

## Bullfrog (*Lithobates catesbeianus*) invasion in Uruguay

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**Abstract** This is the first report of North American bullfrogs, *Lithobates catesbeianus* (= *Rana catesbeiana*), invasion in Uruguay. This Anura was introduced for farming proposes in 1987, but at present most of the farms are closed. At one of these closed farms, located at Rincón de Pando, Canelones, we report the occurrence of a feral population of *L. catesbeianus*. This invasion point is at an early stage and restricted to one or two ponds. We also report the effects of *L. catesbeianus* invasion in the community structure. This includes species composition and species size structure. In this system bullfrog tadpoles constitute a very important proportion of the present biomass. Bullfrog tadpoles appear to be displacing native amphibians and having some type of positive

interaction with fishes. At the invaded system we found more fish species and larger sizes of the shared fish species. We analyze the involved risks of this invasion, the ecological impact by predation, the competition and habitat modification, and the potential of bullfrog to act as pathogens vector. We also recommend taking measures in order to avoid the expansion of this population. There is also the need of studies to search for new invasion points in Uruguay, especially where bullfrog farms were located.

**Keywords** Amphibian invasion · *Lithobates catesbeianus* · Tadpole · Uruguay

### Introduction

Bullfrog, *Lithobates catesbeianus* (Shaw 1802) (= *Rana catesbeiana*), is an important structuring agent of anuran assemblages in their native range, east of the Great Plains, North America (Hecnar and M'Closkey 1997). It has been introduced in the major part of USA and in several countries around the world for aquaculture proposes related with its potential alimentary trade, and also as biological control agent and as an ornamental species (Jennings and Hayes 1985). As a consequence, bullfrog populations could have established at different sites throughout the world: in western North America, Europe, Asia, South America, and the Caribbean (Stumpel 1992; Global Biodiversity Information Facility 2006).

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Its ecological attributes, large body size, broad diet, frequently high population densities, and capacity to invade natural environments, facilitates its potential to impact on different taxa through predation, competition, and habitat modification (Stumpel 1992; Kiesecker and Blaustein 1998; Pearl et al. 2004; Global Invasive Species Database 2005). *L. catesbeianus* is considered one of the major causes of global amphibian population declines (Alford and Richards 1999; Blaustein and Kiesecker 2002). Several experimental (Kiesecker and Blaustein 1997; Lawler et al. 1999; Kiesecker et al. 2001; Boone et al. 2007) and field studies (Dumas 1966; Moyle 1973; Licht 1974; Fisher and Shaffer 1996) have found negative ecological effects of bullfrog invasions on biological communities.

Bullfrog farming in Uruguay started in 1987 with the promotion of local authorities. From 1993 to 2000, 18 private bullfrog farms were established in closed production systems throughout the country. Nevertheless, the farmers did not obtain great economic gains and their interest strongly declined. At present, few bullfrog farms remain working (Carnevia 2005). No control programs have been implemented for the closed farms in order to prevent bullfrog escapes, or releases.

Here we report the presence of a feral bullfrog population at Rincón de Pando, Canelones, Uruguay, whose source could be an unintentional release from one of those farms. We also analyze their interactions and effects on native aquatic species.

## Materials and methods

### Invasion site

Rincón de Pando is located near Pando City, next to Pando Stream (34°44'20"S; 55°55'30"W), in the province of Canelones, Uruguay; 35 km. east from Montevideo. The local landscape consists of highly anthropic modified grasslands and wetlands in the proximity of the Pando Stream. It consists of small agricultural farms, with many artificial ponds for irrigation and cattle watering purposes. Local Anuran assemblages are composed of the following species: *Pseudis minutus*, *Odontophrynus americanus*,

*Hypsiboas pulchellus*, *Chaunus arenarum*, *C. gr. granulatus*, *Leptodactylus gracilis*, *L. mystacinus*, *L. ocellatus*, *L. latinasus*, *Pseudopaludicola falcipes*, *Scinax granulatus*, *S. squalirostris*, and *Elachistocleis bicolor* (Nuñez et al. 2004).

A bullfrog farm was established at the site until 1993, when it was abandoned, leaving the frogs alive, inside the breeding facilities. There are no records about what happened since that moment, but local people informed us that those bullfrogs were released, and that there were many escapes, even when the commercial exploitation was running (Maneyro et al. 2005).

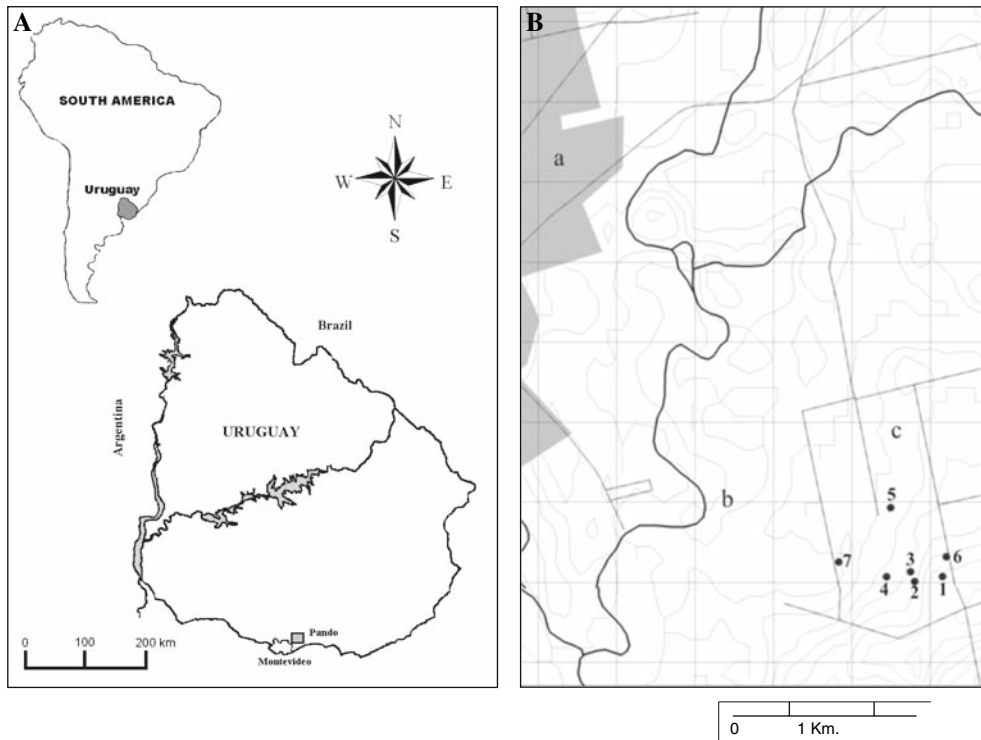
### Sampling methods

In our first exploratory visit to the site (April 2005), we collected five *L. catesbeianus* tadpoles in pond 6 (Fig. 1), with the aid of a hand net (Maneyro et al. 2005). Then we came again to study the site in June 2005 (winter in the Southern Hemisphere) to obtain information about this population.

We sampled seven water systems, ponds and creeks, of the watershed that follows the landscape slope, down from the closed farm point (42 m above sea level) to Pando Stream, as the easiest route of the possible bullfrog dispersion (Fig. 1). Our samples were focused in larval specimens, easier to find and collect than adults (Altig and McDiarmid 1999a). We used a hand trawl net (net diameter 1 m; mesh diameter 5 mm), taking a standardized sample: maximum and cross-section pond axis, and half of pond perimeter. All the animals collected were sacrificed, fixed in 10% formalin, and deposited at the Vertebrate Collection of Facultad de Ciencias (ZVCB), Universidad de la República, Uruguay.

### Data analysis

All the collected tadpoles' species were determined at the laboratory, staged (Gosner 1960), and measured with a digital caliper (to the nearest of 0.01 mm). Measures follow Altig and McDiarmid (1999b): total length (TL) and body length (BL). Fishes were determined and also measured, obtaining standard



**Fig. 1** (A) Site map. Invasion site location in Uruguay and Pando Stream basin. (B) a- Pando City, b- Pando Stream basin, c- Microbasin showing the sampled water bodies, numbered 1–7

length (SL, from snout tip to peduncle end). Total biomass was calculated for each vertebrate species in each sample; proportions in percentages were calculated.

We analyzed the vertebrate assemblage conformation in the invaded system, by comparing it with two other sampled systems of similar geomorphologic and hydric regime characteristics (numbers 2 and 4 in Fig. 1). The other systems were not considered in the analysis due to their different ecosystem characteristics. We analyzed the differences in vertebrate assemblages by a Canonical Correspondence Analysis (CA), associating each community with its species composition (Greenacre 1984). We excluded from the analysis the fish species that only occurred in one system at low densities, in order to obtain clearer results. The two most abundant fishes species were divided into three size classes: *C. interruptus* A (<340 mm), *C. interruptus* B (from 340 to 380 mm), *C. interruptus* C (more than 380 mm), and *C. descenmaculatus* A (<240 mm), *C. descenmaculatus* B (from 240 to 260 mm), and *C. descenmaculatus* C (more than 260 mm).

## Results

### Population occurrence confirmation and invasion status

We did not find any *L. catesbeianus* specimens in pond 6, in June, where we collected the first five tadpoles in April (Maneyro et al. 2005). In pond 1 (Fig. 1), we collected 86 bullfrog tadpoles, ranging from 450 to 978 mm of TL ( $x = 763$  mm and  $SD = 10.16$ ) and from 174 to 351 mm of BL ( $x = 280$  mm and  $SD = 3.41$ ). The developmental stage ranged from stage 25 to 31 (sensu Gosner 1960). Systems characteristics as well as present vertebrate species are numbered in Table 1.

### Invaded pond community: ecological effects

The invaded system (pond 1) was found to have a vertebrate assemblage composed by four fish species (*Cnesterodon descenmaculatus*, *Cheirodon interruptus*, *Cichlasoma facetum*, and *Gymnogeophagus*

**Table 1** Aquatic systems, dimensions, and sampled vertebrate species

System	Area (m <sup>2</sup> )	Tadpoles and relative mass	Fishes and relative mass
1	276	<i>R. catesbeiana</i> (73.8%)	<i>C. descenmaculatus</i> (2.6%) <i>C. interruptus</i> (21.5%) <i>G. meridionalis</i> (1.0%) <i>C. facetum</i> (1.1%)
2	644	<i>H. pulchellus</i> (6%)	<i>C. descenmaculatus</i> (6.6%) <i>C. interruptus</i> (87.4%)
3	391	–	–
4	6,400	<i>H. pulchellus</i> (0.3%)	<i>C. descenmaculatus</i> (1.8%) <i>C. interruptus</i> (97.9%)
5	360	<i>H. pulchellus</i> (100%)	–
6	837	<i>H. pulchellus</i> (2.7%)	<i>C. descenmaculatus</i> (97.3%)
7	350	<i>H. pulchellus</i> (37.3%)	<i>C. descenmaculatus</i> (62.7%)

The water bodies are numbered following the numeration of the map (see Fig. 1)

*meridionalis*) and *L. catesbeianus* tadpoles. This assemblage composition was different from that found at the other sampled systems.

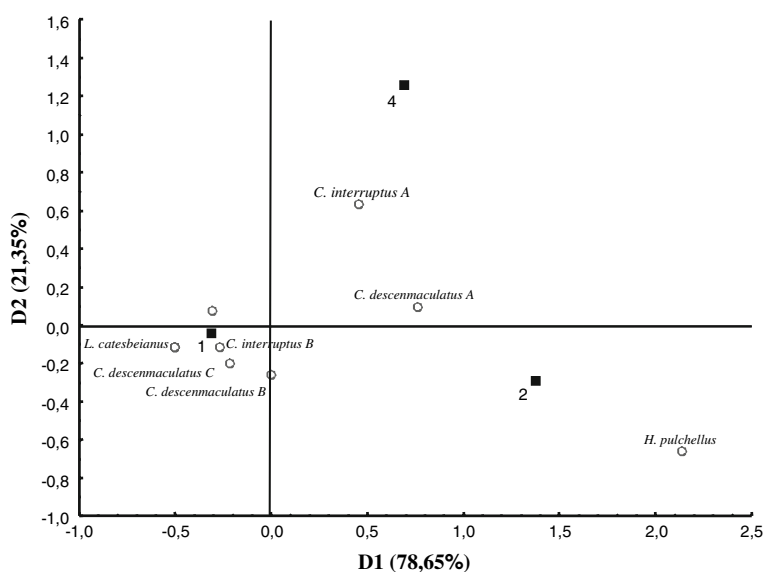
The CA first two eigenvalues explains 79% ( $\chi^2 = 161.0712$ ), and 21% ( $\chi^2 = 43.7267$ ) of the total system inertia. The first component shows a differentiation in the community structure between the invaded pond (pond 1) and the other two (ponds 2 and 4). This differentiation is explained by differences at species (and size classes) composition; on the right of axis 1 (Fig. 2) are placed the ponds containing *H. pulchellus* tadpoles, and the small-sized fishes (*C. interruptus* A and *C. descenmaculatus* A). On the left, is located the invaded pond, containing *L. catesbeianus*, and the mid- and large-sized

(B and C) of *C. interruptus* and *C. descenmaculatus*. The second dimension makes a differentiation between the two ponds that were not invaded and that are therefore of no relevance to our research.

## Discussion

Our finding focuses on the first recorded feral specimens of *L. catesbeianus* in Uruguay. Bullfrog is well studied as an alien species in Northern Hemisphere, but its invasions in other countries are not well known. *L. catesbeianus* have been recently reported in southern South America, in the south, southeast and center of Brazil (Borges-Martins et al.

**Fig. 2** Correspondence analysis for ponds 1 (invaded by *R. catesbeiana*) and other two comparable systems (pond 2 and pond 4). ■ Indicates each community and ○ indicates each species. *C. interruptus* and *C. descenmaculatus* are divided into three size classes, according to their SL ( $SL_A < SL_B < SL_C$ )



2002; Rocha-Miranda et al. 2006), and in Argentina (Sanabria et al. 2005; Global Biodiversity Information Facility 2006; Pereyra et al. 2006), in the surroundings of aquaculture facilities. The mentioned authors suggest negative effects of the invasion on local biota, but not giving evidence about this. Our contribution consists of the report of a new invasion site and the analysis of its effects in aquatic communities.

There is a small feral population of *L. catesbeianus* at Rincón de Pando, confined to a small area (one or two ponds), and located at a short distance from the original frog farm facilities. Most of our collected specimens were tadpoles (winter sampling, June), but at a later visit to the site (November 2005, Spring in the Southern Hemisphere) we also collected one mature female and an egg mass, and we registered vocalizing males, in the same site (not published data). This invasion could be in the stage of establishment of a self-sustaining population within the new habitat. Studies on initial stages are not abundant and necessary in order to detect exotic species before they can affect community structure and ecosystem function (Puth and Post 2005).

Despite of having data on only one invaded pond, due to the wide spread of bullfrog in the study site, is important to analyze its effects on native communities. The invaded pond community exhibit significant differences in relation with the not invaded ponds: (1) the absence of native *H. pulchellus* larva, (2) the highest aquatic vertebrate richness, (3) a higher body size of the common fish species, and (4) *L. catesbeianus* tadpoles are the highest fraction of total vertebrate biomass.

Bullfrogs have negative effects on other amphibian fitness, reducing the premetamorphic period by competition (Seale and Beckvar 1980; Kupferberg 1997; Lawler et al. 1999; Adams 2000; Boone et al. 2004) and affecting survivorship by predation (Stewart and Sandison 1972; Ehrlich 1979; McAlpine and Dilworth 1989; Blaustein and Kiesecker 2002). The effect of the bullfrog on other amphibian is context dependent (Kiesecker et al. 2001) and could be asymmetrical (Pearl et al. 2004). Native aquatic frogs, as *P. minutus*, could have longer potential exposure to *L. catesbeianus* predation due to the habitat affinities of both species (Stewart and Sandison 1972; Melchioris et al. 2004). However, since these hypotheses are based on patterns observed in

other countries, at moment, we can only speculate as to what could happen in Uruguay.

Many ecological mechanisms could be explaining the differences of the invaded pond community structure. Bullfrog tadpole's can control the primary productivity because of their high algal consumption rate (Alford 1999; Hamer et al. 2003) and can also have positive indirect interaction with fishes (Smith et al. 1999; Adams et al. 2003; Boone and Semlitsch 2003). This fact could explain the differences in the fish assemblages, increasing grazing species (*C. descenmaculatus* and *C. interruptus*) sizes and the presence of a higher trophic level (*C. facetum* and *G. meridionalis*) at pond 1. We also did not observe invertebrate predators (Odonata larvae and belostomatids) at pond 1, which were present at the other ponds. Werner et al. (1995) attributed the facilitation to the lack of invertebrate predators in presence of fishes.

The local conditions—high anthropogenic disturbances and considerable amount of suitable habitat—may facilitate *L. catesbeianus* population expansion (Ryan 1978; Kupferberg 1997; Walker and Busack 2000; Boone et al. 2004). There are no geographical barriers in the area that could stop this invasion and, if bullfrogs reach the wetlands and the Pando Stream, it will become difficult to control. A recent prediction of the potential global distribution of *L. catesbeianus*, assigns the region as one of the highest suitability for this species (Ficetola et al. 2007).

Other involved risk is that *L. catesbeianus* can act as a vector for pathogens micro-organisms, especially *Batrachochytrium dendrobatidis*, responsible for chytridiomycosis, an emerging disease identified as one of the main causes of amphibian mass mortality events and global amphibian declines (Berger et al. 1998; Longcore et al. 1999; Ron and Merino 2000; Bonaccorso et al. 2003; Daszak et al. 2003). *L. catesbeianus* is relatively resistant to chytridiomycosis, so it can be considered as an efficient carrier of the pathogen (Daszak et al. 2004; Hanselmann et al. 2004; Garner et al. 2006). *B. dendrobatidis* was recently reported in farmed bullfrogs in Uruguay (Mazzoni et al. 2003). Our findings are an alert, not only by the ecological risk of *L. catesbeianus* invasion, but also by the sanitary risks implicated. Predation and interspecific competition could be manageable, but the possible presence of *B. dendrobatidis* must be explored, considering amphibian pathogens also invasive (Garner et al. 2006).

Due to the earliest stage of the here reported invasion, there is a unique opportunity to act in order to control an invasion focus in a primary step so, the implementation of an eradication plan should be easier and cheaper than later actions. Management strategies, such as the combination of shooting adults and draining ponds, seem to have a positive effect for certain regions (Doubledee et al. 2003).

Considering this experience it is important to monitor what happened with the 18 farms that once existed in Uruguay. In addition, authorities would be more efficient in the control of such dangerous species such as the bullfrog. We therefore recommend extreme precautions with the introduction of new species in Uruguay, as well as with already introduced species that have not yet invaded.

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