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Emerging conflicts for the environmental use of water in high-valuable rangelands. Can livestock water ponds be managed as artificial wetlands for amphibians?

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ABSTRACT

Continental freshwater, irrespective of its origin, natural or artificial, may contribute significantly to biodiversity conservation. Because of the decline of natural aquatic habitats, an increasing concern exists about the role of water ponds as spots of biological richness. Amphibians are strongly at risk since the loss of aquatic habitats, among other factors, causes the isolation of their populations. The implementation of livestock ponds as artificial wetlands may be an effective measure for enhancing amphibian decaying communities. This policy assumes that managing ponds for wildlife conservation purposes joins livestock welfare requirements, but this hypothesis has not been specifically studied. The purpose of this research is to evaluate this premise in the Urbasa-Andia Natural Park, a high-valuable environmental area that holds a relevant amphibian community and has an extended grazing history. We analyse the relationship between the amphibian assemblages present and the design and attributes of a variety of drinking points previously chosen by embodying a high environmental heterogeneity of water resources. The results of this study indicate that the quality of the water stored varies largely along the season, degrading severely in summer because of the wading of animals (in unfenced ponds) and the low water recharge. The contamination, caused by increased enteric microorganisms and dissolved N, is likely to affect livestock more severely than amphibian populations, since the sensitive breeding stage of many amphibians occurs before the loss of water quality. Although the quality of the water is essential, and mammals (wild and domestic) have an influence on it, other factors that are less considered by environmental managers emerge as main drivers of amphibian assemblages, such as hydroperiod, predator occurrence and the environmental quality of the surrounding habitat.

1. Introduction

Wetlands are being created and restored with great frequency around the world because they represent hot spots of biological richness (Mistch et al., 2005). To compensate for the loss of natural wetland habitats, a persuasive trend towards building and adapting water ponds as artificial wetlands is occurring in many countries (Vymazal, 2010). In regions largely transformed by human activities or where natural wetlands are scarce because of climate (such as Mediterranean regions) or soil (such as karstic areas, Cirovic et al., 2008), artificial ponds created for agricultural or stockbreeding purposes represent valuable, strategic breeding habitats for wildlife, such as migratory birds (Schaffer et al., 2006; Shuford et al., 1998), wild mammals (Dalbeck and Weinberg, 2009) and amphibians (Knutson et al., 2004).

In the last decades, amphibian populations have endured a sharp decline around the world (Houlahan et al., 2000). The Iberian Peninsula has not escaped this decay (Barbadillo et al., 1999; Beja and Alcazar, 2003). Because of this, a growing body of publications advocates livestock ponds for amphibian assemblages (Brainwood and Burgin, 2009; Rannap et al., 2009). As we know, most literature has focused on the adequate attributes of artificial ponds for ensuring amphibian growth and survival, but it has neglected the dual role of these reservoirs and the potential trade-offs that may occur when managing water for amphibians' wildlife and for stockbreeding needs (Oerti et al., 2009). The starting point of livestock welfare is the recognition that domestic animals under human care are sentient beings and should be treated accordingly. EU policy has legislated during the last decade in order to warrant the full physic and mental health of domestic animals. However, several gaps have

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still to be covered, such as access to a source of good-quality water, the most important nutrient for animals (Habich and Kamphues, 2009). Contrary to drinking quality for humans, strictly regulated in the majority of countries, water quality for livestock lacks specific legislation and control in most cases. In intensive stockbreeding systems, water is provided by the general distribution network and meets drinking requirements. Conversely, in extensive systems, water derives from natural, less-monitored sources and remains stagnant in reservoirs during long periods of time. Despite its importance, water supply to livestock is a worrying consideration in most extensive rangeland systems around the world.

Owing to these reasons, environmental managers need information about how water reservoirs have to be designed and how they can be managed to maximise their utility. In the field, many questions arise that need a response: Can livestock ponds be managed for dual purposes? Does the surveillance of wildlife prescriptions affect negatively the quality of drinking water to livestock? Is it possible to design ponds where amphibians and livestock coexist conveniently or should we promote the creation of specific ponds for wildlife? All these questions arise in the Urbasa-Andia Natural Park, two mountain ranges of great environmental value and grazing use located south of the Western Pyrenees (950 m a.s.l.). The karstic landscape of these ranges, originated by the calcareous nature of the bedrock and the rainy climate, has promoted the existence of numerous aquifers, while surface streams are mostly absent. As a consequence, from historical times, the supply of water to livestock has been handled by artificial reservoirs, very abundant in the area (up to 40). In 1997, the declaration of the area as Natural Park involved a management more devoted to conservation objectives and led to sensible changes on sectorial policies, as the one related to water. For environmental managers, livestock ponds acquired a singular ecological relevance due to the presence of the alpine newt (*Triturus alpestris*). This amphibian, well distributed over Central and Eastern Europe, is scarcely represented in the Iberian Peninsula, where it is a threatened species (Recuero and Martínez, 2002; Diego-Rasilla, 2009). From then on, Park managers have focused conservation efforts on enhancing the reproductive success and survival of this emblematic amphibian, and advocated the construction of *naturalised* ponds, namely, livestock ponds created and managed to satisfy amphibian habitat requirements.

In this manuscript we address the emerging conflicts and trade-offs that occur when managing water for wildlife and for livestock welfare. This research particularly addresses how different attributes of ponds affect the quality of the water and what are the potential effects on animal populations depending on that water. To achieve this goal, we (1) survey a set of ponds that track the constructive trends occurred over the last century in the Park (1905–2005), (2) assess the chemical and microbiological quality of their waters in two key periods of the year and, (3) evaluate the role of other abiotic and biotic attributes of the ponds on amphibian assemblages. Results of this study are then used to ascertain the main existing conflicts and to set up which are the central attributes of ponds that environmental managers have to take into account in order to allow the dual use of livestock ponds as artificial wetlands for amphibians.

2. Methods

2.1. The study site

The Urbasa-Andia Natural Park, located south of the Western Pyrennees (42°48′–42°52′N and 1°22′–1°32′E) (Fig. 1); is constituted by two karstic mountain ranges with an averaged altitude of 950 m a.s.l. From a climatic point of view, the area is transitional since it receives Atlantic and Mediterranean influences. The mean annual temperature is 8.4 °C and the 1275 mm of rainfall received annually is irregularly distributed, being summer the driest period. The Park is covered by 16,100 ha of grasslands, heathlands and beech forests, most of them enclosed in the Natura 2000 network, and supports a high faunistic biodiversity, up to 145 species of vertebrates (94 of birds, 34 of mammals and 17 of amphibians and reptiles), several of them within status of protection. From a socioeconomic viewpoint, the area is a valuable, traditional pastureland that has been grazed by extensive livestock since prehistory. Nowadays, the Park holds a census of 11,700 Animal Units, which belong to 310 farmers (34,700 sheep, 4200 cattle and 2600 horses), that graze in during 6–7 months/year (Canals and Sebastià, 2000).

2.2. Selection and description of ponds

Because of the karstic landscape, natural surface streams are extremely scarce in the area and water for livestock is supplied from historical times by artificial ponds that, in recent years, have acquired a singular environmental relevance. During 2006 and 2007, a complete inventory of livestock facilities was done in the Park as part of a Grazing Management Planning document (Canals et al., 2008). As a result, 40 livestock ponds were described and catalogued. Most ponds offered direct access to livestock (25), and the remaining were fenced and connected to a trough (15). Seven unsealed ponds offered a temporary water supply and only 2 ponds were sealed with a waterproof artificial lining in the bottom. Pond sizes and depths were variable, from 300 to 18,000 m^2, and from 30 cm to more than 2 m deep.

For this study, we carefully selected seven ponds which traced the constructive changes occurred over the last century, from very simple (small, unfenced, unsealed) to sophisticated reservoirs (deep, fenced, sealed). Table 1 synthesizes the main constructive and environmental attributes of these ponds. Two of them (*Raso Urbasa* and *Bardoitza*) were old, unfenced ponds, created by excavation and unsealed. These reservoirs, filled by rain and rainoff, retained shallow waters and experienced periodic fluctuations of the water table, drying out during the hottest summers. Three selected ponds (*Bekosare*, *Majadas de Alsasua* and *Olaberri*), were also unsealed, but were fenced, connected to a trough and deeper than the previous ones. In these three ponds, an extended strip of vegetation separated water edges from fences. Finally, we included in the study the unique two sealed ponds, provided with an impermeable artificial bottom, *Mármol* and *Ilusiar*. *Mármol* had a plastic base of black polyethylene with steep edges, provided by log palisades in corners to allow amphibians traffic. *Ilusiar*, the last reservoir constructed according to the new environmental criteria (*naturalised* pond), was sealed with a waterproof lining of bentonite replenished with a layer of 30 cm of excavated soil, had flat shore edges and an adjacent, wide strip of vegetation left to provide a post-breeding habitat to amphibians and an ample space for big birds (i.e. vultures) take-offs and landings.

In addition, we collected information on the biotic attributes, floristic and faunistic, of the ponds. We described fisionomically the floristic communities surrounding the ponds and reported the occurrence and cover of aquatic vegetation and terrestrial vegetation in the pond edges. Regarding fauna, we gathered information on the occurrence of potential amphibian predators and competitors inhabiting the ponds, such as fishes and crayfishes.

2.3. Amphibian richness studies

Amphibians have been the focus of several studies within the Urbasa-Andia Natural Park (Alcalde and Patiño, 1989; Gosá et al., 2004), since the area fulfils many environmental conditions for

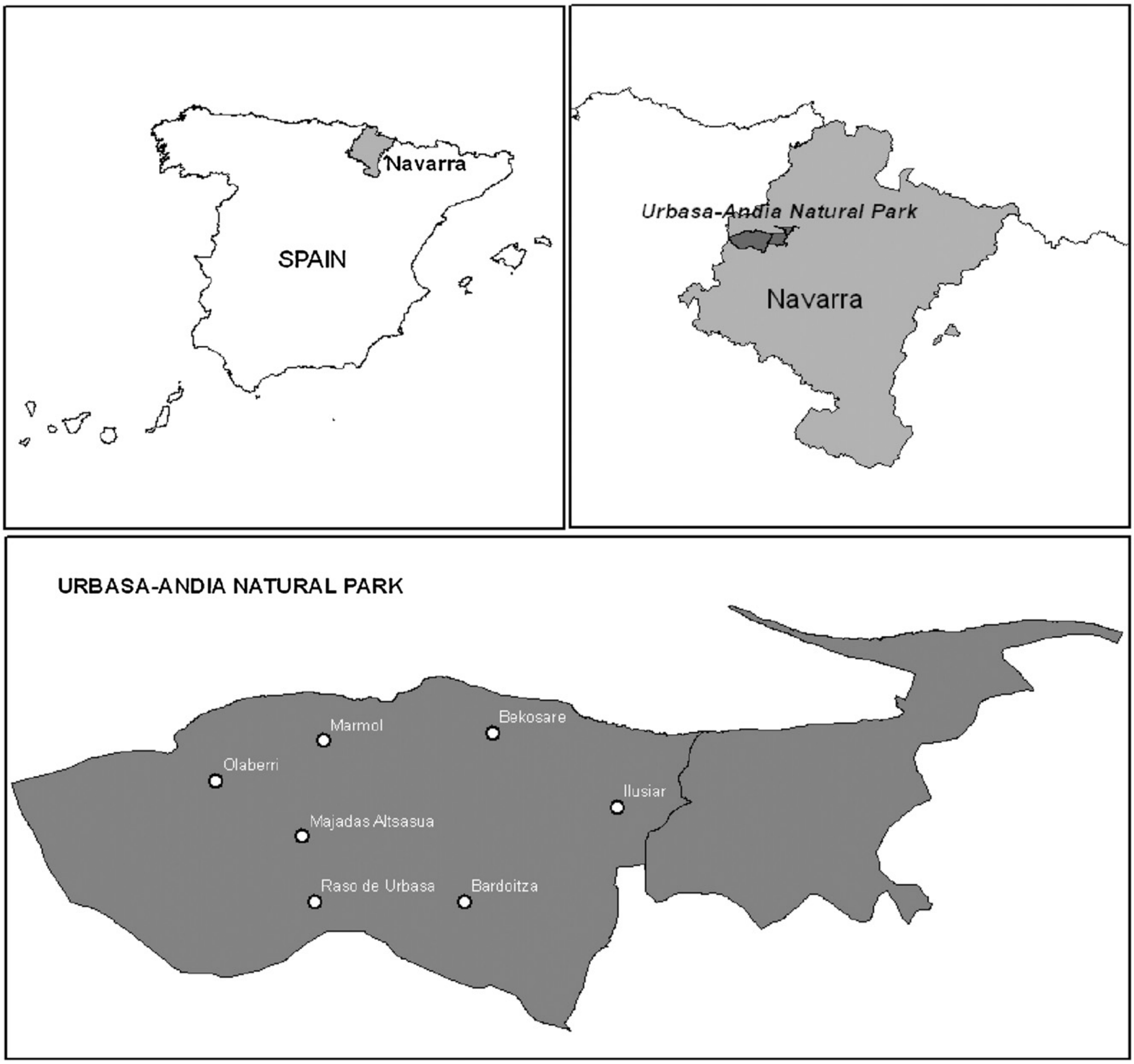

Fig. 1. Location of the sampled ponds at the Urbasa-Andia Natural Park (N Spain).

Table 1
Construction features and environmental characteristics of the sampled ponds.

Pond	R. Urbasa	Bardoitza	Bekosare	Olaberri	M. Alsasua	Mármol	Ilusiar
Construction year	<1950	<1950	<1950	<1950	<1950	1998	2005
Pond fencing	No	No	B wire	B wire	St wall	G-B wire	G-B wire
Pond sealing	No	No	No	No	No	Plastic	Bentonite
Water collection	Rain	Rain/run	Rain/Spr	Rain/run/Spr	Aquif/rain	Rain/run	Spr/rain
Pond surface (m^2)	875	825	1950	260	1200	540	1000
Average depth (cm)	30	60	>130	>130	>130	>200	>200
Water body permanency	No	Yes	Yes	Yes	Yes	Yes	Yes
Number of troughs/pond	0	0	1	1	2	1	1
Floating vegetation (%)	0	0	<5	0	<25	0	0
Submerged vegetation (%)	<25	25–75	<25	>75	>75	>75	25–75
Pond edge shrubs (%)	<25	<25	<25	>75	>75	<25	<25
Pond edge herbs (%)	>75	>75	>75	<25	<25	>75	>75
Surrounding community	Grassland	Grassland	Clearing	Beech	Grassland	Beech	Shrubland

G-B, grid-barbed; B, barbed; St, stones; Spr, spring; run, runoff; Aquif, aquifer.

Table 2
Populations of amphibians, fishes and crayfishes in ponds. Light grey on the left is open and unsealed ponds (Raso Urbasa, Bardoitza), white colour in the middle is unsealed but fenced ponds (Bekosare, Olaberri, Majadas Alsasua), and dark grey on the right is sealed and fenced ponds.

Animals in ponds	Raso Urbasa	Bardoitza	Bekosare	Olaberri	Majadas Alsasua	Mármol	Ilusiar	N. ponds present
Fish occurrence								
Tinca tinca	0	0	0	1	0	0	0	1
Crayfish occurrence								
Austropotamobius pallipes	0	0	1	0	0	0	0	1
Amphibians occurrence								
Rana temporaria	1	1	0	1	0	1	0	4
Rana perezi	0	1	0	1	1	0	1	4
Salamandra salamandra	0	0	0	0	0	1	0	1
Hyla arborea	0	1	0	0	1	0	1	3
Triturus helveticus	0	1	0	1	1	1	1	5
Triturus marmoratus	0	0	0	0	1	1	1	3
Triturus alpestris	0	1	0	1	1	1	1	5
Alytes obstetricans	0	1	1	1	1	1	1	6
Amphibian species richness	1	6	1	5	6	6	6	

them. We compiled the existing data on the occurrence of amphibians of the extensive study of Gosá et al. (2004), with new samplings in previously selected ponds (Cárcamo, 2008) during the growing period. In these survey ponds, connected troughs (when existing) and potential surrounding shelters (fallen leaves, mud, stones,. . .) were carefully examined. Within ponds, samplings were done with a manual fishing net, crossing the reservoir on foot in different directions. Catches including adults, juveniles, larvae and eggs were stored, identified by species, and returned to the ponds.

2.4. *Water sampling and quality analyses*

First water samplings in ponds were done between April and early May 2009, when amphibian populations initiated the breeding stage and livestock had not accessed the rangeland yet. Samples were collected as close to the middle of the pond as possible, by means of long ropes. The samplings were repeated two months later, between late July and early August, at the middle of the grazing period. Of the two unfenced ponds in the study, one dried up in summer, and could not be resampled. A total of 23 water samples were collected in sterilised high-density polypropylene bottles of 1500 mL capacity. Filled bottles were labelled, immediately refrigerated in ice coolers and transported to the laboratory.

Water samples were analysed within 24 h of collection. Laboratory analyses included measurements of physical, chemical and microbiological parameters. In total, 13 water quality variables were determined: colour and odour, particulate matter (turbidity and conductivity), pH, reactive forms of inorganic nitrogen in water (nitrate, nitrite and ammonium), organic matter (oxidability), fecal indicator bacteria (total coliforms and enterococci) and two specific enteric pathogens (*Clostridium perfringens* and *Escherichia coli*). Odour and colour were determined following ISO 5492/1992 and ISO 7887/1995 respectively. Turbidity was measured quantitatively with a turbidimeter Dinko, according to ISO 7077/2005. Oxidability was determined by the permanganate method (ISO 8467/1993). Nitrates and nitrites in water were measured by spectrophotometry and ammonium by means of a flow injection analyser (FIAStar 5000). Microbial detection and enumeration were based in the miniaturised method (most-probable-number, MPN). Coliforms were enumerated by the Colilert-18/Quanti-Tray method (IDEXX Laboratories Inc), which has proved to be more rapid and sensitive than other standardised methods (Eckner, 1998). *Enterococci* and *E. coli* were measured by inoculation in a liquid medium: BEA agar for enterococci (ISO 7899/1) and MLGA and Mc Conkey agars for *E. coli* (ISO 9308-3/AC). Eventually *C. perfringens* was determined by membrane filtration using TSC agar.

2.5. *Statistics*

The use of numerical or statistical methods to analyse data was constrained by the low number of sampled ponds (seven), the high number of variables studied within each pond (more than 26), the different nature of the data collected (quantitative, qualitative, categorical,. . .) and the absence of replicates in several cases. In spite of this, we did multivariate ANOVAs on quantitative variables when possible (i.e. to compare sealed and unsealed ponds) and performed correlation analyses to ascertain potential relationships between water quality parameters and amphibian richness. Previous to the use of parametric statistics, data were checked for normality and for homogeneity of covariances, and transformed when necessary. Eventually, we completed the study with a descriptive analysis of the amphibian assemblages present in each pond and the individual attributes of the pond.

3. Results

3.1. *Amphibian richness*

Despite the karsticity and the scarceness of natural reservoirs, eleven species of amphibians have been reported in the Park, which comprise 80% of the species present in the Navarra County. This fact is explained by the high number of artificial ponds and the environmental quality and variety of surrounding habitats. In this study focused in seven ponds, we reported eight of the eleven species present in the Park. All taxa were distinctive of temperate European environments. *Alytes obstetricans* was the most frequent species, appearing in 6 ponds, followed by two newts *Triturus helveticus* and *T. alpestris*, present in 5 ponds. *Salamandra salamandra* was the less reported taxa, only present in one pond. Three species encountered were included in a status of protection: *T. alpestris* and *S. salamandra* as “Vulnerable” and *A. obstetricans* as “Near threatened” (Pleguezuelos et al., 2002).

Table 2 reports the amphibian assemblages in ponds and the occurrence of fishes and crayfishes. Four ponds were inhabited by six amphibian populations: the most environmentally concerned pond (Ilusiar), the plastic-isolated Mármol, the unsealed Majadas Alsasua and the open and unsealed pond Bardoitza. In two ponds only one amphibian species was reported, Raso Urbasa (used by *Rana temporaria*), and Bekosare (used by *A. obstetricans*). The former pond dried during summer months and the latter was used as a bedding reservoir for the native crayfish *Austropotamobius pallipes*. In addition, in these two ponds the development of submerged vegetation was low (<25%), compared to the rest of reservoirs.

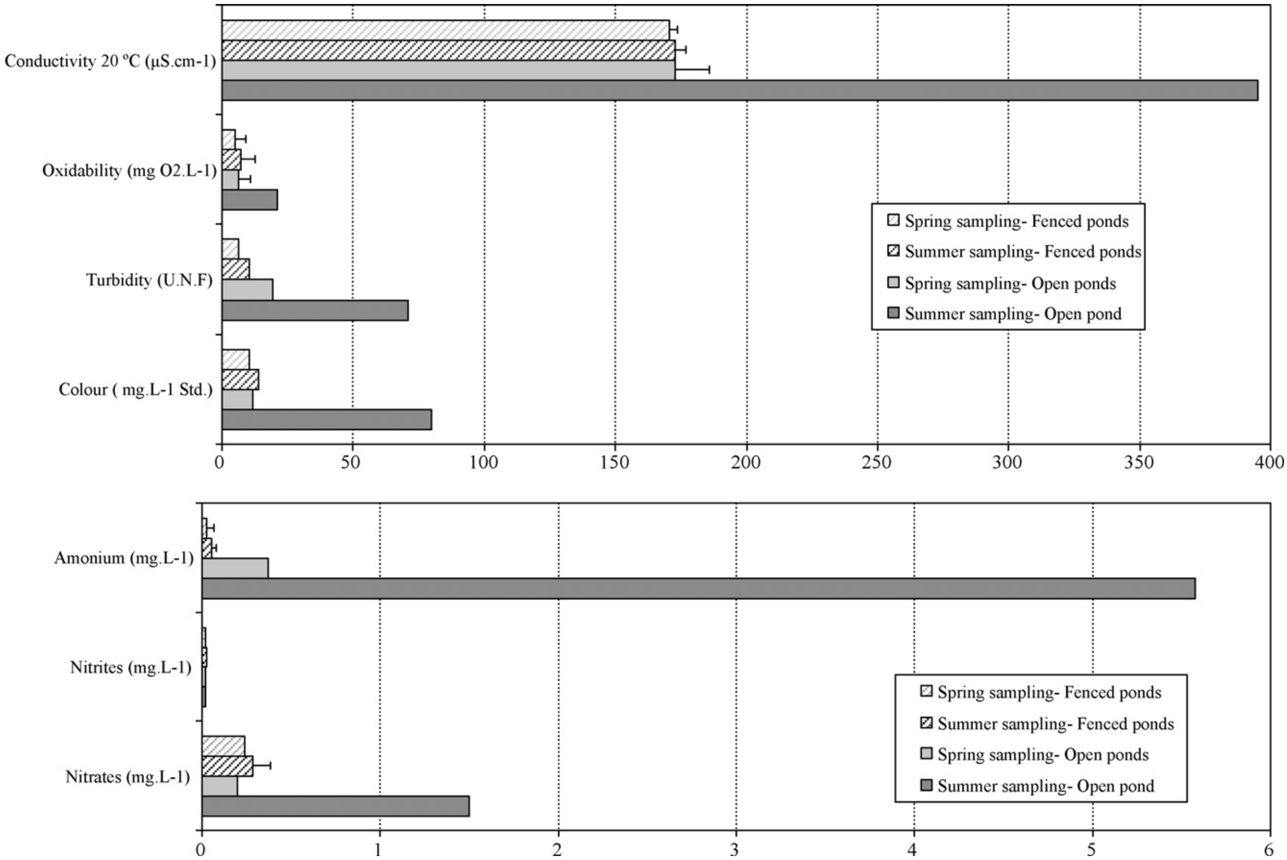

Fig. 2. Chemical quality of the water in fenced and open ponds in spring and summer samplings. Mean values are indicated within the bars. Error bars are standard errors.

Regarding correlations between amphibian richness and quantitative abiotic parameters, we found a tendency to negative relationships between amphibian richness, water turbidity and ammonium concentrations during the spring period ($R^2_{\text{turbidity-amphibians}} = -0.905$; $p < 0.005$; $R^2_{\text{ammonium-amphibians}} = -0.638$; $p < 0.120$), that disappeared in summer samplings ($R^2_{\text{turbidity-amphibians}} = 0.082$; $p < 0.878$; $R^2_{\text{ammonium-amphibians}} = 0.252$; $p < 0.630$). These data suggested that amphibian assemblages in ponds were particularly influenced, and constrained, by the quality of the water in spring, even though, as we will see, water degradation was particularly intense in summer.

3.2. Chemical water quality

Chemical water quality was largely affected by fencing. All ponds in the study had an acceptable chemical quality of water except Bardoitza during the second sampling. This pond, unfenced to livestock, displayed during summer the highest values of turbidity (71 UNF), conductivity (395 µS/cm), oxidability (20.9 mg O_2/L) and colour (80 mg Pt/Co) (Fig. 2). Values came close or exceeded most upper limits recommended for drinking water and for environmental uses. Turbidity was much higher than 40 UNF, conductivity was close to the limit of 500 µS/cm and oxidability farther exceed 5 mg O_2/L. Regarding the occurrence of a pond liner, we did not report significant differences between sealed (plastic/bentonite) and unsealed ponds for any quality variable studied (Table 3).

Nitrogen (N) concentration in water is determined by the geology of the watershed and by potential contaminations. Since inorganic-N forms highly dissolve in water, N may enter the ponds by surface runoff, soil filtering and by direct contributions. In unpolluted freshwaters, ammonium and nitrate are typically at low contents (<1 mg/L), and concentrations of nitrate are usually higher than those of ammonium, since the latter tends to be oxidised to nitrate (even at low levels of dissolved oxygen). We did find this expected pattern in all samplings except for the open pond in the summer time, where ammonium (5.6 mg NH_4^+/L) largely exceeded nitrate (and nitrite) contents (1.5 mg NO_3^-/L). These results indicated that most contamination was caused by urine, and to a lesser extent by feces, of animals accessing the pond. N content in waters correlated significantly with colour ($R^2_{\text{ammonium-colour}} = 0.864$; $p < 0.001$, $R^2_{\text{nitrate-colour}} = 0.761$; $p < 0.01$), turbidity ($R^2_{\text{ammonium-turbidity}} = 0.692$; $p < 0.05$; $R^2_{\text{nitrate-turbidity}} = 0.670$; $p < 0.05$), and conductivity ($R^2_{\text{ammonium-conductivity}} = 0.733$; $p < 0.01$, $R^2_{\text{nitrate-conductivity}} = 0.616$; $p < 0.05$). In addition, we observed in the unfenced pond a sharp pH decline (from 8.4 to 7.6) between sampling periods, which suggested that part of the ammonia had volatilised to ammonium.

Ammonia is the most toxic form of N in water, and its toxicity increases with pH and temperature. We applied the US EPA[1] 2009 algorithms to determine the upper limit contents accepted for this toxic form. Considering a pH of 7.6 and a temperature of 20 °C in the open pond during summer time, thresholds calculated were 8.3 mg NH_4^+/L (acute criterion in salmonid absent waters) and 3.4 mg NH_4^+/L (chronic criterion when early-fish life stages present). According to this, ammonium concentrations exceed the chronic criterion during summer time in the open pond, but were below the acute criterion. With respect to nitrate, all data were

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[1] United States Environmental Protection Agency.

Table 3
Chemical and microbiological quality of the water in ponds in the spring and summer samplings.

Pond	R. Urbasa	Bardoitza		Bekosare		Olaberri		Majadas		Mármol		Ilusiar	
Sampling period	Spring	Spring	Summer	Spring	Summer	Spring	Summer	Spring	Summer	Spring	Summer	Spring	Summer
Chemical variables													
pH	8.1	8.4	7.7	7.7	8.2	7.9	7.5	7.4	7.5	7.2	7.9	7.9	8.2
Colour (mg/L in Pi/Co)	7	16	80	6	<5	13	28	24	25	6	8	<5	7
Turbidity (UNF)	32.1	6.1	71	18.8	18.7	2.9	8	1	1.2	7.1	4.3	2.3	19.4
Conductivity (μS/cm)	216	130	395	177	198	194	201	206	252	40	48	236	166
Oxidability (mg O_2/L)	6.3	6.2	20.9	3.5	3.5	5.6	9.9	9.8	8.2	4.8	5.5	1.9	9.9
Nitrate (mg/L)	<0.4	<0.4	1.5	<0.4	<0.4	<0.4	0.6	0.4	<0.4	<0.4	<0.4	<0.4	0.4
Ammonium (mg/L)	0.71	<0.05	5.58	<0.05	<0.05	<0.05	<0.05	<0.05	0.16	<0.05	<0.05	<0.05	<0.05
Microbial variables													
Total coliforms (ufc/100 mL)	26	5	24,200	816	310	47	3260	219	24,200	260	750	1553	24,200
Enterococci (ufc/100 mL)	<10	<10	179	<10	<10	<10	15	<1	144	<1	15	<10	15
E. coli (ufc/100 mL)	<10	<10	6581	<10	<10	<10	15	<1	2182	<1	<10	<10	179
C. perfringens (ufc/100 mL)	30	<1	30	<1	10	<1	10	<1	<1	50	<1	<1	<1

much below the guidelines for safe use (<10 mg NO_3^-/L; US EPA, 2009), and recommendations for most sensitive freshwater species (8.9 mg NO_3^-/L; Camargo et al., 2005).

3.3. *Microbial water quality*

Similar to chemical quality, water increased its microbiological charge during the summer period (Table 3). Degradation occurred in most ponds, but was particularly severe in the unfenced Bardoitza where total coliforms boosted up to 20,000 ufc/100 mL (Fig. 3). *Enterococci* and *E. coli* are known to provide a better relationship with disease risk than total coliforms (Jin et al., 2004). *Enterococci* encompass a genus of lactic acid, facultative anaerobic bacteria, easier to detect than *E. coli* (Picone et al., 2003; Higgins et al., 2008). We recorded *E. coli* in all ponds, and found a tight relationship between *enterococci* and *E. coli* counts ($R^2 = 0.708$; $p < 0.01$ in spring and $R^2 = 0.914$; $p < 0.0001$ in summer). *Enterococci* increased in all ponds (except Bekosare) during summer, and achieved more than 140 ufc/100 mL in the open pond. Accordingly, the facultative anaerobic *E. coli* highly proliferated in summer in the unfenced pond (6581 ufc/100 mL) and, to a lesser extent, in the rest of ponds (i.e. 2182 ufc/100 mL Majadas Alsasua, 179 ufc/100 mL Ilusiar, Table 3), except Bekosare. In all cases, values exceed for the long reference thresholds established for drinking waters and indicated that water stored in ponds involved a serious microbiological health hazard for mammals.

With regard to the anaerobic *C. perfringens*, it exhibited a different pattern than the rest of microbial indicators. The highest concentrations of *C. perfringens* (50 ufc/100 mL.) occurred in Mármol pond during the spring sampling (Table 3). Contrary to *Enterococci* and *E. coli*, which have a short survival in water, *C. perfringens* spores are extremely persistent and resistant to environmental stresses (Medema et al., 1997; Horman et al., 2004; Savichtcheva and Okabe, 2006), and may be indicators of past fecal inputs (Sorensen et al., 1989; Lisle et al., 2004). Mármol pond, located within a beech forest, accumulated a substantial layer of litter at the end of the winter on its plastic bottom and often stank. Data suggest anoxic conditions favouring the survival and growth of *C. perfringens* spores, coming from present or from remote fecal pollution.

4. Discussion

4.1. *Open ponds, wading of livestock and water contamination*

Direct access of livestock to unfenced ponds caused an intense degradation of water chemical quality and boosted the contents of enteric microorganisms. Although previous works indicate a loss of water quality by the wading of animals in open ponds (Wolinski and Bourassa, 2005; Chandler et al., 2008), we are not aware of studies that have attempted to quantify it. In the spring sampling, before livestock arrival to rangeland, water quality in unfenced ponds was similar to that of fenced ones. Two months later the picture changed completely, and the water degraded rapidly (Figs. 2 and 3). Excrements of livestock concentrated in water, manure and urine, increased the microbial charge and responded for the large nitrogen dissolved in water, mostly ammonium that in turn, correlated with parameters such as conductivity, oxidability, turbidity and colour. The scarce water recharge (Fig. 4) and the high temperatures during summer months, typical from Mediterranean-influenced climate, intensify the problem: the intense evaporation leads to a severe reduction of the water level in ponds whose recharge depends on rain and rainoff – even till dry off in some cases – and the lack of water refill concentrates contamination and creates a muddy floor that enhances the entry of sediments by animals' trampling.

In open ponds during summer months, livestock access the reservoir to get wet and cool down, but avoid drinking and decrease water intake if degraded (*pers. obs*). Consequently, livestock welfare may be largely affected by its own wading in ponds. The consumption of low-quality water entails severe risks: (1) increases animal health problems and pathogen waterborne diseases, (2) affects negatively animal fitness and production, and (3) increases health hazards for humans when consuming animal-derived raw products. Regarding microbiological charge, the risk of enteric contamination was present in all samplings in all ponds. The occurrence of bacteria such as *E. coli* and *C. perfringens* in fenced reservoirs revealed the existence of other sources of fecal contamination apart from livestock. Effectively, some wild, warm-blooded animals have been seen to access fenced ponds. Migratory birds use their muddy shorelines for resting and feeding of invertebrates in spring and autumn, and prey birds, particularly vultures, which are very numerous in the area, refresh in ponds during summer. Also small sized mammals (such as squirrels, mice, voles and moles) may frequent ponds. Even though fencing did not prevent water microbial contamination, it fairly reduced it: *E. coli*, which was detected in all ponds, boosted in the open pond (6581 ufc/100 mL) because of the wading of livestock. *E. coli* is the primary facultative organism of the intestine of warm-blooded animals and its detection in drinking waters entails a risk *per se* (some strains are pathogenic or produce lethal toxins, i.e., O157:H7; Bach et al., 2002), and warns for the potential occurrence of other enteric pathogens (such as *Campylobacter jejuni*, *Salmonella* sp. and *Listeria monocytogenes*). Despite pathogenic charge may be carried asymptomatically by adult stock,

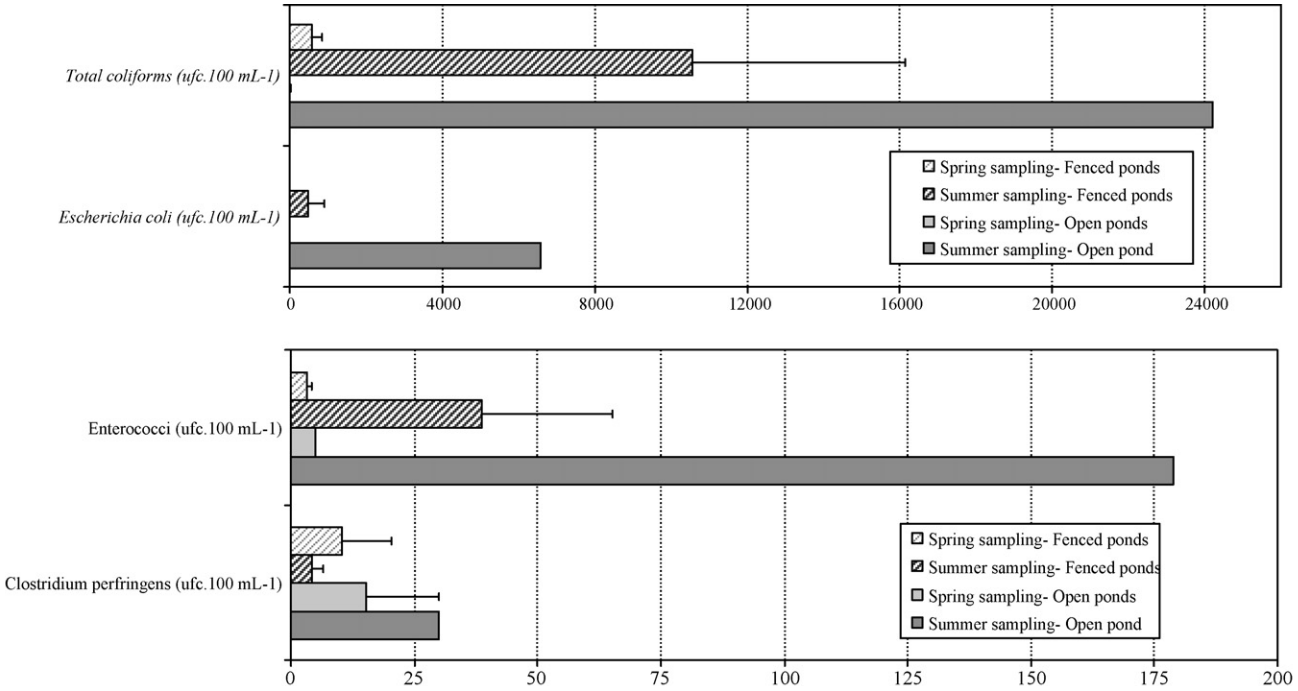

Fig. 3. Microbiological charge of the water in fenced and open ponds in spring and summer samplings. Mean values are indicated within the bars. Error bars are standard errors.

it largely affects young or immunosuppressed animals and may be transmitted to animal products consumed by humans (such as raw ground beef and milk, Kammerer and Ganiere, 1998; Vogt and Dippold, 2005).

Regarding N contamination, moderate levels of nitrate may cause health problems in livestock such as poor growth, infertility, abortions and vitamin A deficiencies. In the ponds, nitrate in water was below the guidelines for safe consumption but the high dissolved ammonia caused a disgusting smell and odour that influenced the voluntary intake of water (*pers.obs*). An animal deficient hydration depresses the ingestion of solids and leads to a reduction of body condition (Hyder et al., 1968; Willms et al., 2002; Lardner et al., 2005). Of all the factors regulating feed intake, a negative water balance triggered by abiotic factors (such as water quality, long distances to water, water temperatures,. . .) is more prone to occur in extensive grazing systems (Schlecht et al., 1999). In spite of this, farmers are hardly aware on the role of the water regulating feed intake and animal's weight losses in extensive farming are seldom related to water deficiencies.

With respect to amphibians, it is well documented its sensitivity to high concentrations of N compounds (Blaustein et al., 1994; Knutson et al., 2004). Ammonia has strong negative effects on larval growth, development and survival, even at low concentrations (Rouse et al., 1999), and nitrate may act as an endocrine disruptor of the sexual selection process (Secondi et al., 2009) and affect larval development and survival (Camargo et al., 2005; Oromí et al., 2009). In our study, we failed to detect a significant relationship between amphibians and N in summer samplings. In fact, Bardoitza unfenced pond, displaying the highest N in summer, supported an amphibian assemblage similar to that of fenced ponds. In addition,

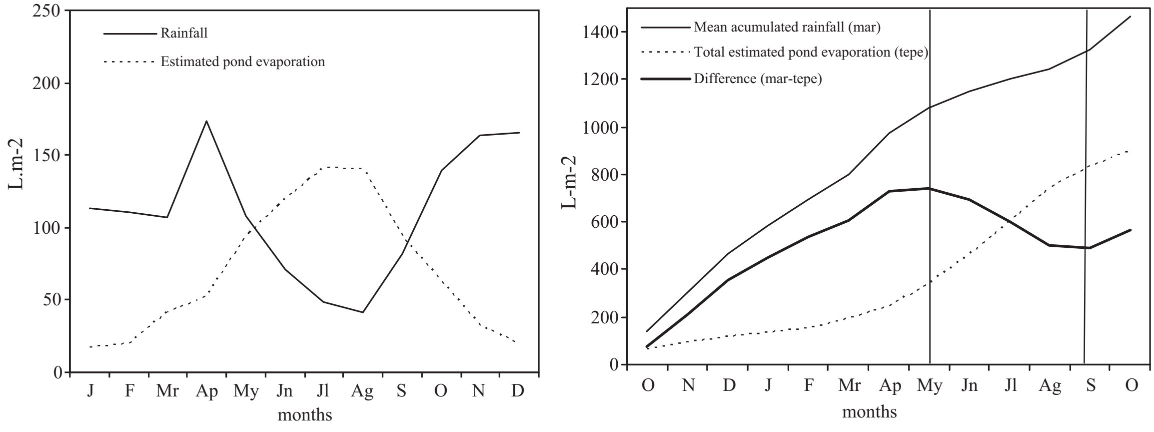

Fig. 4. Mean water imports (by rain and snow) and estimated exports in ponds, based on climatic data from the Urbasa meteorological station for the period 1987–2004. Pond evaporation has been estimated from ETo (Reference Crop Evapotranspiration) calculated by Thornthwaite, considering a Kp (Pan coefficient) of 0.75 (which depends on moisture and winds). In the second graph accumulated values are displayed from October (first month with precipitation > evaporation) and vertical lines encompass the period of the year with the lowest water recharge.

Table 4
Livestock grazing period (in dark grey) and breeding (reproduction and larval development) aquatic periods of amphibians present in the sampled ponds (in light grey).

		J	F	Mr	A	M	J	Jl	Ag	S	O	N	D
Grazing period						■	■	■	■	■	■	◧	
Rana temporaria	R		■	■	■								
	D		■	■	■	■	■						
Rana perezi	R				■	■	■	■					
	D						■	■	■	■	■		
Salamandra salamandra	R				■	■							
	D				◨	■	■						
Hyla arborea	R				■	■	■						
	D					◨	■	■	■				
Triturus helveticus	R		■	■	■	■	■	■					
	D					◨	■	■	◧				
Triturus marmoratus	R		■	■	■	■	■	■					
	D						■	■	■				
Triturus alpestris	R				■	■	■	■	■				
	D						◨	■	■				
Alytes obstetricans	R				■	■	■	■	■				
	D	■	■	■	■	■	■	■	■	■	■	■	■

this pond held the amphibian species *Hyla arborea* described in the literature as very sensitive to ammonium (Ortiz et al., 2004). The lack of clear effects of high dissolved N on amphibian assemblages led us to consider the following hypotheses: (1) amphibian aquatic development occurred before the loss of water quality, (2) concentrations of N compounds were below the sensitivity threshold of the amphibian species present in ponds, and (3) other factors at least as important as water contamination affected amphibian assemblages. Regarding the first point, the most sensitive stages of the amphibian biphasic life cycle, the reproductive and larval phases, occur in late winter and spring, when water quality parameters are good, and most amphibians, except hibernating species, leave the ponds before water degradation occurs (Tables 3 and 4). Concerning the second point, in all studies consulted N concentrations tested were far above the values reported in Urbasa ponds, and the negative effects of N largely varied among species (Marco et al., 1999), and even among populations (Johansson et al., 2001). Regarding ammonium, Ortiz et al. (2004) and Ortiz-Santaliestra et al. (2006) found significant negative affections in amphibians with values of 10 mg NH_4^+/L or higher. In our most degraded pond, concentrations were 5.7 mg NH_4^+/L. With respect to nitrate, Oromí et al. (2009) and Secondi et al. (2009) observed negative effects on amphibians when values reached 50 mg NO_3^-/L. Our highest concentration was 1.5 mg NO_3^-/L. Following EPA's chronic criteria, threshold values ensuring the long-term survival of water-life, sensitive animals are 3.4 mg NH_4^+/L and 10 mg NO_3^-/L. Summing up, nitrate concentrations in ponds were not by far alarming, but ammonium concentrations might reach relevant peaks because of urines of animals accessing ponds. While most nitrogen in livestock wastewaters is in the form of ammonium (Knight et al., 2000), high nitrate concentrations mostly occur in water reservoirs receiving run-off and lixiviates from intensively fertilized agricultural lands (where levels can exceed 25 mg/L, Steinheimer et al., 1998; Camargo et al., 2005).

4.2. Pond attributes not related to water quality

In addition to water quality, we hypothesised that other pond attributes may influence amphibian survival. In the area of study, two ponds hold the lowest number of amphibian populations, Raso Urbasa and Bekosare. The former had a temporary water body and the latter supported a native crayfish population.

Hydroperiod determines the time interval during which a pond is available for colonisation and breeding, and thus ascertains the set of species that may colonise it (Díaz-Paniagua, 1990; Pleguezuelos et al., 2002; Beja and Alcazar, 2003). Water temporality emerged as a key factor of amphibian richness in the Park. In the temporary pond, Raso Urbasa, only one amphibian species was present, *R. temporaria*. In this species reproduction and larval development occurs early in the season, and finishes by the end of June, before most of the reported amphibians do. These data suggest that this early-season breeder may have a competitive advantage on the rest of species on ephemeral ponds, through escaping the drying out during the breeding stage. Even in permanent water bodies, in areas with a manifested warming and in the absence of late spring frosts that affect initial breeding, early-season amphibians may display advantages over late-season breeders. In the Park, the water level of many ponds lowers during summer months. This decrease affects water quality, as previously reported, and has other important consequences. In Mármol pond, the edges of black plastic-sealing attained extreme temperatures in summer (> 40 °C), that heated water over 35 °C (Cárcamo, 2008). It is possible that sealing overheating influences negatively the growth of late season breeders, such as *Rana perezi* (Table 4), a ubiquitous species in the Park that is absent from the plastic-sealing pond (Table 2). External temperatures have profound effects on amphibians since they are ectothermic organisms. Although amphibians in temporary habitats regulate the speed of their metamorphic phases depending on water temperature, encouraging pond abandon when necessary (García-París et al., 2004), extreme thermal increases may result in population declines triggered by low fertilities and poor larval developments (Bowers et al., 2000; Bury, 2008; Galloy and Denoël, 2010).

Regarding predation, many studies report the negative effect of exotic predators such as introduced fishes (Bressi and Stoch, 1999; Adams, 2000; Kloskowski, 2009) and invasive crayfishes (such as red swamp crayfish, Cruz et al., 2005) on native amphibian species. In the area of study a native predator was particularly hazardous for amphibians. *A. obstetricans* was the unique amphibian reported in Bekosare, a pond devoted to preserve the sparse populations of the highly endangered crayfish *A. pallipes* (Table 2). A negative trade-off among preservation objectives emerges in Bekosare, and managers need to be concerned on the predictable negative effect that the native crayfish population exerts on the amphibian community. Regarding the introduced fish *Tinca tinca* in Olaberri pond, two amphibian species sensitive to fish predation, *Triturus marmoratus* and *H. arborea*, missed the reservoir (Ildos and Ancona, 1994; Alarcos et al., 2003), but other five species inhabited it, which suggested that the effects of *T. tinca* on the amphibian community were less negative than those of the native crayfish (Table 2).

Eventually, regarding aquatic vegetation, some studies have correlated the presence of *R. perezi* and some species of *Triturus* spp. with its development (Alarcos et al., 2003; Cirovic et al., 2008). In accordance, we reported *R. perezi*, *T. helveticus* and *T. alpestris* in ponds with percentages of submerged vegetation higher than 25%. However, an excess of nutrients and vegetation may entail a severe risk of pond eutrophication and cause a sharp reduction of dissolved oxygen. Both livestock and amphibians may be largely affected by this process. In order to avoid an excessive development of aquatic vegetation and an increase of water oxidability, regular cleaning tasks are important in ponds. The harvest and removal of the aquatic vegetation helps control nutrients retained in ponds, such as nitrates and other contaminants more important in anoxic conditions (Matheson and Sukias, 2010). These jobs should be restricted to autumn, when most amphibians develop its terrestrial cycle and grazers abandon the area, taking special care on surrounding vegetation, where adult amphibians may hibernate. Pond design is specially relevant at that point (type of fencing, pond edges, pond sealing, existence of drainpipes for drainage,. . .) to ensure the efficiency of the cleaning process.

5. Final remarks

Artificial ponds in extensive rangelands are expected to provide a suitable habitat for amphibian populations and to satisfy livestock demands. Most scientific published works deal with the environmental use of ponds, overlooking domestic animals that drink on them and from which ponds were initially created. Contrarily, technical reports focus mostly on the optimal design that ensures an adequate livestock water supply, neglecting wildlife requirements. In this manuscript, we have intended to join both points of view in order to identify negative trade-offs that have to be considered in pond design.

Results of this study indicate that risks associated to the dual use of ponds mostly occur in summer when there is a scarcity of water recharge and a rising domestic stocking rate. A scarce water recharge reduces the hydroperiod, concentrates pollutants, lowers water level and increases water temperature. Because of this, an optimal pond design should ensure a continuum water recharge along the dry season in these temperate environments. Concerning the second point, the study demonstrates that the negative effects of livestock on ponds tightly depend on fencing. The existence of fences around ponds reduces water degradation, ensures a strip of surrounding vegetation and decreases the chance of mechanical damage (on amphibian eggs and larvae) caused by the wading and trampling of livestock. Water in unfenced ponds degrades in summer with urine and feces of mammals, which increases the dissolved ammonia and boosts the enteric microbial charge. This chemical and microbiological contamination may largely affect livestock whereas amphibians may escape if accomplishing its breeding stage before water degradation. However, despite fencing is the most effective measure to preserve water quality, it does not prevent the occurrence of an hazardous microbial charge caused by birds and small mammals accessing fenced ponds.

Eventually, even though managers are particularly concerned with constructive traits of ponds, biotic attributes such as the quality of the surrounding habitat and the occurrence of predators in ponds are extremely important for amphibian assemblages. The first variable encouraging the occurrence of a given amphibian species in a pond is the surrounding environment. Ponds sampled in this study were surrounded by different types of vegetation, from open grasslands to dense forests. The endangered species *S. salamandra* was only found in Mármol, a pond enclosed in a woodland habitat, which offered the moist and shady environment needed by the species. At first sight, Mármol constructive traits were not suitable for amphibians (plastic sealing of black polyethylene with steep slopes that warmed excessively during summer), however, the number of species encountered in this pond was as high as in other more amphibians-friendly ponds, such as Ilusiar (bentonite sealing). Further studies should focus on amphibian population densities and dynamics in order to ascertain the long-term viability of the species assemblages encountered in these ponds.

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