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On a second invasive frog (Anura: Leptodactylidae) from San Andrés, Colombian Caribbean islands

Sobre la segunda rana invasora (Anura: Leptodactylidae) de San Andrés, Caribe insular colombiano

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Islands are optimal model systems to understand evolutionary processes such as colonization, speciation, and extinction. Their size and distance to the continent are key factors that shape island biodiversity (Zunino & Zullini 2003, Whittaker & Fernández-Palacios 2007). Human have aided the displacement of species to localities that could have otherwise not taken place through natural dispersal across geographical barriers (Ríos & Vargas 2003). Invasive species, particularly in islands, can cause significant ecological changes in the trophic web structure and habitat composition, including severe impacts on native species and ecosystems (Gutiérrez-Bonilla 2006). Invasive species are included among the severe threats that habitats face (*i.e*., overexploitation of natural resources, urban development, pollution, ecosystem modification, among others; see Russel & Kueffer 2019).

The Archipiélago de San Andrés, Providencia, and Santa Catalina (Colombian Caribbean islands) is part of the Western Caribbean, an insular ecoregion recognized by its high diversity and endemicity (Hedges *et al.* 2019). Among the Colombian Caribbean islands, San Andrés is a coral reef island, which is the largest, most diverse, and more impacted by anthropogenic disturbance (Caicedo-Portilla 2014). This is located 483 km northwest of the Colombian Caribbean coast and 190 km east of the Caribbean coast of Central America (Fig. 1). The island is part of the dry tropical zonobiome (Hernández-Camacho & Sánchez 1992), with an iso-megathermal warm climate and unimodal rainfall regime (rainfall season from May to November, the driest periods from March to April, Vargas-Cuervo 2004). The natural ecosystems of San Andrés include tropical dry and mangroove forests, but the predominant coverage are small remnants of the original forest, surrounded by pastures, crops, and shrubs (Caicedo-Portilla 2014).

The herpetofauna of San Andrés has been previously documented with a list of 19 terrestrial species (17 reptiles and two anurans; Dunn 1945, Dunn & Saxe 1950, Tamsitt & Valdivieso 1963, Valdivieso & Tamsitt 1963, McNish 2011, Cubillos-Abrahams *et al.* 2021). The potential origin of the island species was recently summarized by Cubillos-Abrahams *et al*. (2021). Before this publication, the only species of frog documented in San Andrés in literature and museums was *Leptodactylus insularum* Barbour, 1906. Nevertheless, *Eleutherodactylus planirostris* (Cope, 1862) was documented for the first time in 2018 (Cubillos-Abrahams *et al.* 2021). A recent expedition to San Andrés (March and June 2021) found hundreds of a leptodactylid frog that did not correspond to the native species *L. insularum*. A detailed examination of specimens and advertisement calls allowed us to identify them as the white-lipped thin-finger frog, *Leptodactylus fragilis* (Brocchi, 1877), a widely distributed species, ranging from southern to northern South America (Heyer *et al*. 2006, Mendez-Narváez *et al*. 2009) and recently reported as

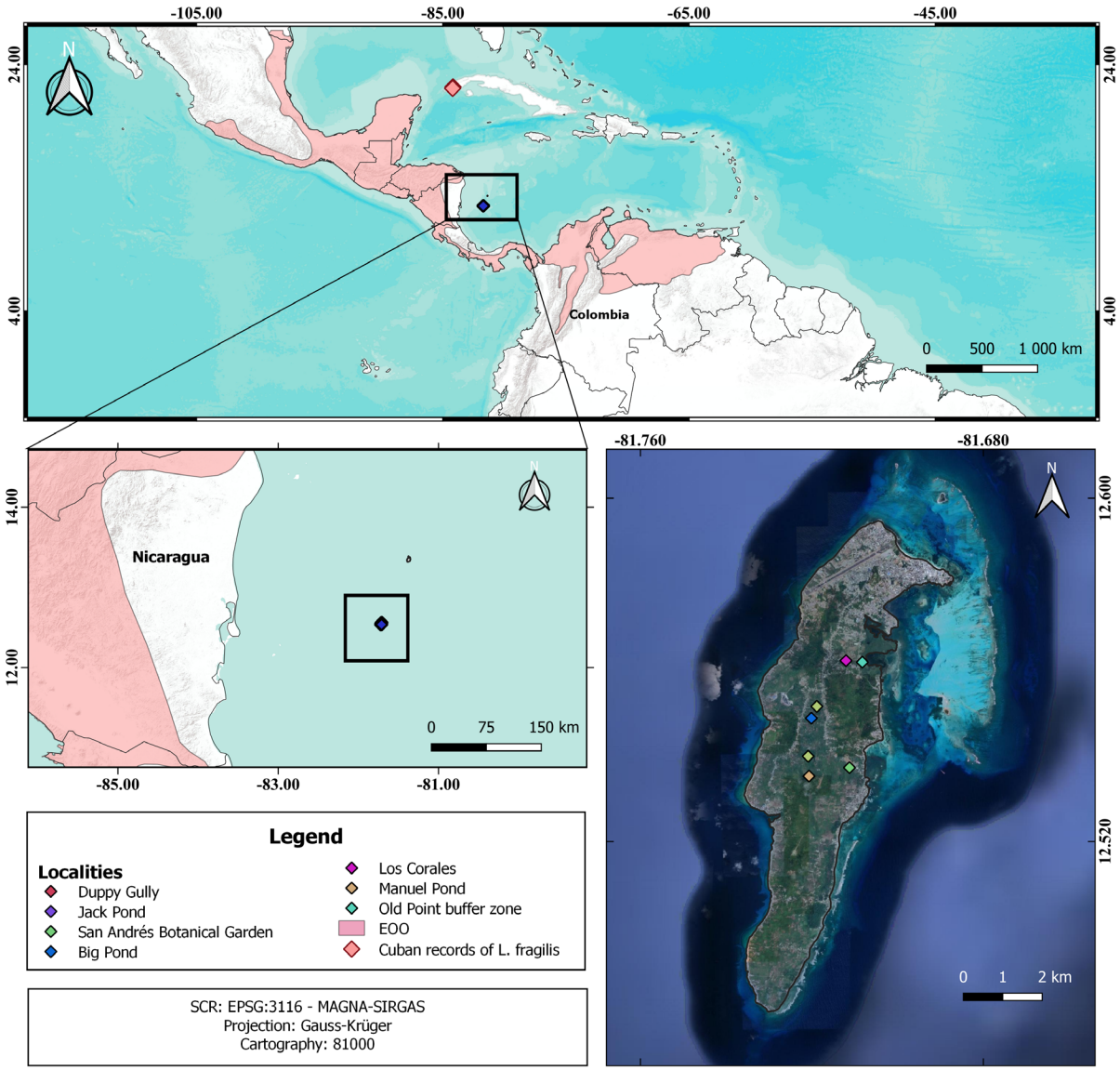

Figure 1. Geographic distribution of *Leptodactylus fragilis,* based on occurrences from GBIF (2021), the extent of occurrences (EOO) from UICN Red List (Heyer et al. 2010), and the recent record from Cuba by Rodríguez-Cabrera et al. (2018).

an introduced species to Cuba (Rodríguez-Cabrera *et al*. 2018). In Colombia, the species is distributed on the continental Caribbean lowlands, inter-Andean valleys, and the western portion of the Orinoquian savannahs (Acosta-Galvis 2021). Here, we report for the first time the presence of the white-lipped thin-finger frog *L. fragilis* from San Andrés, in the Colombian Caribbean islands. In addition, we report and describe some aspects of the species natural history in San Andres.

Leptodactylus fragilis was recorded at seven localities in the central area of the island. All the localities are part of semi-rural to rural areas characterize by the presence of forest remnants, shrubs, pastures, and build-up areas. Taxonomic determination was initially based on the dichotomous key of Cuentas *et al.* (2002) for the anurans of northern Colombia. Subsequently, the identity of the species was confirmed by verifying the descriptions by Heyer *et al*. (2006) and de Sá *et al*. (2014). Specimens collected were euthanized following the ASIH guidelines using a chemical anesthetic, later fixed in a formalin solution (10%) and finally maintained in alcohol (70%). All vouchers are deposited at the amphibian collection of the Centro de Colecciones Científicas at the Universidad del Magdalena (CBUMAG), Santa Marta, Colombia, with the following collection numbers: CBUMAG: ANF: 01188-89, 01199-201, 01203-04.

Herein, we report the new records of *Leptodactylus fragilis*: Colombia, departamento Archipiélago de San Andrés, Providencia y Santa Catalina: San Andrés (Fig. 1): Los Corales (12°33'43.7" N, 81°42'43.4" W, 21 m asl.; CBUMAG: ANF: 01188-89 [Fig. 2A], CBUMAG: ANF: 01199-201), buffer zone of Old Point Mangrove Regional Park (12°33'42.4" N, 81°42' 29.7" W, 4 m asl.), Jack Pond (12°33'05.3" N, 81°43'07.9" W, 50 m asl.), Big Pond (12°32'55.4" N, 81°43'12.5" W, 44 m asl.; CBUMAG: ANF: 01203-04), Duppy Gully (12°32'23.5" N, 81°43'15.2" W, 32 m asl.), Manuel Pond (12°32'06.8" N, 81°43'14.5" W, 32 m asl.), and Botanical Garden of San Andrés (12°32'13.9"N, 81°42'40.5" W, 28 m asl.). At these new localities, specimens were found sharing their habitat with *E. planirostris* and *L. insularum* in pastures, pond shores, tropical dry and mangrove forests edges. In San Andres, the species is found on different substrates, *i.e.*, mud, leaf-litter, wet grasses, roots, bare soil, and cattle footprints and droppings. *Leptodactylus fragilis* vocalizes throughout all the night, but apparently, the peak of vocalizations is in the early hours (19:00–21:00 h). The advertisement calls of *L. fragilis* was so deafening that generally we did not hear advertisement calls of *L. insularum*. In fact, the species was abundant and exceeding by far *L. insularum* in all localities.

We found hundreds of individuals in the surroundings of water sources such as Jack Pond, Manuel Pond, and Big Pond, the latter is permanent pond. Seasonal water bodies are heavily sedimented and have a large mass of floating macrophytes (*Pistia stratiotes*), which almost completely cover the muddy lake beds exposed during the dry season. This seems to be an important microhabitat for *L. fragilis*, although it was also frequently found in the leaf-litter of remnants vegetation surrounding the water bodies. In localities without ponds, such as the Botanical Garden of San Andrés, Los Corales, and the buffer zone of Old Point Regional Park, the specimens gathered in humid shelters, *e.g.*, wet surfaces due to water leaks, irrigation zones, drainage gutters, greenhouses, and gardens. During rainy days, specimens were found at the edges of tropical dry forest.

We recorded the reproductive activity of *Leptodactylus fragilis* from the beginning to the end of the fieldwork period. We heard the advertisement call and found foam nests with tadpoles in the driest time of the year (March to April). Foam nests were deposited in humid cavities at the floor level (Fig. 2B), under fallen logs, boards, boulders, and coral rocks, which were not in direct contact with water. We observed parental care in four foam nests that involves the males sitting next to the nest within the burrow.

Specimens were identified as *Leptodactylus fragilis* by the following combination of characters (Heyer *et al*. 2006, de Sá *et al*. 2014): relatively small size (maximum size in snout-vent length [SVL]: 44 mm in females, 43 mm in males), head longer than wide, spatulated snout, bulging in lateral view and rounded canthus rostralis. Two to four longitudinal folds, two weak dorsolateral and two discontinuous lateral. Lateral fringes of toes absent. All specimens had a white stripe on the upper lip and a dark supratympanic fold. Two lateral vocal sacs, from pigmented grayish to heavily dark. *Leptodactylus fragilis* is easily distinguished from *L. insularum* (morphological characters of *L. insularum* in parenthesis) by its considerable smaller size (maximum size 120 mm SVL), snout shape spatulated, rounded in lateral view (not spatulated), absence of lateral fringes in toes (lateral fringes present), and the paired lateral vocal sacs (single subgular vocal sac). Likewise, *L. fragilis* can be distinguished from *Eleutherodactylus planirostris* (morphological characters of *E. planirostris* in parenthesis) by a considerably larger size (maximum size: 23.2 mm SVL) and the absence of digital ornamentation (lateral fringes present, digital discs slightly expanded and truncated, with pads and circumferential groove).

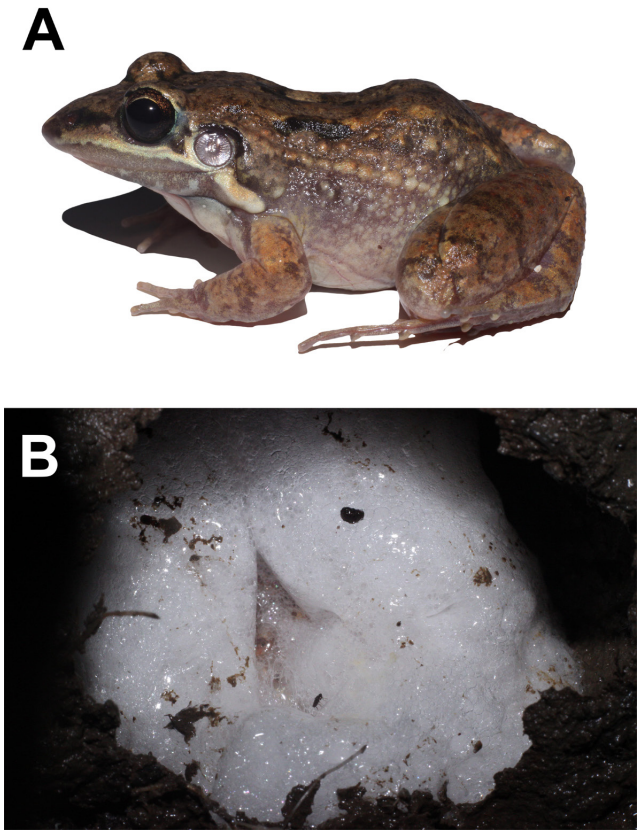

Figure 2. Adult specimen (A, CBUMAG: ANF: 1189) and foam nest (B) of *Leptodactylus fragilis* from Los Corales, San Andrés, Colombia.

The record of *L. fragilis* in San Andrés represents the third known anuran, the second invasive frog species, and the ninth non-native herp documented from the Colombian Caribbean islands (Cubillos-Abrahams *et al.* 2021). *Leptodactylus fragilis* is a very adaptable species with a high invasive potential in San Andrés (and other Caribbean islands, see Rodríguez-Cabrera *et al*. 2018), since as suggested by its high relative abundance (0.81 individuals/hours/observers, Cubillos-Abrahams & Montes-Correa, unpublished data). Given that this species can reproduce even in the driest season (*i.e.* using artificial water sources, the small remnants of seasonal water bodies, or the only one permanent pond), we assume that the species could have a continuous reproductive cycle throughout the year. The latter is rare in amphibians found in tropical dry forests in northern Colombia, which exhibit explosive breeding events mediated by the availability of water resources (Vargas-Salinas *et al*. 2019). The reproductive cycle of *L. fragilis* appears to be somewhere between a prolonged and explosive breeding pattern (*sensu* Wells 2007, Vargas-Salinas *et al*. 2019), because reproductive activity was extended throughout the study (from the driest season to the beginning of rainfalls), frogs densely gathered in reproductive habitats of variable stability (both temporary to permanent water bodies or humid shelters), and males vocalized permanently every night at all sampling sites.

The response of amphibians to water scarcity in tropical dry forest depends on a constant interaction of physiological, ecological, and behavioral strategies (Urbina-Cardona *et al*. 2014). Five ecological and behavioral strategies of tropical dry forest frogs from northern Colombia were described to tolerate desiccation in dry periods (Cuentas *et al*. 2002), two active strategies (horizontal and vertical movements towards microhabitats with optimal humidity) and three passive ones (staying in water bodies or humid shelters, and estivation). According to Cuentas *et al*. (2002), *L. fragilis* remains in the surroundings of water bodies, and they did not obtain records of the species during prolonged dry periods (which could suggest that there is a process of estivation). However, we found specimens of *L. fragilis* at the time of maximum drought densely grouped around permanent water bodies, in the wet mud of seasonal water bodies, and in humid areas not necessarily related to the presence of water bodies. Therefore, it seems that *L. fragilis* displays a wide range of ecological and behavioral responses to desiccation, which could be influenced by the variation of climatic conditions both in natural and non-native distribution area. Laying eggs in a foam nest is a *Leptodactylus* frog trait that improves tadpole survival against desiccation, a common threat in tropical dry forests, and even in South temperate areas (among other functions, see Gould 2021). Terrestrial and underground foam nests were previously considered as a synapomorphic character for the *Leptodactylus fuscus* species group, but recognition of the derived or primitive condition for the genus *Leptodactylus* depends on the discovering what kind of nesting prevails in the members of the genus *Hydrolaetare* (Heyer 1969, see de Sá *et al*. 2014). This reproductive mode can provide adaptive advantages compared to species (like *L. insularum*) that lay their eggs on the water surface (Gould 2021).

The invasive populations of *L. fragilis* in San Andrés show high abundance, ecological tolerance to multiple habitats, and reproduce successfully in all sites where it was recorded. For all the above, we consider that the invasive population of this species in San Andrés is in the "spread stage" (category D2) according to the framework for biological invasions proposed by Blackburn *et al.* (2011). This stage is assigned to self-sustaining non-native populations which survive and reproduce at a significant distance from the original point of introduction. In the non-native population of *L. fragilis* stablished in Cuba (Rodríguez-Cabrera *et al.* 2018), the invasion is at an intermediate stage (between establishment and dispersal stages *sensu* Blackburn *et al*. 2011). Although the species may be locally abundant, it has not dispersed enough to increase its extent of occurrence significantly. On the other hand, the extent of occurrence of *L. fragilis* in San Andrés (2.68 km^2) occupies 22.3% of the total area of the island, and it is likely that this is greater than documented here. Therefore, it is evident that the invasive potential of *L. fragilis* is greater in small islands.

Future research is critically need it to understand the ecological aspects of *L. fragilis* and how it shares its ecological niche with the native *L. insularum*, to determine if the apparent reproductive advantages and greater relative abundance of *L. fragilis* may threaten the survival of *L. insularum* in San Andrés. Similarly, it is important to assess if the invasion of *L. fragilis* has any effect on the acoustic niche of *L. insularum*, as estimated in Cuba with the endemic toad *Peltophryne empusa* Cope, 1862 (del Castillo-Domínguez *et al.* 2021). In the same way, genetic studies are required to determine the origin of this invasive *L. fragilis* population. Cuban population of *L. fragilis* is the product of a recent invasion from northern South America facilitated by trade between the countries of Cuba and Venezuela (Rodríguez-Cabrera *et al.* 2018). Since San Andrés is a free port and receives merchandise from multiple Central American countries, as well as continental Colombia, the populations of *L. fragilis* could come from almost anywhere in their natural distribution range.

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