%), Monocotyledons (Liliopsida, 47.2%) and algae (5.7%). Twenty-two families were represented and the most relevant in terms of species richness were Cyperaceae (8), Poaceae (6) Juncaceae (5), Ranunculaceae and Potamogetonaceae (4). Among the algae three genera: *Chara*, *Nitella* and *Cladophora* were identified. Mosses (Bryophyta) were

Table 2
Floristic composition of the wetlands. The origin (Nat.: native, End.: endemic, Exot.: exotic, *: cosmopolitan) and habit (Em.: emergent, Fl-l.: floating-leaved, Fr-f.: free-floating, Sub.: submersed) are detailed.

Taxa	Codes	Origin	Habit
Bryophyta spp.	Brio		Em.
Characeae			
Chara sp.	Char		Sub.
Nitella sp.	Nitel		Sub.
Cladophoraceae			
Cladophora sp.	Clad		Sub.
Apiaceae			
Hydrocotyle chamaemorus Cham. & Schltdl.	H.cha	Nat.	Sub.
Lilaeopsis macloviana (Gand.) A.W. Hill	L.mac	Nat.	Sub.
Asteraceae			
Matricaria recutita L.	M.rec	Exot.	Em.
Taraxacum officinale Weber ex F.H. Wigg.	T.off	Exot.	Em.
Xanthium spinosum L.	X.spi	Nat.	Em.
Brassicaceae			
Cardamine variabilis Phil.	C.var	End.	Em.
Nasturtium officinale W.T.Aiton	N.off	Exot.	Em.
Callitrichaceae			
Callitriche lechleri (Hegelm.) Fassett	C.lec	End.	Sub.
Callitriche truncata Guss.	C.tru	Exot.	Sub.
Cyperaceae			
Carex gayana E. Desv.	C.gay	Nat.	Em.
Carex macrorrhiza Boeck.	C.mac	Nat.	Em.
Carex nebularum Phil.	C.neb	Nat.	Em.
Eleocharis pseudoalbibracteata S. González & Guagl.	E.pab	Nat.	Em.
Eleocharis macrostachya Britton	E.mac	Nat.	Em.
Eleocharis melanomphala C.B. Clarke	E.mel	Nat.	Em.
Eleocharis melanostachys (d'Urv.) C.B. Clarke	E.mes	End.	Em.
Schoenoplectus californicus (C.A. Meyer) Soják	S.cal	Nat.	Em.
Fabaceae			
Trifolium repens L.	T.rep	Exot.	Em.
Haloragaceae			
Myriophyllum quitense Kunth	M.qui	Nat.	Sub.
Hippuridaceae			
Hippuris vulgaris L.	H.vul	Nat.	Em.
Juncaceae			
Juncus balticus Willd.	J.bal	Nat.	Em.
Juncus burkartii Barros	J.bur	End.	Em.
Juncus involucratus Steud. Ex Buchenau	J.inv	Nat.	Em.
Juncus sp.	J.sp.		Em.
Juncus scheuchzerioides Gaudich.	J.sch	Nat.	Em.
Juncaginaceae			
Triglochin palustris L.*	T.pal	Nat.	Em.
Lamiaceae			
Mentha aquatica L.	M.aqu	Exot.	Em.
Lemnaceae			
Lemna gibba L.*	L.gib	Nat.	Fr-f.
Plantaginaceae			
Plantago major L.	P.maj	Exot.	Em.
Veronica anagallis-aquatica L.	V.a-aq	Exot.	Em.
Veronica serpyllifolia L.	V.ser	Exot.	Em.
Poaceae			
Alopecurus magellanicus Lam.	A.mag	Nat.	Em.
Alopecurus pratensis L.	A.pra	Exot.	Em.
Distichlis spicata (L.) Greene	D.spi	Nat.	Em.
Glyceria multiflora Steud.	G.mul	Nat.	Em.
Poa lanuginosa Poir.	P.lan	Nat.	Em.
Poa pratensis L.	P.pra	Exot.	Em.
Polygonaceae			
Rumex crispus L.	R.cri	Exot.	Em.
Potamogetonaceae			
Potamogeton linguatus Hagstr.	P.lin	End.	Fl-l.
Stuckenia striata (Ruiz & Pav.) Holub	S.str	Nat.	Sub.
Stuckenia filiformis (Pers.) Boehm. ssp. *alpina* (Blytt) R.R. Haynes, Les & M. Král	S.fil	Nat.	Sub.
Primulaceae			
Samolus spathulatus (Cav.) Duby	S.spa	End.	Em.
Ranunculaceae			
Caltha sagittata Cav.	C.sag	Nat.	Em.
Ranunculus hydrophilus Gaudich.	R.hyd	End.	Sub.
Ranunculus trichophyllus Chaix	R.tri	Nat.	Sub.
Ranunculus uniflorus Phil. ex Reiche	R.uni	Nat.	Sub.
Rosaceae			
Acaena magellanica (Lam.) Vahl	A.mge	Nat.	Em.
Scrophulariaceae			
Mimulus glabratus Kunth	M.gla	Nat.	Em.

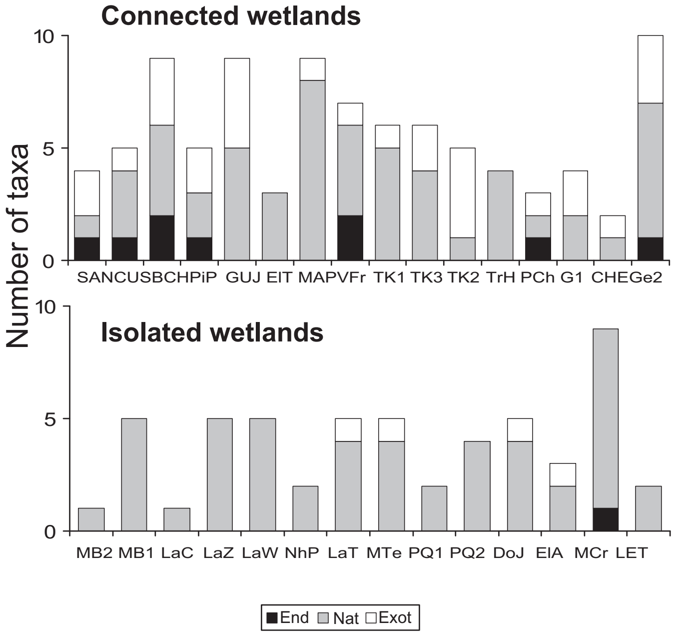


Fig. 3. Aquatic plant richness according to their origin: End: endemic, Nat: native and Exot: exotic, in 30 connected and isolated wetlands from Chubut Province, Patagonia, Argentina (December 2006 and December 2007).

less represented and occurred only at Crespo, Nahuel Pan and Genoa wetlands. Although most of the plants recorded corresponded to aquatic forms, five terrestrial species that are likely to be found in wet areas were also recorded *Matricaria recutita*, *Taraxacum officinale*, *Xanthium spinosum*, *Trifolium repens* and *Veronica serpyllifolia*.

According to our results 36 species were native (75%), and from these seven species were endemic of Chilean and Argentinean Patagonia. However, 25% of the total species inventory was exotic; these were more frequently recorded at hydrologically connected wetlands (Fig. 3). Among hydrophytes 36 species were emergent, 14 submersed, one floating-leaved, and just one was free-floating (Table 2).

From all species recorded 37.7% were found in one site. The most frequent species (>8 sites) were *Eleocharis pseudoalbibracteata*, *Myriophyllum quitense*, *Veronica anagallis-aquatica* and *L. macloviana.*

The lowest total richness corresponded to isolated sites: La Cancha and Benetton 2 (one taxon), being dominated by *Juncus* sp. and *E. pseudoalbibracteata* respectively, both sites were disconnected, intermittent and also displayed medium to high land use intensity. In comparison, the highest species richness was found at connected sites such as Genoa 1 (12 taxa), Arroyo Pescado (10 taxa), Bracito Chubut and Gualjaina (9 taxa). The species richness of emergent plants at low disturbance sites was almost twice that of those displaying high disturbance, whereas the richness of submersed life forms was practically the same among different disturbance categories (Fig. 4). Examination of species richness patterns by wetland location, type and condition did not show any significant difference (Mann Withney and Kruskal Wallis test, $p > 0.05$) (Fig. 5). Nevertheless, at isolated wetlands we observed a significant and decreasing trend in the range and mean values of species richness, native richness and aquatic plant coverage as land use intensity increased. For emergent plants, a trend was apparent but not significant ($p = 0.10$) at these sites. When the same analyses were performed at connected wetlands none of these patterns was seen (Fig. 4).

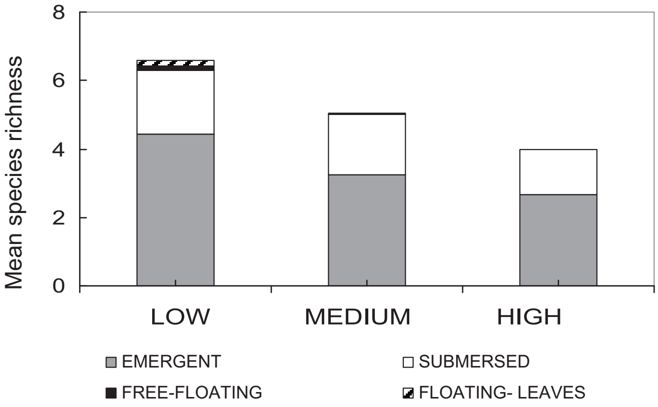


Fig. 4. Mean species richness of emergent, submersed, free-floating and floating-leaved aquatic plants in the studied wetlands spanning 3 intervals or intensities of pasture pressure.

3.3. Community ordination

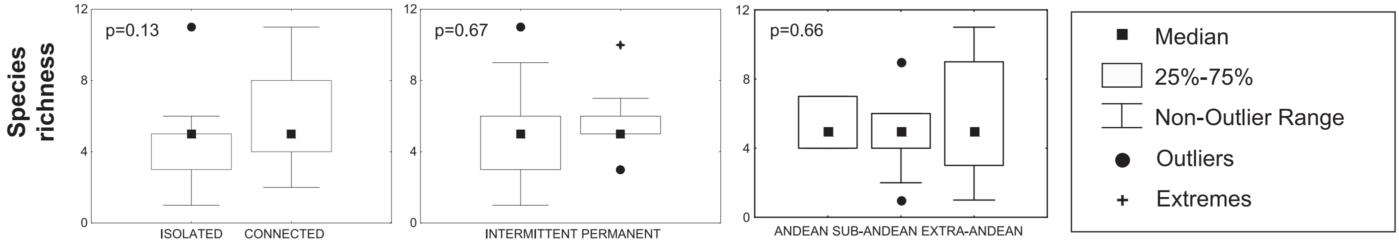
Results of the CCA are summarized in Table 3 and shown in Fig. 6. The environmental variables selected in the analysis are represented in the biplot by arrows, which point in the direction of maximum change in the value of the associated variable (Fig. 6). The species-environmental correlations were: 0.95, 0.95 and 0.91 for the first, second, and third axes, respectively (Table 3), indicating strong relationships with the environmental variables selected. Monte Carlo tests were significant for all axes (Table 3).

The strongest explanatory factors were physical and chemical variables, but only 20.3% of variation in the species data was accounted by the environmental variables measured (Table 3). CCA axis 1 reflects the distribution of species along the gradient of salinity and ammonia values, highlighting a natural but also an anthropogenic effect. Variables most strongly related to axis 2 were depth, wetland size, elevation and dissolved oxygen (Fig. 4). *Rumex crispus*, *M. recutita*, *X. spinosum*, *Distichlis spicata* and *Eleocharis melanphomala*, typically halophytes or xerophytes species associated to disturbed environments were positioned on the upper right quadrant. On the other hand, the species *Cardamine variabilis*, *Mentha aquatica*, *V. anagallis-aquatica*, Juncaceae and Cyperaceae taxa are emergent species and they were placed to the negative end of the CCA2 axis, were related to connected or flowing

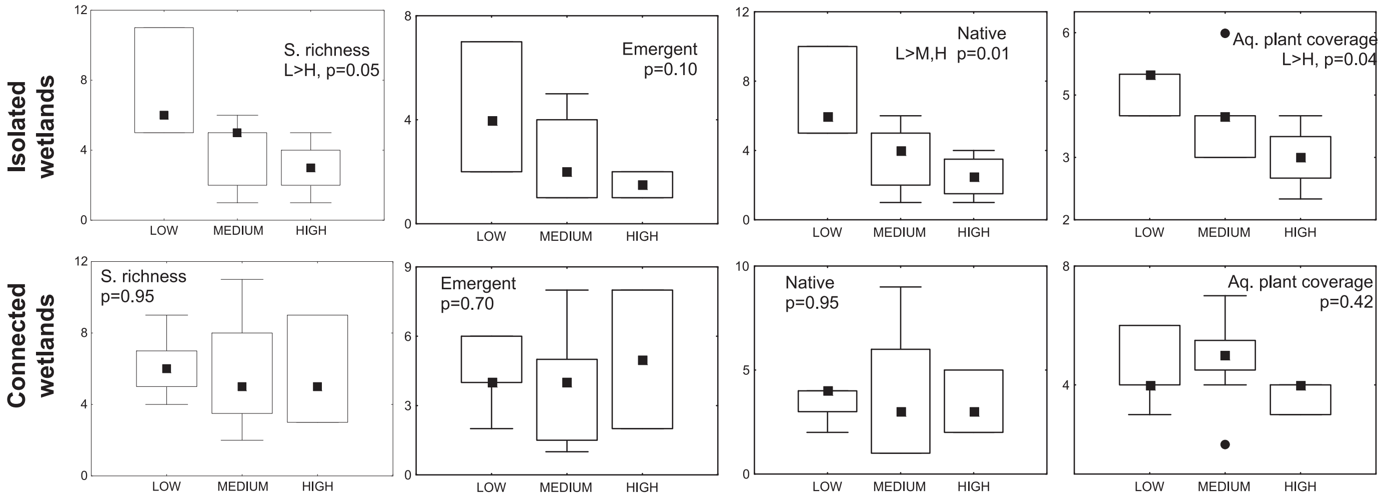
Table 3
Eigenvalues and intraset correlation of environmental variables with the axes of CCA of aquatic plants species data. Study carried out in 30 wetlands of Northwest Chubut, Patagonia Argentina (December 2006–2007), environmental variable codes in Table 1.

	CCA1	CCA 2	CCA3
Eigenvalue	0.67	0.52	0.46
Species-environmental correlations	0.95	0.95	0.91
Cumulative percentage variance			
Of species data	8.2	14.6	20.3
Of species-environmental relation	21.2	37.5	52.1
Correlations			
Altitude	−0.47	−0.44	−0.19
Water temperature	0.27	−0.39	0.12
Depth	−0.39	0.83	−0.04
Wetland size	−0.09	−0.32	0.41
Salinity	0.62	0.04	−0.39
Dissolved oxygen	0.38	−0.43	0.13
NH_4	0.41	0.04	0.44
SRP	0.11	0.07	−0.03
pH	0.08	−0.27	−0.60

Test of significance of first canonical axis: $F = 1.79$, $p < 0.02$.
Test of significance of all canonical axes: $F = 3.14$, $p < 0.0009$.



a) Wetland types and location



b) Land Use Intensity

Fig. 5. (a) Species richness patterns of aquatic plants per wetland according to connectivity, permanency, and biozones ($n = 30$). (b) Responses of total species richness, richness of emergent and natives, and aquatic plant coverage to land use intensity at isolated ($n = 14$) and connected wetlands ($n = 16$) from Chubut Province, Patagonia, Argentina. Significant relationships after Kruskal Wallis or Mann Whitney U test are shown. Symbols in box and whiskers graph are: black squares: medians, boxes: percentiles (25–75%), bars: non outlier range, black circles: outliers, and crosses: extreme values.

environments. Submersed and floating-leaved species as *Callitriche truncata*, *Nitella* sp., *Potamogeton* sp., *Stuckenia* spp. and the emergent *Hippuris vulgaris* were located to the positive end of CCA2 and markedly associated with lacustrine fringe wetlands.

4. Discussion

Macrophytes assemblages in the studied wetlands were rich and diverse and reflected the landscape attributes. Whereas wetlands had distinctive environmental features from the cordillera to the Plateau that have influence on macrophytes, our analyses suggests that some factors as increased salinity and ammonia levels (eutrophication symptoms), which can be related to grazing pressure, also had consequences on aquatic plant composition across all biozones. Nevertheless, as has been shown in other studies conducted along wide gradients (Miserendino, 2009; Sass et al., 2010) there was an overlapping of natural and anthropogenic effects making difficult to separate controlling factors. Changes in macrophyte life-form and floristic composition were clearly distinguished across environmental gradients related with wetland typologies, hydrological features location and size (depth, altitude and water temperature). As in other landscape approaches conducted in other regions of the world those variables associated to geographic and morphological heterogeneity of studied wetland had a high predictive value on macrophytes assemblages (De Steven and Toner, 2004; Bornette and Puijalon, 2011).

Nutrients levels, conductivity and pH values in water column also appeared as significant factors governing macrophytes composition. Several authors had reported that wetlands influenced by agricultural activity and pasture development often have higher levels of total phosphorus, phosphorus in the sediment, turbidity and conductance than do non disturbed wetlands (Freeland and Richardson, 1997; Helgen and Gernes, 2001; Gleason et al., 2003a,b; Chipps et al., 2006; Bornette and Puijalon, 2011). According to Brock et al. (2005) reduction in the diversity of plants in both freshwater and terrestrial systems has been attributed to salinity increases as a symptom of land use change.

Moreno et al. (2001) stated that salinity is one of the main constraints conditioning diversity and development of macrophytes communities, and Hart et al. (1991) identified this trend in other primary producers such as periphyton. In agreement with these observations James et al. (2009) documented that macrophyte richness tends to decline with increasing salinity in Australian wetlands. In line with our results, Gaitán et al. (2011) found that variables as moisture, alkalinity and salinity conditions of the soil were also important determining distribution of terrestrial plant communities associated with wetlands in a recent study in Patagonia.

A consistent pattern among land use intensity and species richness, native richness and cover of macrophytes was observed at isolated wetlands, suggesting that grazing can have a pervasive effect on aquatic plants at this type of environments. Similar

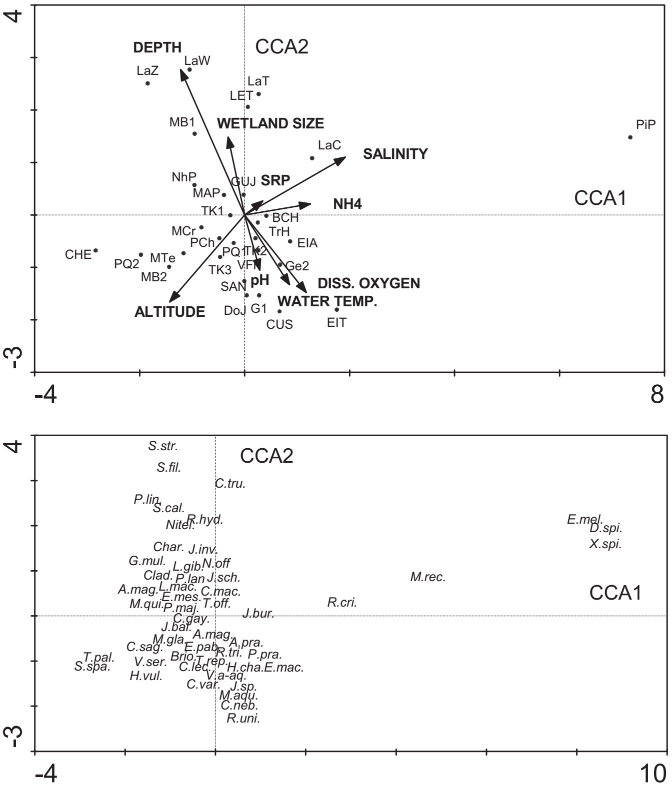


Fig. 6. Canonical Correspondence Analysis ordination plot for (A) sites and environmental variables and (B) aquatic plants species. Ordination performed on 30 wetlands data from Chubut Province, Patagonia, Argentina. (December 2006 and December 2007). Full environmental variables and sampling sites names are in Table 1. Aquatic plants species codes are in Table 2.

results have been observed at littoral zones of water bodies located in developed basins, thus compositional and richness measures appears as consistent metrics in biological surveillance (Rothrock et al., 2008; Sass et al., 2010). The total number of plant species and the proportion of exotic species have resulted good candidates to assess disturbance at floodplain wetlands (Chipps et al., 2006). Nevertheless, in our study none of the different compositional and cover measures showed the expected response at connected wetland. At these places the water movement (longitudinal and lateral), which is a major agent of dispersal of seed or vegetative fragment of aquatic plants appeared to ameliorate the impact of livestock (Bornette and Puijalon, 2011).

Some activities such as plowing, planting, and/or cattle grazing can alter wetland hydrology and affect plant communities (Wilcox, 1995). Chipps et al. (2006) observed that wetland disturbance was associated with reduced plant species richness and a higher proportion of exotic species. In our study the lowest species richness was recorded at the higher disturbed sites, however, exotic richness was significantly higher in connected than isolated sites and did not vary consistently with the level of disturbance. The proportion of exotic species recorded in this work (25%) was similar to that reported by Hauenstein (2006) for Chilean continental aquatic ecosystems (20.7%), and lower than mentioned by Ramírez et al. (1991) (32.5%) for wetlands in recreational areas in Valdivia, Chile. In our inventory exotic macrophytes corresponded to emergent species, that are broadly distributed in wet environments like *T. repens*, *T. officinale*, *V. serpyllifolia*, *Plantago major* and to the submersed *C. truncata*. In addition to the mentioned land forms (*T. repens*, *T. officinale* and *V. serpyllifolia*), the species *M. recutita* and *X. spinosum*, which were present in disturbed sites, are weeds that correspond to indicators for grazing and trampling (Hauenstein et al., 2008).

It is well known that Patagonian steppe environments are subjected to a higher grazing pressure than mountain ones; wetlands in the steppe are highly productive for foraging (Collantes and Faggi, 1999). There is evidence that these environments show a replacement of species, as for example Juncaceae by *Distichlis* sp. This phenomenon appears associated with erosion processes which increase soil salinity. In fact *Distichlis* sp. is considered an indicator species of wetland deterioration and also of eutrophication processes (Hauenstein et al., 2008). Our work is in line with those observations, *Distichlis* was recorded in Piedra Parada (PiP), one of the most saline wetland of our study. Raffaele (1999) suggests that probably Juncaceae and Cyperaceae taxa show a low rate of sprouting under saline conditions, while *Distichlis* sp. grows successfully on saline inundated soils. Nevertheless we also found *Eleocharis melanomphala* (Cyperaceae) associated to Extra-Andean zone (PiP site) suggesting certain plasticity in this cyperacean species.

Schoenoplectus californicus dominated the lacustrine fringe areas and was associated with *Hippuris*, *Myriophyllum* and *Potamogeton* at wetlands in the Andean biozone. Clausen et al. (2006) reported same assemblages mostly in open water areas at higher latitudes. The Patagonian endemic *Samolus spathulatus* appeared constituting compact communities with Cyperaceae and Juncaceae, which is in agreement with observed in isolated wetlands in Aysen (Chile) (Saldivia Pérez, 2006). The submersed *L. macloviana*, would be a tolerant species being recorded in a wide range of environmental conditions and intensity of land uses consistently with reported by Affolter (1985) regarding the dispersion patterns and ecological requirements in this species.

The cosmopolitan *Triglochin palustris* was collected in the highest wetland (918 m.a.s.l.), a site with high level of nutrients. *T. palustris* is frequently associated to rich fens in circumpolar regions (Glaser et al., 1990; Johnson and Steingraeber, 2003). Other wide spread species *Lemna gibba* was recorded only at Pescado site. Canales-Gutiérrez (2010) stated that this species occurs in a wide range of pH and temperature, whereas Hernández and Mitsch (2002) reported *L. gibba* in places showing circumneutral pH values and high nitrate contents, which is similar to our observations.

Given the importance of wetlands in maintaining aquatic biodiversity, the knowledge of the biota and environmental characteristics are crucial for management actions. In particular macrophytes contribute greatly to the structural diversity of wetland environments, providing important refuge areas for macroinvertebrates communities and are food source for water fowl (Cazzanelli et al., 2008). This study provides a first look at natural and anthropogenic controls on aquatic plants in Patagonian wetlands. Although we were not able to clearly separate natural from grazing effects, there was a correspondence between macrophyte assemblages and the level of disturbance. According to our observations isolated wetland were more affected than sites connected by grazing practices.

It is necessary to conduct further studies focused on spatial and temporal issues on these communities to investigate compositional and structural measures of macrophyte communities for biological assessment. Currently there is a strong concern about activities that are changing the land use in Patagonia that could have pervasive effects on wetlands, e.g. replacement of native species by exotic ones with commercial value (*Pinus* sp.) and change of the historical extensive livestock practices for intensive ones (Mazzoni and Vázquez, 2004). It would be desirable that managers and planners understand the functioning of these ecosystems in order to minimize the possible environmental damage and to develop appropriate strategies for wetland conservation in Patagonia.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.ecoleng.2013.12.007. These data include Google maps of the most important areas described in this article.

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