



# Grazing for biodiversity: Assessing the effects of cattle management practices on wetlands and amphibian communities in central Argentina

Maria Gabriela Agostini<sup>a,b,\*</sup>, David Bilenca<sup>a,c</sup>

<sup>a</sup> Universidad de Buenos Aires-CONICET, Instituto de Ecología, Genética y Evolución de Buenos Aires, IIEGEB, CABA, Argentina

<sup>b</sup> COANA. Conservación de Anfibios en Argentina, CABA, Argentina

<sup>c</sup> Universidad de Buenos Aires, Facultad de Ciencias Exactas y Naturales, Departamento de Biodiversidad y Biología Experimental, CABA, Argentina

## ARTICLE INFO

### Keywords:

Pampas  
Anurans  
Livestock  
Grassland  
Conservation

## ABSTRACT

Argentina is widely recognized as a key player in the production and consumption of beef, occupying a prominent position in the global context. However, there is a lack of comprehensive research on the environmental consequences of beef production, particularly concerning wetlands and aquatic biodiversity. We aim to assess the effects of cattle management practices (grazing types and stocking density) on wetland attributes and associated amphibian diversity in central Argentina. During two amphibian breeding seasons, we surveyed 117 wetland sites in paddocks with different cattle grazing types (continuous grazing, glyphosate-promoted pastures and rotational grazing) and stocking densities (low and high). We analyzed the effects of cattle management practices on wetland attributes, including water quality parameters, nutrient concentrations, wetland morphometry, and vegetation cover. Effects on amphibian communities were explored using richness, abundance, and species occurrence. General Mixed-effects Models revealed significant increases in nutrients (total solid dissolved, total nitrogen and soluble reactive phosphorous) and a significant decrease in vegetation cover in sites with high cattle stocking density. Likewise, we observed increased soluble reactive solids in wetlands associated with rotational grazing and decreased vegetation cover in those wetlands related to glyphosate-promoted pastures. Generalized Mixed-effects Models revealed that amphibian communities were negatively affected mainly by high stocking density, while abundance and species occurrence of hylids (*Boana pulchella* and *Scinax squaleirostris*) were also affected by glyphosate-promoted pastures and rotational grazing treatments. Our results indicate that different types of grazing at low stocking density favor amphibian diversity and improve wetland water quality, therefore, reduced stocking density may result in improved wetland conditions and more diverse amphibian communities. Our results also suggest that glyphosate-promoted pastures and rotational grazing, although beneficial in terms of pasture management, may have unintended consequences on water quality in wetlands and amphibian communities. This study will contribute to our understanding of how cattle management practices influence wetland ecosystems and aquatic biodiversity in the most important cattle breeding area of South America, providing valuable insights for conservation efforts in cattle ranching landscapes.

## 1. Introduction

Humans have been transforming the landscape for hundreds of years to obtain food, fibers, fuels, and other goods and services provided by ecosystems (Sanderson et al., 2002). Approximately one-third of the calories produced by the world's crops are currently allocated for feeding animals grown for human consumption, and when pasture and grazing lands are considered, livestock production accounts for approximately 70% of the global agricultural land area (Cassidy et al.,

2013). Thus, the livestock industry is currently the single major driver of habitat loss and degradation, which is, in turn, the primary cause of species declines and extinctions worldwide (Ducatez and Shine, 2017). Amphibians play a leading role in the ongoing biodiversity crisis, as the vertebrate group experiencing the steepest global population decline and the highest species extinction rate (IUCN, 2023). Habitat alterations, especially those related to land use and land cover changes, are among the main causes of amphibian decline (Campbell Grant et al., 2020).

\* Correspondence to: Instituto de Ecología, Genética y Evolución de Buenos Aires, GEBA, Ciudad Universitaria, Pabellón II, Güiraldes 2160, C1428EGA, CABA, Argentina.

E-mail addresses: [gabrielaagostini18@gmail.com](mailto:gabrielaagostini18@gmail.com), [gagostini@ege.fcen.uba.ar](mailto:gagostini@ege.fcen.uba.ar) (M.G. Agostini).

<https://doi.org/10.1016/j.agee.2023.108801>

Received 3 June 2023; Received in revised form 27 October 2023; Accepted 5 November 2023

Available online 17 November 2023

0167-8809/© 2023 Elsevier B.V. All rights reserved.

Globally, amphibian distributions and breeding habitats overlap with lands devoted to livestock grazing. Amphibian-livestock interactions, particularly those involving cattle, have been relatively well-documented in grassland biomes of the Palearctic and Nearctic realms. In a global review, Howell et al. (2019) summarized the amphibian response to the use of wetlands by livestock, concluding that climatic conditions, species habitat type, and the diverse life histories of amphibians make it difficult to predict general responses to livestock grazing. There are many potential pathways for livestock to affect amphibians and their habitat, and its consequences can be positive, negative, or neutral.

Negative impacts include the direct trampling of amphibians by cattle, water quality degradation associated with livestock waste and changes in the hydroperiod and successional trajectory of wetlands (Jansen and Healey, 2003; Cole et al., 2016). Studies have shown a negative association between grazing cattle and amphibian abundance (Riedel et al., 2008), and a positive association between grazing and increased incidence of ranavirus infection (Hoverman et al., 2012) and parasite abundance (McKenzie, 2007). Grazing may also have positive effects in areas with naturally low levels of nutrient inputs or regions with an evolutionary history that includes large grazers (Plăiașu et al., 2010). The combination of increased fertilization and removal of senescent vegetation stimulates new plant growth, which may provide extra habitat and foraging opportunities for amphibian communities in oligotrophic aquatic ecosystems (Denton et al., 1997; Plăiașu et al., 2010).

Argentina is globally known as a beef producer and consumer (Arrieta et al., 2020). The beef industry holds significant importance for the national food system and economy. Over the past three decades, there have been notable changes in Argentine cattle breeding and beef production. Cattle ranching went through a territorial reorganization, partially driven by the expansion of the agricultural frontier (Baeza and Paruelo, 2020). This reorganization resulted in the expansion of cattle towards previously untapped areas, particularly in Chaco, the Espinal, and the Paraná Delta Islands (Graesser et al., 2015), and in increased stocking density/rate in areas already occupied by cattle, many of which were already experiencing overgrazing (Bilenca et al., 2018). Finally, the emergence of confined systems such as feedlots has allowed for a larger concentration of animals and is associated with more localized pollution. This transformative process has primarily occurred in the central part of the country, specifically in the Pampas Region (Viglizzo et al., 2011; Baeza and Paruelo, 2020).

We conducted this study in the Flooding Pampas, the most important cattle grazing region in Argentina, which also includes a vast extension of semi-natural grasslands and wetlands with diverse hydrological regimes (Soriano, 1991). The vegetation characteristics determine that this area is destined for cow-calf rearing with a cattle stocking density ranging from 0.7 to 1 head of cattle ha<sup>-1</sup> (Codesido and Bilenca, 2021). The most common livestock management in the region is continuous grazing in natural grasslands (Gonzalez Fischer and Bilenca, 2020). The alternative management that emerged in recent years involves continuous grazing in natural grasslands in which glyphosate is applied to promote the growth of winter Ryegrass (*Lolium multiflorum*), a non-native annual species (Rodriguez and Jacobo, 2010). A third management type is rotational grazing, where cattle are moved between multiple paddocks to improve grazing efficiency and grass growth. Paddocks are rotated every 15–30 days during spring and summer and every 3–4 months during autumn and winter (Jacobo et al., 2006). In addition to the different management practices, animal stocking density can vary depending on forage quality and rancher decisions. Several authors have suggested that under proper management, cattle ranching could coexist with native flora and fauna, presenting a valuable opportunity to conserve the relicts of the South American temperate grasslands (Bilenca and Minarro, 2004; Cardoni et al., 2015; Codesido and Bilenca, 2021). Nonetheless, none of the previous studies examining how cattle grazing affects ecosystems - and wildlife in particular - have

focused on amphibian responses or on the impact of cattle management on semi-permanent or ephemeral wetlands in the Pampean Region.

To address this knowledge gap, we aim to investigate the effects of livestock management practices (cattle grazing types and stocking density) on wetlands attributes and associated amphibian diversity (species richness, abundance, and species occurrence) in the most important cattle-raising region of Argentina. We first tested the *hypothesis* that cattle management practices modulate wetland attributes in the Flooding Pampas. As previously mentioned, livestock access to wetlands results in water pollution and decreased palatable plant species (Jansen and Healey, 2003; Cole et al., 2016). Therefore, we predict that regardless of grazing type, high stocking density will expose eutrophication indicators (increase of nutrient concentrations and decrease dissolved oxygen) and reduce wetland vegetation cover. Our *second hypothesis* is that cattle grazing types and stocking density have different impacts on amphibian communities in the Flooding Pampas. Some authors have argued that, as the use of glyphosate shifts the floristic composition of grasslands (Rodriguez and Jacobo, 2010), it indirectly affects avian communities by reducing habitat availability (Codesido and Bilenca, 2021). Furthermore, glyphosate has been shown to directly affect amphibians, especially during the larval cycle when this agrochemical reaches water bodies (Lajmanovich et al., 2011; Agostini et al., 2020). We predict that, at equal cattle stocking density, the richness and abundance of amphibian communities in wetlands associated with pastures promoted with glyphosate will be more affected than wetlands associated with other grazing types. Likewise, livestock access to wetlands could reduce the availability of aquatic breeding habitats (Howell et al., 2019). Therefore, we predict high cattle stocking density will negatively affect amphibian richness and abundance. Finally, since the effects of human-mediated changes in aquatic vegetation and water quality usually depend on species' natural history traits and result in species-specific responses (Babbitt et al., 2009; Boissinot et al., 2019; Agostini et al., 2021; Perrone et al., 2022) we *hypothesize* that grazing types and cattle stocking density would have a species-specific impact on occurrence. During the summer and spring seasons, the wetlands of the Flooding Pampas support vegetation communities that provide forage resources (Jacobo et al., 2006) and complex habitats for the reproduction of the amphibian species involved in this study (Agostini et al., 2021). Thus, we predict that high cattle stocking density will primarily reduce the availability of aquatic habitats for breeding and that climbing species (Hylidae family) will be the most affected.

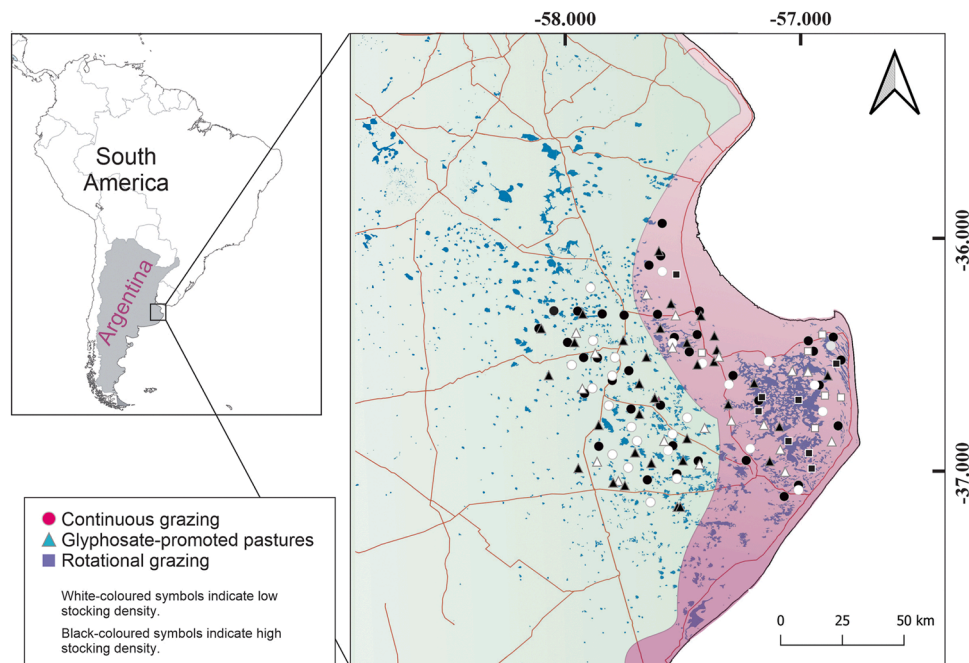
We conducted this study during the breeding season of all the amphibian species involved in this study when livestock has access to wetlands (Agostini et al., 2020; 2021). This approach enabled us to compare the relative use of wetlands by amphibian communities under different cattle management and assess the conservation value of wetlands in cattle ranching landscapes to favor amphibian biodiversity.

## 2. Material and methods

### 2.1. Study area and wetland selection

The study area extends ~11,500 km<sup>2</sup> and comprises 117 sites representing cattle grazing paddocks in which wetland areas are also present (Fig. 1). According to the floristic composition (phytogeographical criteria), the study area comprises a uniform ecological unit named Flooding Pampas (Soriano, 1991). Under the wetland classification criteria, the study area comprises two units: to the east, the Coastal Wetland Region, and to the west, the Pampean Wetland Region (Fig. 1; Benzaquén et al., 2017). The climate is temperate and humid with an average annual rainfall of 900 mm, uniformly distributed throughout the year. The region is frequently exposed to prolonged flooding, due to its depressed relief and low slope (León et al., 1984).

We created a wetland area dataset within the study area by digitizing 2019 aerial photographs in ArcGIS (ESRI, 2011) and recognizing those wetlands areas suitable to conduct the surveys. Then, for each identified



**Fig. 1.** Map of the study area. The green surface indicates the Pampean Wetland Regions, and the purple surface indicates the Coastal Wetland Region, according to Benzaquén et al. (2017). Symbols indicate the 117 wetland sites surveyed.

area, we created a 1 km<sup>2</sup> quadrant grid and then selected 200 at random. When we reached the area, the wetlands were selected based on accessibility and landowner permissions. We only included sites located in paddocks containing natural wetlands surrounded by lands intended for cattle grazing. Amphibians can respond adversely to co-occurrence with fish and proximity to roads (Hartel et al., 2007; Brown et al., 2012). Therefore, aquatic habitats less than 200 m away from main roads or containing large fish (e.g., *Hoplias argentinensis*, *Rhamdia quelen* or *Oligosarcus jenynsi*) were excluded from wetland selection.

Based on the information provided by ranchers and landowners, each site was classified according to the cattle grazing type surrounding the wetlands as follows: continuous grazing (CG), glyphosate-promoted pastures (GlyP), and rotational grazing (RG). It should be noted that surveys in rotational grazing paddocks were conducted only in the Coast Wetland Region. We defined the cattle stocking density as the number of animals (including cows and calves) per unit area at a given time since we conducted the surveys when the cattle were grazing the paddocks. Considering the reported cattle stocking density for the region (0.7–1 head of cattle ha<sup>-1</sup>) and the information provided by ranchers and landowners (See Supplementary material A), each site was also classified into low cattle stocking density (LowSD) when the paddocks had less than 0.6 head of cattle ha<sup>-1</sup> and high cattle stocking density (HighSD) for those paddocks exceeding 0.6 head of cattle ha<sup>-1</sup>. For this study, we included only those sites that had maintained consistent cattle management practices for at least four years.

Surveys were conducted in October/November 2018 and 2019 and February/March 2020 and 2021. To ensure statistical independence, all sites were more than 5 km apart. All sites were sampled three times and measurements were averaged to produce one value per variable.

## 2.2. Wetland attributes

At each sampling site (see wetland selection), a surface area of 250×250 m was selected to collect data on wetland attributes. We recorded 12 variables representing four sets of wetland attributes: water quality (temperature, dissolved oxygen, conductivity, pH, and total dissolved solids), nutrients (nitrites, nitrates, ammonium, total nitrogen and soluble reactive phosphorus), wetland morphometry (average

depth), and pond vegetation cover (submerged and emerged vegetation). Water quality parameters were measured using a Hanna HI98194 multiparameter. Water samples were taken for nutrient analysis and then immediately passed through Whatman GF/C filters having a 1.2-μm pore size (Whatman Incorporated, Clifton, NJ, USA) and transported to the laboratory on ice. Nutrient concentrations were measured following Baird and Bridgewater (2017). We obtained the wetland average depth by measuring the depth in five different locations (randomly selected) across the wetland area defined for each sampling site. To do this, we submerged a 2 m folding meter stick until it reached the bottom of the water body. To determine vegetation cover, we estimated the percent of submerged and emerged vegetation in ten randomly selected quadrants (50×50 cm) across the wetland area defined for each sampling site. Then, we constructed an index for vegetation cover per wetland, following Yin et al. (2000).

## 2.3. Amphibian survey

To optimize species detection, surveys were conducted after heavy rainfall (Agostini et al., 2016, 2021). Sampling was restricted to breeding sites combining two methods for detecting adult amphibians: (i) acoustic surveys were conducted for 5 min in three different locations around the wetland in each site, and (ii) visual encounter surveys were conducted using three fixed transects (50×2 m) per site. Each transect was randomly selected, conducted on foot, and covered both the edges and the interior of each wetland. Surveys were conducted by the same person during warm nights (22.00–02.00) with low wind (≈10 km h<sup>-1</sup>). The minimum air temperature for surveys was 12°C (measured in situ using a Kestrel 5500FW Weather Meter). The number of observed individuals was recorded for each species. The combination of these two methods is widely used to study amphibian communities (Petitot et al., 2014; Boissinot et al., 2019; Agostini et al., 2021; Perrone et al., 2022). We followed Frost (2023) for specific names and systematic approach.

## 2.4. Statistical analyses

Mixed-effects models with normal error structure (identity link function) (Crawley, 2007) were used to determine the effects of cattle

management on wetland attributes. Models (one for each response variable) were constructed considering temperature (T), dissolved oxygen (DO), conductivity (C), pH, total dissolved solids (TDS), nitrite ( $\text{NO}_2^-$ ), nitrate ( $\text{NO}_3^-$ ), ammonium ( $\text{NH}_3$ ), total nitrogen (TN), soluble reactive phosphorus (SRP), average depth (AD), and vegetation cover (VC) as response variables. We incorporate this variable as a random effect because the aquatic environments studied are located in different wetland regions.

To test the effects of cattle management on species richness and abundance, we used generalized linear mixed models (GLMMs; Pinheiro and Bates, 2000; Zuur et al., 2009). Species richness was determined based on acoustic and visual surveys, whereas count abundance was recorded only through visual surveys. Both models were fixed to Poisson error structure (log link function for count data). We performed separate GLMMs for each response variable and we also incorporated wetland region as a random effect. By doing so, we controlled the variability contributed by differences in wetland structure and anuran community composition.

The analysis of species occurrence was limited to those species with a presence in between 10% and 90% of the wetlands sampled (Peduzzi et al., 1996). Of these species, we selected the three most prevalent in the amphibian communities to explore the association of the wetland attributes on the species occurrence. We employed separate GLMs (one for each species in each wetland region) with Bernoulli error structure (logit-link function, for binomial data = species detected/not detected).

All models were constructed considering the grazing management type (CG, GlyP and RG), the cattle stocking density (LowSD and HighSD), and their interactions as explanatory variables. Before conducting analyses, we tested the spatial autocorrelation level for both response and explanatory variables employing Mantel's Test (Mantel, 1967) (Supplementary material A). The significance of the random effect was evaluated with a likelihood ratio (LR), and we used the backward-selection procedure to remove non-significant effects in decreasing order of probability. Then, Tukey's HSD post hoc analyses were conducted to test for treatment differences. We conducted all analyses in the R environment, version 4.3.0 (R Core Team, 2023).

### 3. Results

The cattle management practices involved in this study affected four wetland attributes (TDS, TN, SRP, and VC). The TDS and TN values were only affected by cattle stocking density (EST: 0.05, STD: 0.02,  $t$ : 2.43,  $p$  < 0.05; EST: 0.09; STD: 0.04,  $t$ : -1.98,  $p$  < 0.05). Additionally, both cattle grazing types and stocking density affected SRP (EST: 1.16, STD: 0.07,  $t$ : 2.43,  $p$  < 0.05; EST: 2.48, STD: 0.40,  $t$ : 6.08,  $p$  < 0.05) and VC (EST: -0.74, STD: 0.32,  $t$ : -1.59,  $p$  < 0.05; EST: -0.11, STD: 0.32,  $t$ : 3.09,  $p$  < 0.05). We did not detect random effects, so the wetland region variable was removed from the model. After removing random effects, Tukey's HSD post hoc analyses showed that TDS and TN values increased significantly in all HighSD treatments with respect to the LowSD treatments for the same grazing type (Fig. 2A-B). SRP values also showed a significant increase in all HighSD treatments with respect to the LowSD treatments for the same grazing type. The significantly higher values were also observed in RG-HighSD compared to CG-HighSD and GlyP-HighSD treatments (Fig. 2C). Finally, we detected lower values of vegetation cover in all HighSD treatments compared with LowSD for the same grazing type. This value also decreased in the GlyP-LowSD treatment compared to the other cattle grazing types (Fig. 2D). The dataset of wetlands attributes is detailed in Supplementary material B.

During two breeding seasons, we detected a total of 13 species belonging to five anuran families (Table 1). All species were present in both wetland regions except for *C. ornata* which was only found in Coastal Wetland. All species were found in sites with all management types. The species abundance ranked differently for both wetland regions (see Table 1).

GLMMs results showed an effect of cattle stocking density on

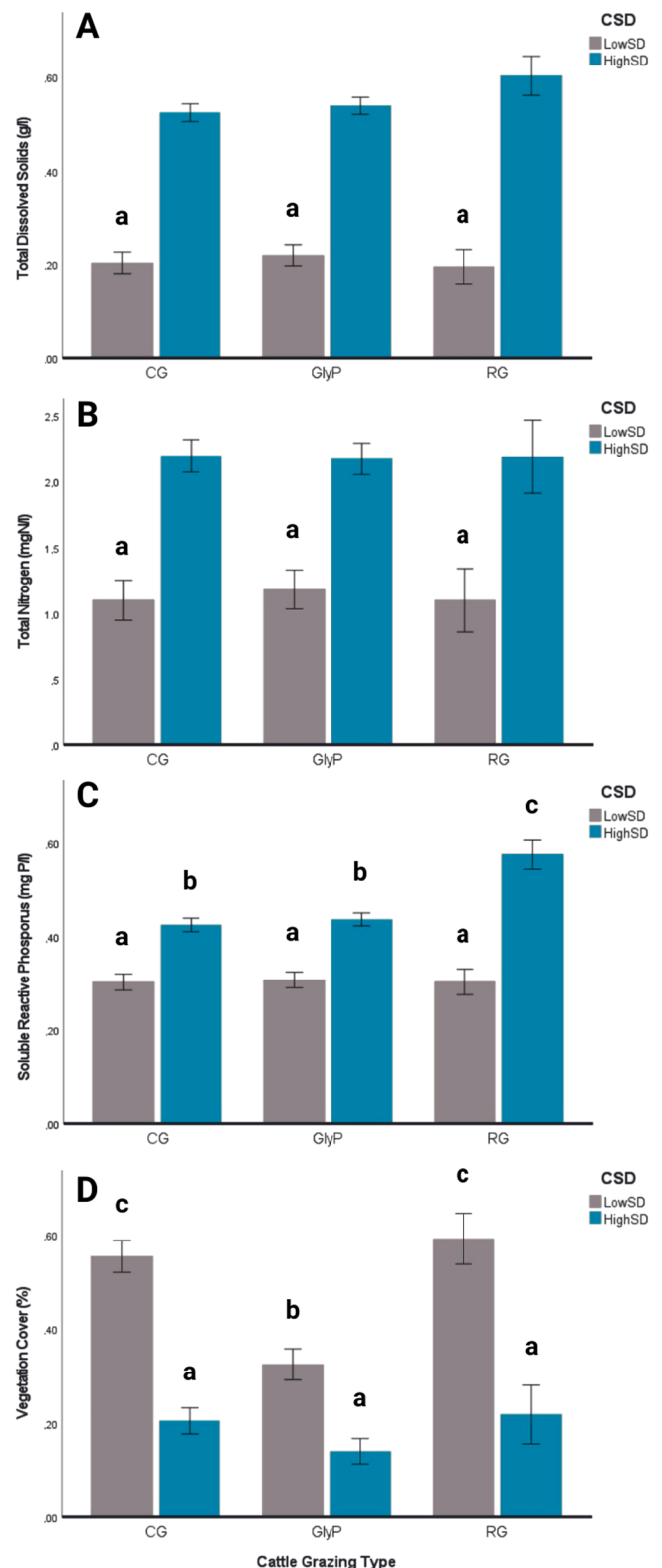


Fig. 2. Bar plots showing the four wetland attributes affected by cattle grazing type or/and cattle stocking density (CSD). The transverse lines inside the bars represent error bars (including media values and 95% confidence intervals). A: Total Dissolved Solids. B: Ammonium. C: Soluble Reactive Phosphorus. D: Vegetation Cover. CG: Continuous grazing. GlyP: Glyphosate-promoted pastures. RG: Rotational grazing. Letters (a, b, and c) indicate significant differences between treatments ( $p$  < 0.05).



**Table 1**

Species composition of the amphibian communities in 117 wetland sites from the Flooding Pampas of central Argentina. Species occurrence is expressed as % of occupied ponds and the abundance is expressed as means (and standard errors) of the total individuals sampled.

Amphibian species	Pampean Wetlands (N=55)		Coastal Wetlands (N=62)	
	Species occurrence	Abundance	Species occurrence	Abundance
Bufonidae				
<i>Rhinella arenarum</i>	6.3	4.6 (±3.1)	7.4	3.1 (±2.5)
<i>Rhinella dorbignyi</i>	77.5†	20.7 (±9.6)	60.5	19.3 (±6.3)
Ceratophryidae				
<i>Ceratophrys ornata</i>	-	-	12.9	6.5 (±5.9)
Hylidae				
<i>Boana pulchella</i>	91.6†	24.7 (±9.2)	86.4†	29.9 (±13.6)
<i>Pseudis minuta</i>	31.6	5.8 (±6.9)	27.2	7.1 (±5.4)
<i>Scinax granulatus</i>	21.7	5.7 (±2.9)	18.9	3.6 (±2.2)
<i>Scinax squalirostris</i>	53.9	13.2 (±6.5)	79.6†	15.8 (±6.9)
Leptodactylidae				
<i>Leptodactylus gracilis</i>	5.3	1.3 (±0.9)	1.3	0.9 (±0.1)
<i>Leptodactylus latinasus</i>	12.9	2.1 (±1.0)	2.7	2.5 (±0.9)
<i>Leptodactylus luctator</i>	60.3†	9.1 (±3.6)	55.3	7.1 (±5.7)
<i>Physalaemus fernandezae</i>	39.2	5.5 (±1.9)	43.5	6.8 (±5.3)
<i>Pseudopaludicola falcipes</i>	57.8	7.3 (±2.9)	54.8	9.2 (±5.9)
Odontophrynidae				
<i>Odontophrynus asper</i>	51.2	7.8 (±2.5)	63.6†	8.1 (±3.0)

† Species used for those models testing the influence of different cattle management practices on amphibian species occurrence. Species occurrence was obtained using acoustic and visual surveys and abundance was registered using visual encounter surveys.

richness, while no effect of grazing type was detected (Table 2). Under the same grazing type, higher stocking densities (HighSD treatments) were associated with lower amphibian species richness (Fig. 3A). Both grazing type and cattle stocking density emerged as significant variables affecting species abundances (Table 2). A posteriori test showed that HighSD treatments were associated with lower richness values compared to the LowSD treatment for the same grazing type (Fig. 3B). Likewise, amphibian abundance values in the RG-HighSD treatment

**Table 2**

Results of generalized linear mixed models. Coefficients estimates, standard error and z-values for explanatory variables: cattle grazing types (CG = Continuous grazing, GlyP = glyphosate-promoted pastures, RG = rotational grazing) and cattle stocking density (LowSD, HighSD) influencing the richness and total abundance of amphibian communities in 117 wetland sites of the Flooding Pampas. Results of full models are presented. Significant explanatory variables are in bold \* $P < 0.05$ , \*\* $P < 0.01$ , \*\*\* $P < 0.001$ ; n.s., not significant.

Parameter	Effect	Estimate	Std. Error	z-value	P
<b>Richness</b>	Intercept	0.12	0.28	0.40	n.s.
	GlyP	-0.18	0.20	-0.88	n.s.
	RG	-0.05	0.05	-0.92	n.s.
	<b>HighSR</b>	<b>-0.56</b>	<b>0.01</b>	<b>-35.1</b>	<b>***</b>
	GlyP* HighSD	0.26	0.22	1.14	n.s.
	RG *HighSD	-0.41	0.08	-0.55	n.s.
<b>Total Abundance</b>	Intercept	<b>1.69</b>	<b>0.38</b>	<b>3.35</b>	<b>**</b>
	GlyP	-0.17	0.10	-1.58	n.s.
	RG	<b>-0.86</b>	<b>0.40</b>	<b>-2.13</b>	<b>*</b>
	<b>HighSR</b>	<b>-8.04</b>	<b>0.97</b>	<b>-8.27</b>	<b>***</b>
	GlyP* HighSD	0.02	0.15	0.17	n.s.
	RG *HighSD	-0.52	0.47	-1.09	n.s.

CG and LowSD treatments are included in the intercept.

were lower than those observed in GlyP-HighSD and CG-HighSD treatments (Fig. 3B).

Since species occurrence and abundance differed between the two wetland regions (see Table 1), the occurrence species analysis was performed separately. The most prevalent species in the Pampean Wetland Region were *Boana pulchella*, *Rhinella dorbignyi*, and *Leptodactylus luctator*, while *B. pulchella*, *Scinax squalirostris* and *Odontophrynus asper* were the most prevalent species in the Coastal Wetland Region.

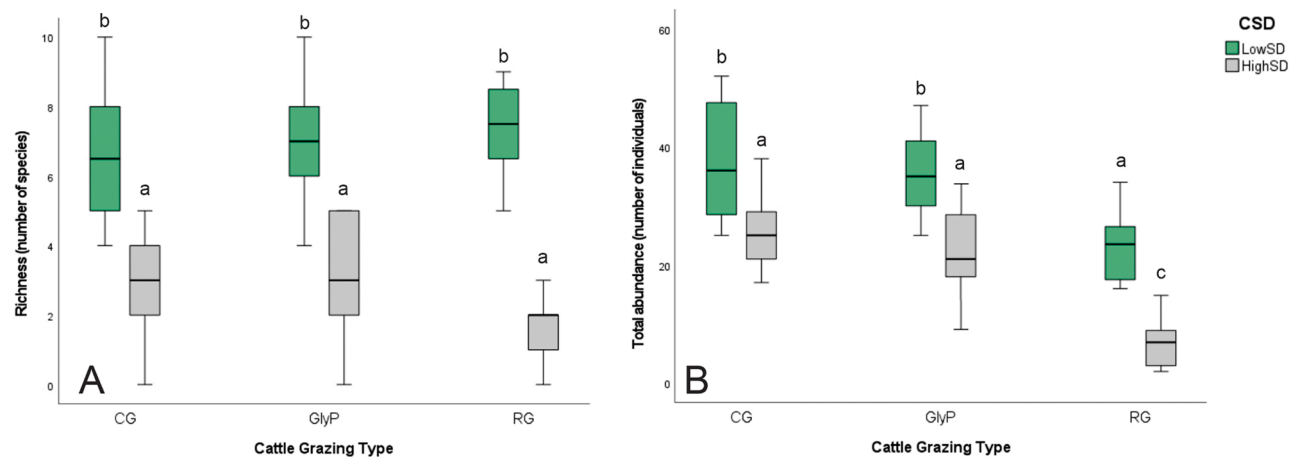
In the Pampean Wetland Region, the occurrence of *B. pulchella* and *L. luctator* was negatively affected by HighSD compared to LowSD of the same grazing type. Furthermore, the occurrence of both species was significantly lower in the GlyP-HighSD treatment compared with all the treatments. The *R. dorbignyi* occurrence was only affected by cattle stocking density, being lower in all HighSD treatments (Fig. 4A, Supplementary material A). Finally, in aquatic habitats of the Coastal Wetland Region, the occurrence of *B. pulchella* and *S. squalirostris* was negatively affected by HighSD compared to LowSD (same grazing type). Considering all the treatments, the lower occurrence values for both species were detected in RG-HighSD (additive effect). *Odontophrynus asper* occurrence was negatively affected by cattle stocking density, being lower in all HighSD treatments (Fig. 4B, Supplementary material A).

#### 4. Discussion

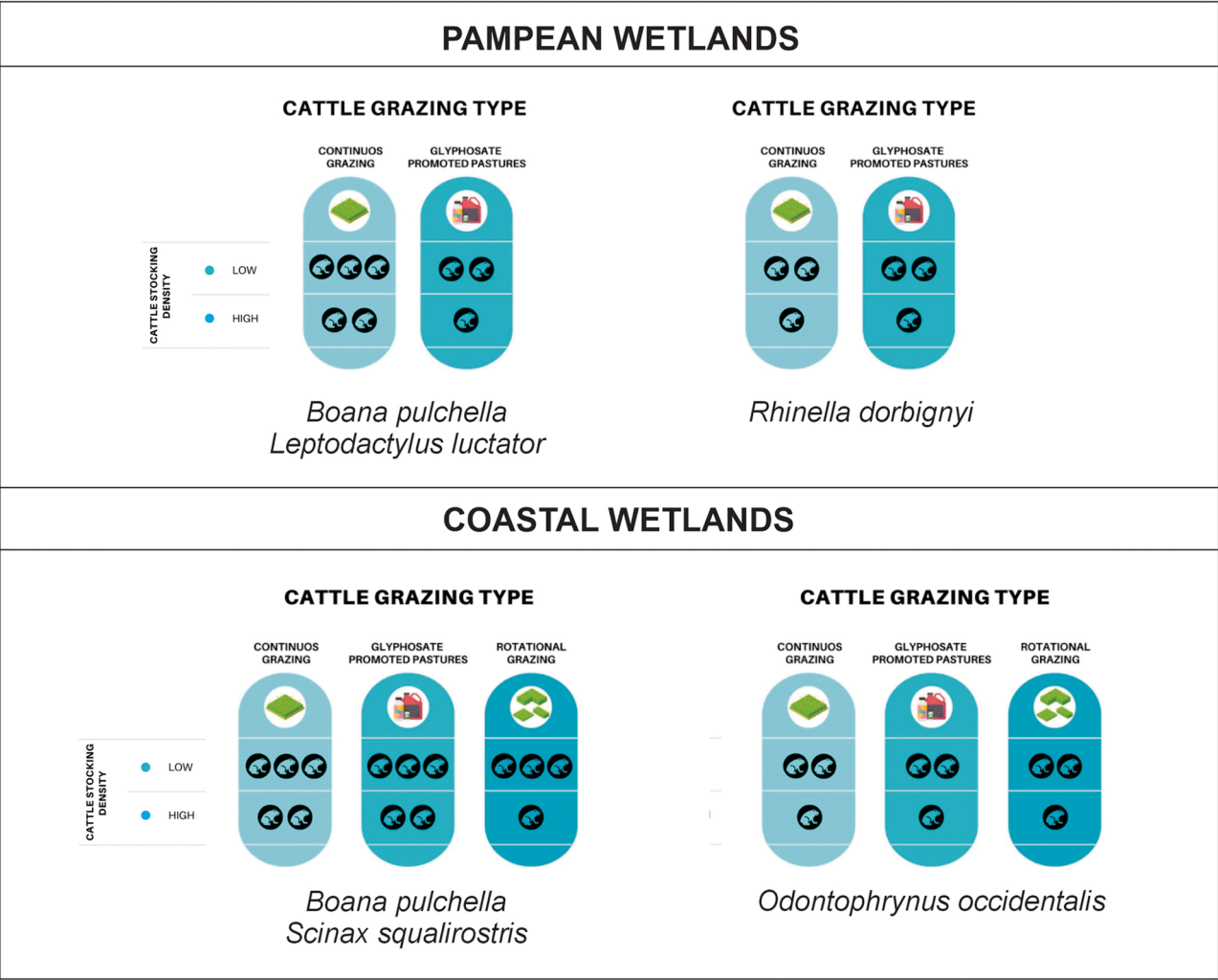
Although Argentina stands as one of the leading countries in beef production, the ecological consequences of cattle on biodiversity, particularly in aquatic environments, remain understudied (Mayora et al., 2021; Seimandi et al., 2021). Globally, conclusive evidence on how grazing affects amphibians is lacking. While open-canopy amphibians are likely to experience positive effects from the presence of livestock, the grazing effects also depend on various factors, including geographical locations and management practices (Howell et al., 2019). Furthermore, the intricate physiology and diverse life histories of amphibians render it challenging to predict their general responses to livestock grazing (Babbitt et al., 2009). Here, we present pioneering evidence elucidating the impact of cattle grazing on wetlands associated with the Argentinean Pampas grasslands, shedding light on their implications for native amphibian communities. Our comprehensive findings reveal that, among the various cattle management practices used in the region, the stocking density emerges as the main modulator of amphibian diversity in the most important cattle-raising region of Argentina, a pattern consistently observed across the multiple wetland attributes studied.

##### 4.1. Effects of cattle management practices on wetlands

We predicted that wetland parameters reflecting water eutrophication (nutrient concentrations and DO) would primarily be affected by high cattle stocking density. Our results partially confirmed the prediction and showed that total dissolved solids, total nitrogen, and soluble reactive phosphorus increased in those sites where cattle access the wetlands at high stocking density. The elevated nutrient concentrations measured in our study can be attributed to the substantial amount of feces that enter aquatic systems when livestock are concentrated. Cow manure (feces and urine) contains organic matter, water, and nutrients such as nitrogen, phosphorus, and potassium (Chaneton and Lavado, 1996; Tweel and Bohlen, 2008). In aquatic environments, both cow feces and urine undergo biodegradation processes; however, excessive amounts of manure have been proven to lead to aquatic eutrophication, where the excess nutrients cause an overgrowth of algae, depleting oxygen levels and harming aquatic life (Schmutzer et al., 2008; Kronberg et al., 2021). Based on this, we expected to find decreases in dissolved oxygen levels in those sites exposed to high cattle loads. The observed results, including no significant difference in dissolved oxygen values,



**Fig. 3.** Boxplot of amphibian richness (A) and total abundance (B) in wetlands under different livestock management practices (cattle grazing types and cattle stocking density -CSD-). The transverse lines inside the boxes represent median values; lower and upper hinges correspond to the first and third quartiles. Whiskers represent the maximum and minimum range (excluding outlier values). CG: Continuous grazing. GlyP: Glyphosate-promoted pastures. RG: Rotational grazing. Letters (a, b, and c) indicate significant differences between treatments ( $p < 0.05$ ).



**Fig. 4.** Results of GLMMs showing significant differences in cattle grazing types and cattle stocking density on the occurrence of five amphibian species from Pampean Wetland and Coastal Wetland Regions.

could be explained as a consequence of the sampling design, which involved surveys conducted after heavy rainfall. We expect that with time following rainfall events, the degradation processes of manure will exacerbate the eutrophication of the water, leading to a more pronounced decline in oxygen levels at sites subjected to high stocking density. Finally, our findings substantiate the detrimental impact of high stocking density on wetland vegetation cover, likely mediated by two processes involving direct grazing on highly palatable plant species and trampling (Seimandi et al., 2021).

When we evaluated differences among grazing types, we observed a reduction of vegetation cover in those wetlands related to glyphosate-promoted pastures. This is consistent with several studies demonstrating that this broad-spectrum herbicide reduces aquatic vegetation when reaching wetlands, whether through spray, drift, or runoff (Richmond, 2018).

#### 4.2. Effects of cattle management practices on amphibians

Glyphosate, the active ingredient in many herbicides, has been a topic of concern due to its potential effects on amphibians (Mann et al., 2009). Research conducted under controlled laboratory conditions has shown that Glyphosate (active ingredient and commercial formulations) and its degradation products (AMPA) can have a range of impacts on amphibians at different stages of their life cycles, including genotoxic and teratogenic effects (Lajmanovich et al., 2003; 2011; Mann et al., 2009). Furthermore, studies carried out under real exposure conditions (when glyphosate enters the aquatic environmental matrix) proved the existence of sublethal effects in amphibian species native to central Argentina (Agostini et al., 2020). Contrary to our prediction, we did not detect a significant decrease in amphibian richness or abundance in wetlands exposed to glyphosate. This unexpected finding could be attributed to the timing of glyphosate applications, which predominantly occur in late summer (Rodríguez and Jacobo, 2010). This period does not coincide with the reproductive activity for most of the amphibian species considered in this study. Thus, effects may not become severe enough to affect detections during the breeding season. To gain a comprehensive understanding of the impact of glyphosate on amphibian communities occurring in wetlands associated with glyphosate-promoted pastures, future research should focus on assessing the effects of this herbicide on aquatic species during late summer and autumn. Species such as *Boana pulchella*, *Scinax squalirostris*, and *Pseudis minuta* could serve as valuable indicators for studying the potential consequences of glyphosate exposure associated with this pasture management since larval stages inhabit wetlands where agrochemical applications are placed.

The findings of this study demonstrate that high cattle stocking densities have a detrimental impact on amphibian richness, abundance, and wetland attributes. Several underlying mechanisms may explain these effects. Firstly, the co-occurrence of livestock and reproductive activities in wetlands may lead to adult exclusion and interfere with male reproductive chorusing (Howell et al., 2009). Secondly, overgrazing can lead to changes in vegetation structure and composition, which in turn decreases the availability of suitable breeding habitats for multiple amphibian species (Jansen and Healey, 2003). Lastly, elevated cattle stocking density is likely to adversely affect recruitment by increasing embryo and tadpole mortality (Schmutzer et al., 2008; Cole et al., 2016).

Comparing management practices with high cattle densities, rotational grazing was found to negatively affect amphibian abundance. It is worth noting that this study did not quantitatively assess the specific number of cows entering wetlands but rather categorized them into high- and low-stocking density groups. However, rotational management involves concentrating a large number of animals in relatively small plots, exploiting the forage resources provided by wetlands (referred to as “bajos dulces”) during the spring and summer seasons (Rodríguez and Jacobo, 2010), which coincide with the reproductive

activity of all species involved in this study. Consequently, the aforementioned factors, such as exclusion and interference of reproductive chorusing, may be intensified, thereby providing an explanation for the observed results. Finally, since changes in wetland attributes may influence the detectability of species (visual surveys), it could be considered a limitation of the sampling design that cannot be completely controlled. For example, decreases in vegetation cover (observed in glyphosate promoted-pasture and high stocking density treatments) could increase detectability. To overcome these limitations, we employed other sampling techniques (auditory surveys) which are used in previous studies (Agostini et al., 2021; Perrone et al., 2022).

The presence of *Ceratophrys ornata* associated only with the Coastal Wetland Region may be explained by the population decline and local extinction that this species is suffering in several areas of its historical distribution, including the Pampean Wetland region (Deutsch et al., 2017; Deutsch et al., 2023). The Coastal Wetland Region has been identified as a priority conservation area for the species (Deutsch et al., 2017), and recent studies have shown a high specificity for native grassland habitats and well-preserved wetlands (Deutsch et al., *in prep*). Future investigations are needed to evaluate the effects of cattle management practices on the occurrence of *C. ornata* aimed to provide recommendations for conserving the remnant populations of this species in Argentina.

In agreement with several studies, the occurrence of the five most representative species within the communities exhibited species-specific responses to livestock management (Babbitt et al., 2009; Boissinot et al., 2019; Agostini et al., 2021; Perrone et al., 2022). Nevertheless, in a general sense, the high cattle stocking density significantly impacted the occurrence of all species when compared to low cattle stocking density within the same grazing type (see Fig. 4). Also, in the Pampean Wetlands, we detected an effect of grazing type on *B. pulchella* and *L. luctator* where wetlands associated with glyphosate-promoted pastures exhibited lower species occurrence. Although we did not find effects of this type of management on most wetland attributes or amphibian richness and abundance, it is plausible that factors occurring outside the study period (e.g., larval exposure to glyphosate, winter grazing exclusion) could show different results than those obtained during the reproductive periods. This emphasizes the need to employ diverse measures of biodiversity, including species-specific responses and various life cycle stages, when evaluating the potential effects of cattle disturbances. In the case of the two most prevalent species from the Coastal Wetlands Region (*B. pulchella* and *Scinax squalirostris*), it was found that cattle management involving rotational practices and high stocking density had a detrimental impact on their occurrence. This outcome was also corroborated through the assessment of wetland parameters (see Fig. 2) and amphibian abundance (see Fig. 3). Our findings are consistent with previous studies that have demonstrated the heightened sensitivity of climbing species, specifically Hylidae, to reductions in vegetation cover (Agostini et al., 2021; Perrone et al., 2022). Overgrazing practices may contribute to the decline in available microhabitats essential for amphibian breeding.

#### 4.3. Recommendations and conservation remarks

The Flooding Pampas harbors the most well-preserved grassland in South America (Bilenca and Minarro, 2004; Baeza and Paruelo, 2020). Recent literature highlights the ability of these moderately anthropized habitats to support moderate to highly diverse vertebrate assemblages (Codesido et al., 2013; Agostini et al., 2020; Codesido and Bilenca, 2021). Nonetheless, most grassland remnants are private lands under the imminent threat of agricultural conversion, which has been dramatically increasing in recent decades (Arrieta et al., 2020). Additionally, meat production is likely to increase globally over the next several decades because of population growth and income-dependent dietary shifts towards more meat-based diets (Clark and Tilman, 2017). Therefore, the challenge is how to conserve grassland

biodiversity in productive landscapes in the context of agricultural expansion and livestock intensification (Bilenca et al., 2018).

Meat production alters habitats not only via land-use changes but also through its outputs of agrochemicals, nutrients, sediments, antibiotics and hormones into wetlands (Howell et al., 2019). Our results suggest that future management efforts to conserve amphibians in the Flooding Pampas should strive to protect wetlands and subject them to minimal cattle use during critical periods (reproduction and larval cycle). Moreover, we highlight the urgency of specific legislation for the protection of natural wetlands neglected by current laws (Straccia and Isla, 2020). We also emphasize the importance of multi-institutional actions from the production sector to promote cattle management practices that minimize risks to biodiversity. Since aquatic habitats are inextricably linked to their surroundings, wetland conservation must be pursued in the context of an integrated systems approach to environmental conservation and sustainable development. This is particularly important for amphibians since many species require both terrestrial and aquatic habitats during their life cycle (Beebe and Griffiths, 2005).

## 5. Conclusion

Despite significant changes to the native landscapes, cattle ranches in central Argentina can support rich and abundant amphibian assemblages. This study reveals that the most common cattle management practices affect amphibian diversity differently and that regulating stocking density could greatly benefit wetland water quality and amphibian diversity. However, the effective implementation of management strategies may require not only an understanding of whether cattle grazing influences local biodiversity but also which characteristics of wetlands must be preserved or restored in order to achieve specific conservation goals.

## Funding

This paper was funded by Neotropical Grassland Conservancy, ANPCyT (PICT 2018–00839) and UBACyT (20020190100244BA).

## Declaration of Competing Interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: M. Gabriela Agostini reports financial support was provided by Neotropical Grassland Conservancy. M. Gabriela Agostini reports financial support was provided by ANPCyT (PICT 2018–00839). David N. Bilenca reports financial support was provided by UBACyT (20020190100244BA).

## Data availability

Data will be made available on request.

## Acknowledgments

We thank Camila Deutsch, Isis Ibañez, Sofia Perrone and COANA volunteers for field assistance. Our thanks to landowners, farm employees, and managers who allowed access to their property and provided us with information on cattle management, especially Pablo Preliasco, Sergio Campos, Nicolás Gonzales, Federico Quiroga y Patricio McLaughlin. Our special thanks to C. Gonzalez-Fisher for language editing. Finally, we thank the associate editor and two anonymous reviewers who made valuable edits and comments that improved this paper.

## Appendix A. Supporting information

Supplementary data associated with this article can be found in the

online version at doi:10.1016/j.agee.2023.108801.

## References

- Agostini, M.G., Roesler, I., Bonetto, C., Ronco, A.E., Bilenca, D.N., 2020. Pesticides in the real world: The consequences of GMO-based intensive agriculture on native amphibians. *Biol. Conserv.* 41, e108355 <https://doi.org/10.1016/j.biocon.2019.108355>.
- Agostini, M.G., Deutsch, C., Bilenca, D.N., 2021. Differential responses of anuran assemblages to land use in agro-ecosystems of central Argentina. *Agr. Ecosyst. Environ.* 311, e107323 <https://doi.org/10.1016/j.agee.2021.107323>.
- Agostini, M.G., Saibene, P.E., Roesler, C.I., Bilenca, D.N., 2016. Amphibians of northwestern Buenos Aires province, Argentina: checklist, range extensions and comments on conservation. *Check List* 12 (6), 1–10. <https://doi.org/10.15560/12.6.1998>.
- Arrieta, E.M., Cabrol, D.A., Cuchieta, A., González, A.D., 2020. Biomass consumption and environmental footprints of beef cattle production in Argentina. *Agr. Syst.* 185, e102944 <https://doi.org/10.1016/j.agsy.2020.102944>.
- Babbitt, K.J., Baber, M.J., Childers, D.L., Hocking, D., 2009. Influence of agricultural upland habitat type on larval anuran assemblages in seasonally inundated wetlands. *Wetlands* 29 (1), 294–301. <https://doi.org/10.1672/07-228.1>.
- Baeza, S., Paruelo, J.M., 2020. Land use/land cover change (2000–2014) in the Rio de la Plata grasslands: an analysis based on MODIS NVDI time series. *Remote Sens* 12, e381. <https://doi.org/10.3390/rs12030381>.
- Baird, R., Bridgewater, L., 2017. *Standard Methods for the Examination of Water and Wastewater*, 23rd ed. American Public Health Association., Washington, D.C.
- Beebe, T.J.C., Griffiths, R.A., 2005. The amphibian decline crisis: A watershed for conservation biology? *Biol. Conserv.* 125, 271–285. <https://doi.org/10.1016/j.biocon.2005.04.009>.
- Benzaquén, L., Blanco, D., Bo, R., Kandus, P., Lingua, G., Minotti, P., Quintana, R., 2017. Regiones de Humedales de la Argentina. MAYDS, Wetlands International, Universidad Nacional de San Martín and Universidad de Buenos Aires, Buenos Aires.
- Bilenca, D., Codesido, M., Abba, A., Agostini, M.G., Corriale, M.J., González Fischer, C., Pérez Carusi, L., Zufiaurre, E., 2018. Conservación de la Biodiversidad en Sistemas Pastoriles. Buenas Prácticas para una Ganadería Sustentable de Pastizal. Kit de Extensión para las Pampas y Campos. Fundación Vida Silvestre Argentina. Buenos Aires.
- Bilenca, D., Minarro, O.F., 2004. Áreas Valiosas de Pastizal (AVPs) en las Pampas y Campos de Argentina. Uruguay y sur de Brasil. Fundación Vida Silvestre Argentina. Buenos Aires.
- Boissinot, A., Besnard, A., Lourdais, O., 2019. Amphibian diversity in farmlands: Combined influences of breeding-site and landscape attributes in western France. *Agr. Ecosyst. Environ.* 269, 51–56. <https://doi.org/10.1016/j.agee.2018.09.016>.
- Brown, D.J., Street, G.M., Nairn, R.W., Forstner, M.R.J., 2012. A place to call home: Amphibian use of created and restored wetlands. *Int. J. Ecol.* 1, e989872 <https://doi.org/10.1155/2012/989872>.
- Campbell Grant, E.H., Miller, D.A., Muths, E., 2020. A synthesis of evidence of drivers of amphibian declines. *Herpetologica* 76, 101–107. <https://doi.org/10.1655/0018-0831-76.2.101>.
- Cassidy, E.S., West, P.C., Gerber, J.S., Foley, J.A., 2013. Redefining agricultural yields: from tonnes to people nourished per hectare. *Environ. Res. Lett.* 8 (3), e034015 <https://doi.org/10.1088/1748-9326/8/3/034015>.
- Cardoni, D.A., Isach, J.P., Iribarne, O., 2015. Avian responses to varying intensity of cattle production in *Spartina densiflora* saltmarshes of south-eastern South America. *Emu* 115, 12–19. <https://doi.org/10.1071/MU13028>.
- Chaneton, E.J., Lavado, R.S., 1996. Soil nutrients and salinity after long-term grazing exclusion in a Flooding Pampa grassland. *J. Rang. Manag.* 49, 182–187. <https://doi.org/10.2307/4002692>.
- Clark, M., Tilman, D., 2017. Comparative analysis of environmental impacts of agricultural production systems, agricultural input efficiency, and food choice. *Environ. Res. Lett.* 12, e064016 <https://doi.org/10.1088/1748-9326/aa6cd5>.
- Codesido, M., Bilenca, D.N., 2021. Avian assemblages associated with different grasslands management in cattle production systems in the pampas of Argentina. *PECON* 19, 464–474. <https://doi.org/10.1016/j.pecon.2021.07.003>.
- Codesido, M., González-Fischer, C.M., Bilenca, D.N., 2013. Land bird assemblages in different agricultural landscapes: a case study in the pampas of central Argentina. *Condor* 115, 8–16. <https://doi.org/10.1525/cond.2012.120011>.
- Cole, E.M., Hartman, R., North, M.P., 2016. Hydroperiod and cattle use associated with lower recruitment in an r-selected amphibian with a declining population trend in the Klamath Mountains, California. *J. Herpetol.* 50 (1), 37–43. <https://doi.org/10.1670/14-014>.
- Crawley, M.J., 2007. *The R Book*. John Wiley & Sons, Chichester.
- Denton, J.S., Hitchings, S.P., Beebe, T.J.C., Gent, A., 1997. A recovery program for the Natterjack Toad (*Bufo calamita*) in Britain. *Conserv. Biol.* 11, 1329–1338. <https://www.jstor.org/stable/1341210>.
- Deutsch, C., Bilenca, D., Agostini, M.G., 2017. In search of the horned frog (*Ceratophrys ornata*) in Argentina: Complementing field surveys with citizen science. *Herpetol. Conserv. Biol.* 12 (3), 664–672. [https://www.herpeconbio.org/Volume\\_12/Issue\\_3/Deutsch\\_et\\_al\\_2017.pdf](https://www.herpeconbio.org/Volume_12/Issue_3/Deutsch_et_al_2017.pdf) (Available at).
- Ducatez, S., Shine, R., 2017. Drivers of extinction risk in terrestrial vertebrates, 186–19. *Conserv. Lett.* 10 (2). <https://doi.org/10.1111/conl.12258>.
- Deutsch, C., Bilenca, D.N., Zurano, J.P., Marin da Fontee, L., Dallagnol Vargas, N., Kindel, A., Pittella, R., Duarte Freire, M., Maneyro, R., Faivovich, J., Agostini, M.G., 2023. Habitat loss and distribution of the Ornate Horned Frog (*Ceratophrys ornata*):



- Implications for its conservation in South American temperate grasslands. PECON In press.
- ESRI, 2011., ArcGIS Desktop: Release 10. Environmental Systems Research Institute, Redlands.
- Frost, D.R., 2023. Amphibian Species of the World: An Online Reference. (<https://amphibiansoftheworld.amnh.org/index.php>) (accessed 20 February 2023).
- Gonzalez Fischer, C., Bilenca, D.N., 2020. Can we produce more beef without increasing its environmental impact? Argentina as case study. PECON 18, 1–11. <https://doi.org/10.1016/j.pecon.2019.12.002>.
- Graesser, J., Aide, J.M., Grau, H.R., Ramankutty, N., 2015. Cropland/pastureland dynamics and the slowdown of deforestation in Latin America. Environ. Res. Lett. 10, e034017 <https://doi.org/10.1088/1748-9326/10/3/034017>.
- Hartel, T., Nemes, S., Cogălniceanu, D., Öllerer, K., Schweiger, O., Moga, C., Demeter, L., 2007. The effect of fish and aquatic habitat complexity on amphibians. Hydrobiologia 583, 173–182. <https://doi.org/10.1007/s10750-006-0490-8>.
- Hoverman, J.T., Gray, M.J., Miller, D.L., Haislip, N.A., 2012. Widespread occurrence of ranavirus in pond-breeding amphibian populations. EcoHealth 9 (1), 36–48. <https://doi.org/10.1007/s10393-011-0731-9>.
- Howell, H.J., Mothes, C.C., Clements, S.L., Catania, S.V., Rothermel, B.B., Searcy, C.A., 2019. Amphibian responses to livestock use of wetlands: new empirical data and a global review. Ecol. Appl. 29 (8), e01976 <https://doi.org/10.1002/eap.1976>.
- IUCN, 2023. The IUCN Red List of Threatened Species. Version 2022-2. (<https://www.iucnredlist.org/>) (accessed 2 February 2023).
- Jacobo, E.J., Rodríguez, A.M., Bartoloni, N., Deregibus, V.A., 2006. Rotational grazing affects on rangeland vegetation at a farm scale. Rangel. Ecol. Manag. 59, 249–257. <https://doi.org/10.2111/05-129R1.1>.
- Jansen, A., Healey, M., 2003. Frog communities and wetland condition: relationships with grazing by domestic livestock along an Australian floodplain river. Biol. Conserv. 109 (2), 207–219. [https://doi.org/10.1016/S0006-3207\(02\)00148-9](https://doi.org/10.1016/S0006-3207(02)00148-9).
- Kronberg, S.L., Provenza, F.D., van Vliet, S., Young, S.N., 2021. Review: Closing nutrient cycles for animal production – Current and future agroecological and socio-economic issues. Animal 15 (1), e100285. <https://doi.org/10.1016/j.animal.2021.100285>.
- Lajmanovich, R.C., Attademo, A.M., Peltzer, P.M., Jungues, C., Cabagna, M., 2011. Toxicity of four herbicide formulations with Glyphosate on *Rhinella arenarum* (Anura: Bufonidae) tadpoles: B-esterases and glutathione S-transferase inhibitions. Arch. Environ. Con. Tox 60, 681–689. <https://doi.org/10.1007/s00244-010-9578-2>.
- Lajmanovich, R.C., Lorenzatti, E., Maitre, M.I., Enrique, S., Peltzer, P., 2003. Comparative acute toxicity of the commercial herbicides glyphosate to neotropical tadpoles *Scinax nasicus* (Anura: Hylidae). Fres. Environmen. Bull. 12 (4), 364–367.
- León, R.J.C., Rusch, G.M., Oesterheld, M., 1984. Pastizales pampeanos – impacto agropecuario. Phytocoenologia 12, 201–218.
- Mann, R.M., Hyne, R.V., Choung, C.B., Wilson, S.P., 2009. Amphibians and agricultural chemicals: review of the risks in a complex environment. Environ. Pollut. 157, 2903–2927. <https://doi.org/10.1016/j.envpol.2009.05.015>.
- Mantel, N., 1967. The detection of disease clustering and a generalized regression approach. Cancer Res 27 (2), 209–220.
- Mayora, G., Piedrabuena, A., Ferrato, J.J., Gutierrez, M.F., Mesa, L., 2021. Water quality dynamics of floodplain lakes in relation to river flooding and cattle grazing. Mar. Freshw. Res. 72, 1496–1505. <https://doi.org/10.1071/MF20297>.
- McKenzie, V.J., 2007. Human land use and patterns of parasitism in tropical amphibian hosts. Biol. Conserv. 137 (1), 102–116. <https://doi.org/10.1016/j.biocon.2007.01.019>.
- Peduzzi, P., Concato, J., Kemper, E., Holford, T.R., Feinstein, A.R., 1996. A simulation study of the number of events per variable in logistic regression analysis. J. Clin. Epidemiol. 49, 1373–1379. [https://doi.org/10.1016/S0895-4356\(96\)00236-3](https://doi.org/10.1016/S0895-4356(96)00236-3).
- Perrone, S.M., Deutsch, C., Bilenca, D.N., Agostini, M.G., 2022. Artificial aquatic habitats impoverish amphibian diversity in agricultural landscapes of central Argentina. Aquat. Conserv. 32, 591–604. <https://doi.org/10.1002/aqc.3792>.
- Petitot, M., Manceau, N., Geniez, P., Besnard, A., 2014. Optimizing occupancy surveys by maximizing detection probability: Application to amphibian monitoring in the Mediterranean region. Ecol. Evol. 4 (18), 3538–3549. <https://doi.org/10.1002/ece3.1207>.
- Pinheiro, J.C., Bates, D.M., 2000. Mixed-Rf-effects Models in S and S-PLUS. Springer-Verlag NY, Inc. New York., Berlin, Heidelberg.
- Plăiașu, R., Băncilă, R., Samoilă, C., Cogălniceanu, D., 2010. Factors influencing the breeding habitat use by amphibians in the alpine area of the Retezat National Park. Rom. Trav. du Mus. éUm. Natl. D. 'Hist. Nat. 53, 469–478.
- R Core Team., 2023. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing (Version 3.0.0). (<https://www.R-project.org/>).
- Richmond, M.E., 2018. Glyphosate: A review of its global use, environmental impact, and potential health effects on humans and other species. J. Envir. Stu. Sci. 8, 416–434. <https://doi.org/10.1007/s13412-018-0517-2>.
- Riedel, B.L., Russell, K.R., Ford, W.M., O'Neill, K.P., Godwin, H.W., 2008. Habitat relationships of eastern red-backed salamanders (*Plethodon cinereus*) in Appalachian agroforestry and grazing systems. Agr. Ecosyst. Environ. 124, 229–236. <https://doi.org/10.1016/j.agee.2007.10.001>.
- Rodríguez, A., Jacobo, E., 2010. Glyphosate effects on floristic composition a species diversity in the Flooding Pampa grassland (Argentina). Agric. Ecosys. Environ. 138, 222–231. <https://doi.org/10.1016/j.agee.2010.05.003>.
- Sanderson, E.W., Jaiteh, M., Levy, M.A., Redford, K.H., Wannebo, A.V., Woolmer, G., 2002. The human footprint and the last of the wild. BioScience 31, 891–904. [https://doi.org/10.1641/0006-3568\(2002\)052\[0891:THFATL\]2.0.CO;2](https://doi.org/10.1641/0006-3568(2002)052[0891:THFATL]2.0.CO;2).
- Schmutzer, A.C., Gray, M.J., Burton, E.C., Miller, D.L., 2008. Impacts of cattle on amphibian larvae and the aquatic environment. Freshw. Biol. 53, 2613–2625. <https://doi.org/10.1111/j.1365-2427.2008.02072.x>.
- Seimandi, G., Leticia, M., Sánchez, M.L., Saigo, M., Gutiérrez, H., 2021. Effect of rotational grazing management on vegetation of floodplain wetlands. Wetl. Ecol. Manag. 29, 565–580. <https://doi.org/10.1007/s11273-021-09802-y>.
- Soriano, A., 1991. Río de la Plata Grasslands. In: Coupland, R. (Ed.), Natural Grasslands: Introduction and Western Hemisphere. Elsevier., Amsterdam, pp. 367–407.
- Straccia, P.H., Isla, L., 2020. Leyes de presupuestos mínimos de protección ambiental. Sobre glaciares, humedales y la emergencia del carácter político de categorías despolitizadas. Ecol. Austral 30 (1), 085–098. <https://doi.org/10.25260/EA.20.30.1.0.971>.
- Tweel, A.W., Bohlen, P.J., 2008. Influence of soft rush (*Juncus effusus*) on phosphorus flux in grazed seasonal wetlands. Ecol. Eng. 33, 242–251. <https://doi.org/10.1016/j.ecoleng.2008.05.003>.
- Viglizzo, E.F., Frank, F.C., Carreño, L.V., Jobbagy, E.G., Pereyra, H., Clatt, J., Poncén, D., Ricard, M.F., 2011. Ecological and environmental footprint of 50 years of agricultural expansion in Argentina. Glob. Change Biol. 17 (2), 959–973. <https://doi.org/10.1111/j.1365-2486.2010.02293.x>.
- Yin, Y., Winkelman, J.S., Langrehr, H.A., 2000. Long Term Resource Monitoring Procedures: Aquatic Vegetation Monitoring. LTRMP 95-P002-007. US Geological Survey, Upper Midwest Environmental Sciences Center, Wisconsin.
- Zuur, A.F., Ieno, E.N., Walker, N.J., Saveliev, A.A., Smith, G.M., 2009. Mixed Effects Models and Extensions in Ecology with R. Springer., New York.