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Reproductive features of *Chaltenobatrachus grandisonae* (Anura: Batrachylidae) within a protected area in Patagonia, Chile

Características reproductivas de *Chaltenobatrachus grandisonae* (Anura: Batrachylidae) en un área protegida en Patagonia, Chile

JAVIERA CISTERNAS[1,2,*], CLAUDIO CORREA[1,3], NELSON VELÁSQUEZ[2] & MARIO PENNA[2]

[1]Aumen o el Eco de los montes, Organización No Gubernamental, P. O. Box 393, Coyhaique, Chile
[2]Universidad de Chile, Facultad de Medicina, Instituto de Ciencias Biomédicas, P. O. Box 70005, Santiago, Chile
[3]Pontificia Universidad Católica de Chile, Departamento de Ecología, Alameda 340, P. O. Box 6513677, Santiago, Chile
[*]Corresponding author: javiera.cisternas.tirapegui@gmail.com

Basso et al. (2011) assigned the monotypic genus *Chaltenobatrachus* for the species described originally as *Telmatobius grandisonae* Lynch, 1975 (later transferred to the genus *Atelognathus* by Lynch 1978). The type locality of *Chaltenobatrachus grandisonae* (Lynch, 1975) is Puerto Edén (Wellington Island) in the Magallanes Region, Chile, where the species has not been found again. Basso et al. (2011) added two new localities from Argentina and provided a detailed description that includes morphological and osteological characteristics of adult specimens, external morphology of tadpoles, karyotype and phylogenetic relationships. They also provide a few field observations, highlighting the lack of knowledge of the natural history and population biology of this anuran.

In this work we report a new locality for *C. grandisonae* in Chile, extend its altitudinal distributional range and report on its reproductive mode. The study site is within the protected area Laguna Caiquenes (LCPA), located in the Aysén Region, which holds 9.000 ha of evergreen forests of *Nothofagus betuloides* and regrowth of *Drimys winteri* with bogs of *Poaceae* and *Cyperaceae*. In this locality, *C. grandisonae* cohabits with other five anuran species: *Alsodes coppingeri* (Günther, 1881); *Batrachyla antartandica* Barrio, 1967; *Batrachyla taeniata* (Girard, 1855); *Eupsophus calcaratus* (Günther, 1881) and *Nannophryne variegata* Günther, 1870. Data on the reproductive activity of these species are also reported here.

Reproductive mode is defined by a combination of characteristics including breeding site, clutch structure, location of egg deposition, larval development site and parental care (Wells 2007, Vitt & Caldwell 2009). Clutch structure was defined as the type of oviposition (rosary-like strings or clusters) and as parental care we considered any form of post-ovipositional parental behavior that could increase the survival of the offspring, such as nest attendance. Because temperature and rainfall are determinants of seasonality in anuran reproduction (Vitt & Caldwell 2009), we report phenological changes of reproductive indicators (Fig. 1A) in relation to these environmental factors (Fig. 1B).

Field observations were performed during nine trips between October 2007 and December 2012. Individuals were searched in areas with water availability, captured and checked with a standard table of field observations and then released at the same place where they were observed. Overall, 35 points were sampled, most of which were resurveyed on each trip. All procedures used comply with the laws of animal welfare in Chile (capture permit from the Servicio Agrícola Ganadero (SAG), resolution 5090 / 2011).

Our results indicate that *C. grandisonae* lays its eggs in clusters attached to branches or stones under the water (Fig. 2A and 2B). We found three clusters of 14, 21 and 30 eggs. Tadpoles develop in lentic waters (mainly temporary ponds) (Fig. 2C and 2D). Breeding sites are located in the pond edges (altitudinal

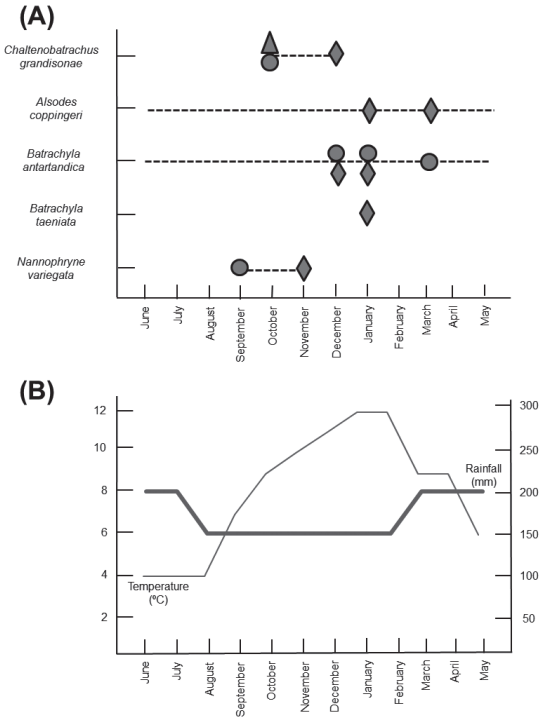

Fig. 1: Phenological changes of reproductive indicators in relation to environmental factors. (A)Temporal distribution of reproductive events of five amphibian species in Laguna Caiquenes Protected Area. (B) Monthly Mean Temperature and Monthly Mean Precipitation for Laguna Caiquenes Protected Area shown in the Atlas Climatológico de Chile (Dirección Meteorológica de Chile 2007). The figure summarizes accumulated field observations made in nine field trips between October 2007 and December 2012. There are no observations of *Eupsophus calcaratus*. Symbology: ▲ amplexus, ⬮ egg clutches, ---- larval development, ◆ metamorphosing tadpoles.

Fenología de indicadores reproductivos en relación a factores ambientales. (A) Distribución temporal de eventos reproductivos de cinco especies de anfibios presentes en el Área de Protección Laguna Caiquenes. (B) Temperatura media mensual y Precipitación media mensual para el Área de Protección Laguna Caiquenes publicado en el Atlas Climatológico de Chile (Dirección Meteorológica de Chile 2007). La figura resume la acumulación de observaciones realizadas en nueve visitas entre Octubre 2007 y Diciembre 2012. No se registraron observaciones para *Eupsophus calcaratus*. Simbología: ▲ amplexo, ⬮ ovipostura, ---- desarrollo larval, ◆ individuos postmetamórficos.

range: 288 - 942 masl; both records extend the altitudinal limits of this species). The amplexus is axillary (Fig. 2E) and parental care was not observed. This reproductive mode differs from that of the other five species found in the area; none of these paste their eggs to a substrate. General descriptions of the reproductive modes of the other species were reported by Soto et al. (2008).

Observations throughout the six-year period showed that reproduction for *C. grandisonae* in this area begins with the amplexus and egg deposition in October (middle spring), following with larval development for 10 - 12 weeks, and ending with metamorphosing tadpoles in December (early summer) (Fig. 2F). Oviposition of *N. variegata* was observed in September and larval development was observed until November. Egg clutches of *B. antartandica* were observed from December through March and larval development occurred throughout the year. *Alsodes coppingeri* also exhibits larval development throughout the year. Metamorphs of *A. coppingeri* and *B. taeniata* were observed in January. No reproductive behavior of *Eupsophus calcaratus* was observed.

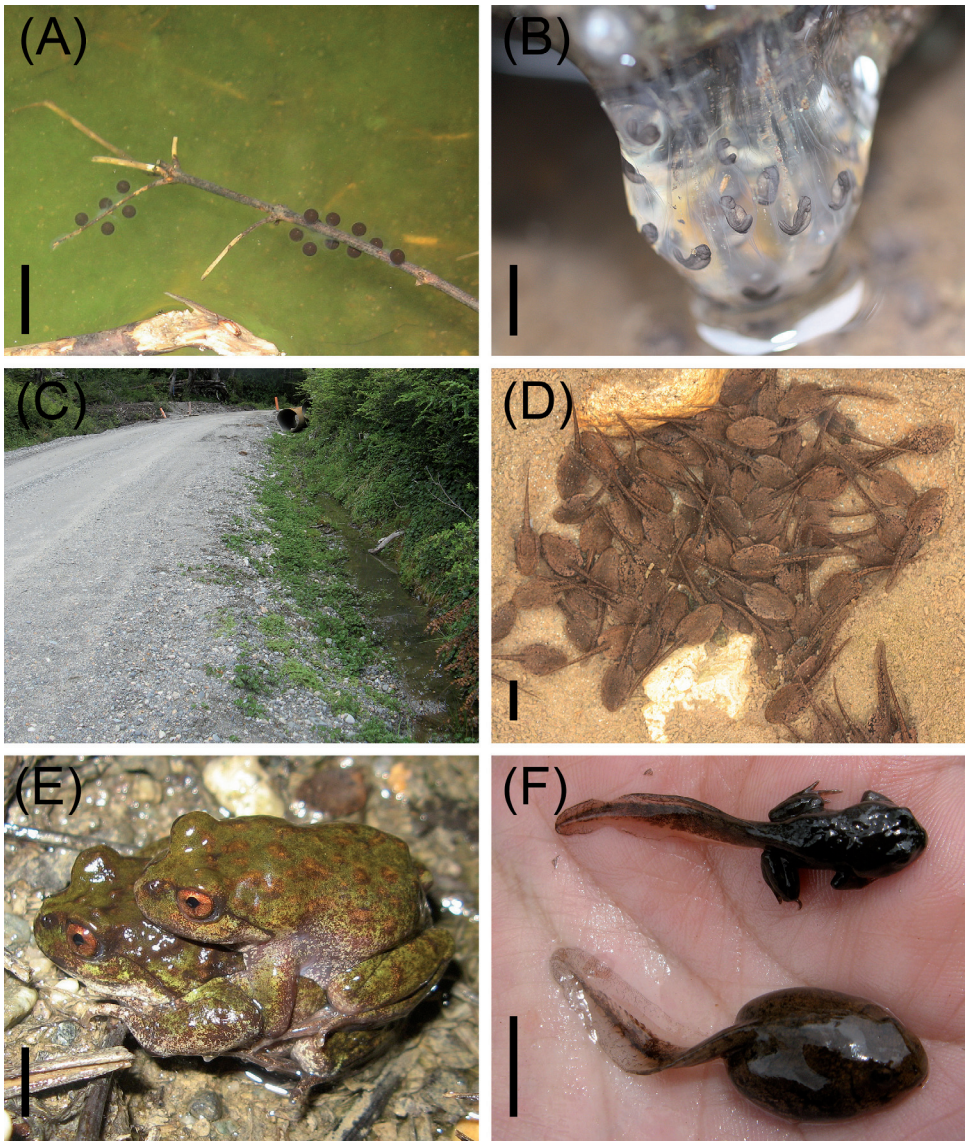

Fig. 2: Microhabitat and life cycle stages of *Chaltenobatrachus grandisonae*. A) Eggs pasted to a branch under water. B) Enclosed embryos in a clutch pasted to a stone. C) Temporary pond located at the border of the Austral highway. D) Tadpoles in a pond with organic matter on the bottom. E) Adults in amplexus. F) Larvae in Gosner stages 32 and 43. Scale bars correspond to 1 cm.

Microhábitat y etapas del ciclo de vida de *Chaltenobatrachus grandisonae*. A) Huevos adheridos a una rama bajo el agua. B) Embriones adheridos a una piedra. C) Poza temporal situada al borde de la Carretera Austral. D) Larvas en una poza con materia orgánica en el fondo. E) Adultos en amplexo. F) Larvas en estadíos Gosner 32 y 43. Barras de escala corresponden a 1 cm.

Antecedents reported here are the first on the breeding biology of *C. grandisonae*. Our data differ from those of Basso et al. (2011), who suggested a prolonged development and overwintering of larvae in the water bodies. We observed two different types of ponds where the larval development of this species occurs. One corresponds to ponds within areas of forest re-growth where the substrate is organic matter that maintain temperatures above 0 °C even when water surface freezes. The second type corresponded to temporal ponds located along the highway left after the road construction, in which water persists for four months. In fact, road construction also contributes with the

creation of reproductive sites for *A. coppingeri*, *B. antartandica* and *B. taeniata*.

This preliminary report on the phenology of reproduction of *C. grandisonae* and sympatric species in LCPA provide the basis for further studies on the autoecology and monitoring of these populations (e.g. Buckley & Beebee 2004). In particular, the poorly known *C. grandisonae* and *A. coppingeri* could be preferential targets for conservation and management in this and other protected areas in the region.

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