Phyllomedusa 23(1):79–85, 2024

ISSN 1519-1397 (print) / ISSN 2316-9079 (online)
doi: http://dx.doi.org/10.11606/issn.2316-9079.v23i1p79-85

Short Communication

Maximum egg mass size of *Ambystoma altamirani* (Caudata: Ambystomatidae)

Elisa Reyes-Olivares,[1] Hublester Domínguez-Vega,[1] Armando Sunny,[2] and Yuriana Gómez-Ortiz[1]

[1] Universidad Intercultural del Estado de México, División de Desarrollo Sustentable. Libramiento Francisco Villa s/n, 50640 San Felipe del Progreso, Estado de Mexico, Mexico. E-mail: hublester.dvega@gmail.com.

[2] Universidad Autónoma del Estado de México, Centro de Investigación en Ciencias Biológicas Aplicadas. Instituto Literario 100, Colonia Centro, 50000 Toluca, Estado de Mexico, Mexico. E-mail: sunny.biologia@gmail.com.

Keywords: Brook axolotl, Eggs, Natural history, Oviposition, Stream salamander.

Palavras-chave: Axolote-de-riacho, História natural, Ovipostura, Ovos, Salamandra-de-riacho.

Salamanders of the genus *Ambystoma* Tschudi, 1838, commonly known in Mexico as axolotls or achoques, are a group of amphibians represented by 25 species distributed from southern Canada to central Mexico (Frost 2024). Axolotls have a robust, elongated body, a compressed tail, smooth skin, and three pairs of external gills (Canseco-Márquez and Gutiérrez-Mayén 2010, Stebbins and McGinnis 2018). These amphibians live in lentic and lotic aquatic ecosystems and hide among aquatic plants and within openings formed by rocks and walls. They breed in water and are polygamous (Feder and Lynch 1982, Stebbins and McGinnis 2018, Ávila-Akerberg *et al.* 2021).

Of the 25 species that comprise the genus *Ambystoma,* 11 are found in Mexico and 10 are endemic to this country (Everson *et al.* 2021, Balderas-Valdivia and González-Hernández 2024, Frost 2024). Axolotls can be divided into two groups depending on their habitat: those that live in lagoons and dams and those that live in high mountain streams (Casas-Andreu *et al.* 2003). *Ambystoma altamirani* Dugès, 1895 belongs to the latter group and was previously considered to be three species: *A. altamirani,* found in the Lagunas de Zempoala and the Sierra de las Cruces, in Morelos, Mexico City and Mexico state; *A. leorae* Brandon, 1989, found in the Sierra Nevada in Puebla and Mexico state; and *A. rivulare* Frost, 2004, found in the Sierra de Taxco in Guerrero and Sierra Chincua in Michoacan and Mexico State (Everson *et al.* 2021). It is one of the two river axolotls found in the Trans-Mexican Volcanic Belt, between 2720 and 3479 m a.s.l. (Lemos-Espinal *et al.* 1999, Woolrich-Piña *et al.* 2017, Lemos-Espinal and Smith 2020, Sánchez-Sánchez *et al.* 2022). Although the species is apparently widespread, its populations are restricted to small patches within its habitat. Populations of *A. altamirani* are surrounded by some of the most highly urbanized areas of central Mexico; thus, the species faces serious environmental problems due to anthropogenic pressure, even within protected areas (Heredia-Bobadilla and Sunny 2021).

Received 30 January 2024
Accepted 25 March 2024
Distributed June 2024

The relatively few studies of *A. altamirani* are in contrast to other species such as *Ambystoma mexicanum* (Shaw and Nodder, 1798). Some of these studies on *A. altamirani* include field observations describing the eggs, larvae morphology, and size (Campbell and Simmons 1962, Lemos-Espinal and Ballinger 1994), larval diet (Lemos-Espinal *et al.* 2015), behavior and physiology (Sánchez-Sánchez *et al.* 2022), health condition, parasites, and deformities (Barriga-Vallejo *et al.* 2015, Sánchez-Manjarrez *et al.* 2022, Hernández-Luría *et al.* 2023), relationships between coloration and use of the substrate (Villarreal-Hernández *et al.* 2020), analysis of genetic variability, population size and structure (Heredia-Bobadilla *et al.* 2016, 2017), and evaluations of the impact of some habitat characteristics and the presence of livestock on their abundance (Gómez-Franco *et al.* 2022).

Adult males of *A. altamirani* reach a snout–vent length (SVL) ranging from 77 ± 1.6 mm to 95 ± 6.03 mm, while females range in size from 76 ± 1.4 mm to 99 ± 5.6 mm (Lemos-Espinal *et al.* 2017, Sánchez-Manjarrez 2017). The dorsal coloration of this species is blackish to olive green, and the color is less intense on the sides. The tail and belly are blackish gray and yellow or black spots may occur on the back (Taylor 1940). Only about six records of the egg mass size of *A. altamirani* have been reported. The number of eggs varies between 1 and 463, which have been found in masses or isolated on different elements of the habitat, such as roots, vegetation, rocks, and pine needles (Brandon and Altig 1973, Bille 2009, Legorreta-Balbuena *et al.* 2014, Sunny *et al.* 2014, Monroy-Vilchis *et al.* 2015, Lemos-Espinal *et al.* 2016, 2017). Herein we report an egg mass of *A. altamirani* that represents the maximum size known to date. Characteristics of the oviposition site and hatching time are described.

On 21 March 2022, an egg mass of *A. altamirani* (previously considered *A. rivulare*) was observed in San Francisco Oxtotilpan, municipality of Temascaltepec, state of Mexico, (19.161723° N, 99.903279° W at 2633 m a.s.l.). The location is within two protected areas: the Nevado de Toluca Flora and Fauna Protection Area and the Natural Resources Protection Area in the catchment of the rivers Valle de Bravo, Malacatepec, Tilostoc and Temascaltepec (CONANP 2023; Figure 1). It is a high mountain ecosystem with a temperate subhumid to semi-cold climate composed of fir (*Abies* spp.), pines (*Pinus* spp.), oaks (*Quercus* spp.) and aile (*Alnus acuminata* Kunth) (CONANP 2014). Numerous watersheds provide aquatic habitats for a diversity of organisms such as axolotls.

To estimate the size of the egg mass, the number of eggs was estimated from a sample of 93 eggs, which accounted for about 10% of the total mass; thus, the egg mass was estimated to contain between 930 and 1000 eggs (Figure 2). The eggs were black and had diameters of approximately 6 mm. They were located at the edge of a stream in a single mass attached to rocks and roots. In addition, more than half of the egg mass was covered by floating aquatic vegetation (*Lemna minor* L.) and small pieces of bark. The stream depth was 16 cm and its width was 36 cm, water temperature was 14°C, and the bottom had a sandy–muddy substrate. After observation, the collected sample was deposited in the same place together with the rest of the mass and observed periodically. All eggs hatched between 336 and 360 hours.

The size of the egg mass of *A. altamirani* in this observation was more than twice as large as previously reported egg masses, where between 1 and 463 eggs were observed, either isolated or in a single mass (Brandon and Altig 1973, Bille 2009, Legorreta-Balbuena *et al.* 2014, Sunny *et al.* 2014, Lemos-Espinal *et al.* 2016, 2017). The number of eggs per mass varies greatly in *A. altamirani;* nonetheless, such variation is not surprising considering that this species belongs to the *A. tigrinum* complex, whose species show enormous variation in their life history due to proximal factors (temperature, predation, competition, etc.) and genetic factors (Collins 1981, Collins *et al.* 1993, Sorci *et al.* 1996,

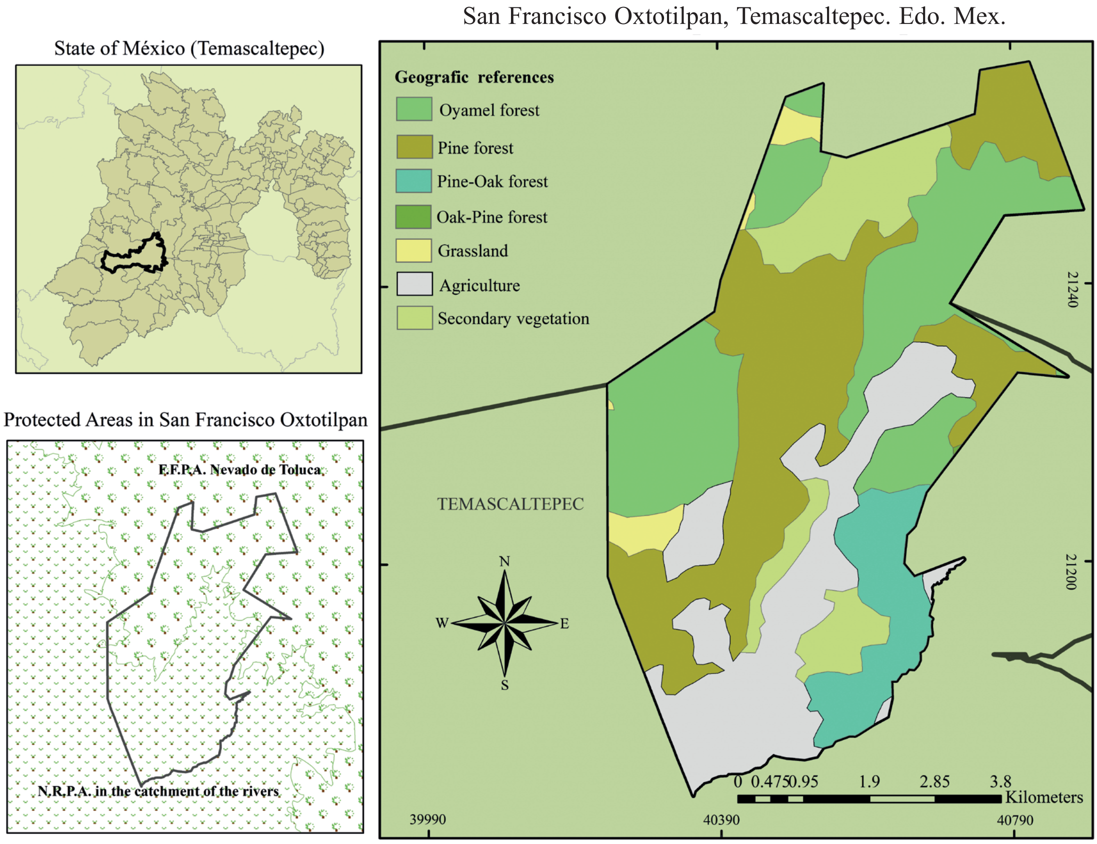

Figure 1. Map showing location of the egg mass of *Ambystoma altamirani*. The center of the image shows the location of San Francisco Oxtotilpan and the left side of the image shows its location within the Natural Areas of the zone.

Everson *et al.* 2021). Large egg masses such as that observed may be due to a proportional relationship between the size and age of the female and the number of eggs per mass (Parker and Begon 1986, Flemming 1994). Larger females and young females tend to lay a greater number of smaller eggs (Kuramoto 1975, Morrison and Héroe 2003).

The observed egg mass was attached to rocks and roots covered with bark and aquatic plants on a sandy–muddy bottom. Bille (2009) stated that *A. altamirani* seems to randomly select the places for egg deposition. For example, Brandon and Altig (1973) found an egg mass on roots and under a rock; Bille (2009) found numerous isolated eggs and egg masses on submerged pine branches on a rocky–muddy bottom, while Sunny *et al.* (2014) and Lemos-Espinal *et al.* (2016, 2017) found isolated eggs or egg masses on aquatic plants on a muddy bottom. Axolotls also appear to select sites that have structures that provide protection and sufficient support for egg deposition (Martínez 2007), such as vegetation, rocks, or roots. The sandy–muddy substrate in which the egg mass in this study was found coincides with other records of the characteristics of the habitat chosen by the species (SEMARNAT 2018).

Figure 2. (**A**) Egg mass of *Ambystoma altamirani* within a small stream. (**B**) Sample of 93 eggs from the clutch deposited by *A. altamirani.*

The depth of the stream in which the egg mass was located was 16 cm with a width of 36 cm and a velocity of 0.36 m/s. The velocity was estimated based on the time it takes for a 10-gram ball to travel a distance of one meter above the water surface of the stream. We used this technique because we did not have a flow meter at hand; thus, the measure must be taken with caution. The recorded depth is within the range of 10 cm to 57 ± 7.4 cm reported in other observations, while the width is close to the 40 and 69 ± 4.8 cm previously reported by other authors (Lemos-Espinal *et al.* 2016, 2017). The current velocity is also within the previously reported 0.3 to 0.4 m/s (SEMARNAT 2018), which is consistent with habitat characteristics of *A. altamirani,* which lives in small high mountain streams with low water flow.

The time from sighting to hatching of eggs in the field was between 336 and 360 hours at a temperature of 14°C with 100% of the eggs hatching, which is a longer period than that reported by Legorreta-Balbuena *et al.* (2014). Those authors reported that eggs in captivity hatched from 288 to 312 hours at a temperature of 18°C, with 86% of the eggs hatching. Temperature has been shown to strongly influence the growth and development of embryos and larvae (Brown 1976, Pepin 1991). In *Ambystoma* in general, eggs hatch relatively quickly at high temperatures but at a lower percentage, which is consistent with our observations and other literature reports (Light and Bogart 1989).

Our observations synthesize and add to what is known about some aspects of reproduction in *A. altamirani* in the central region of Mexico. We emphasize the need to conduct further research to gain a better understanding of the natural history of the species in the states of Guerrero, Michoacán, Morelos, Mexico City, and Puebla.

Acknowledgments.—We thank the local owners (ejidatarios) and community members of the town of San Francisco Oxtotilpan for their permission and willingness to allow us to conduct exploratory surveys in their community. SEMARNAT granted the collection permit SPARN/DGVS/06200/23. The Universidad Intercultural del Estado de México (UIEM) provided support and the Facultad de Estudios Avanzados de Zaragoza of the Universidad Autónoma de México placed the egg mass photograph in the digital collection of the Zoological Museum with the catalog number (MZFZ-IMG540). Finally, we thank the two anonymous reviewers and to Janalee Caldwell whose comments significantly improved this manuscript.

References

Ávila-Akerberg, V., T. González-Martínez, A. González-Hernández, and T. González-Martínez. 2021. El género Ambystoma en México ¿Qué son los ajolotes? *CIENCIA ergo-sum 28:* e127.

Balderas-Valdivia, C. J. and A. González-Hernández. 2024. Inventario de la Herpetofauna de México. *Herpetología Mexicana 6:* 13.

Barriga-Vallejo, C., O. Hernández-Gallegos, I. Hunt von Herbing, A. E. López-Moreno, M. L. Ruíz-Gómez, G. Granados-Gonzalez, M. V. Garduño-Paz, J. F. Méndez-Sánchez, J. Banda-Leal, and A. K. Davis. 2015. Assessing population health of the Toluca Axolotl *Ambystoma rivulare* (Taylor, 1940) from México, using leukocyte profiles. *Herpetological Conservation and Biology 10:* 592–601.

Bille, T. 2009. Field observations on the salamanders (Caudata; Ambystomatidae, Plethodontidae) of Nevado de Toluca, Mexico. *Salamandra 45:* 155–164.

Brandon, R. A. and R. G. Altig. 1973. Eggs and small larvae of two species of *Rhyacosiredon*. *Herpetologica 29:* 349–351.

Brown, H. A. 1976. The time-temperature relation of embryonic development in the northwestern salamander, *Ambystoma gracile. Canadian Journal of Zoology 54:* 552–558.

Canseco-Márquez, L. and M. G. Gutiérrez-Mayén. 2010. *Anfibios y Reptiles del Valle de Tehuacán-Cuicatlán.* Mexico City. Comisión Nacional para el Conocimiento y Uso de la Biodiversidad, Fundación para la Reserva de la Biosfera Cuicatlán, Benemérita Universidad Autónoma de Puebla. 302 p.

Campbell, H. W. and R. S. Simmons. 1962. Notes on the eggs and larvae of *Rhyacosiredon altamirani* (Duges). *Herpetologica 18:* 131–133.

Casas-Andreu, G., R. C. Aviña, and X. Aguilar-Miguel. 2003. Un regalo poco conocido de México al mundo: el ajolote o axolotl (*Ambystoma*: Caudata: Amphibia). Con algunas notas sobre la crítica situación de sus poblaciones. *CIENCIA ergo-sum 10:* 304–308.

Collins, J. P., 1981. Distribution, habitats and life history variation in the tiger salamander, *Ambystoma tigrinum,* in eastcentral and southeast Arizona. *Copeia 1981:* 666–675.

Collins, J. P., K. E. Zerba, and M. J. Sredl. 1993. Shaping intraspecific variation: development, ecology and the evolution of morphology and life history variation in tiger salamanders. *Genetica 89:* 167–183.

CONANP. 2023.Información Espacial de las Áreas Naturales Protegidas. CONANP. Eletronic Database accessible at http://sig.conanp.gob.mx/website/pagsig/info_shape.htm. Captured on 17 January 2024.

Everson, K. M., L. N. Gray, A. G. Jones, N. M. Lawrence, M. E. Foley, K. L. Sovacool, J. D. Kratovil, S. Hotaling, P.M Hime, A. Storfer, G. Parra-Olea, R. Percino-Daniel, X. Aguilar-Miguel, E. M. O'Neill, L. Zambrano, H. B. Shaffer, and D. W. Weisrock. 2021.Geography is more important than life history in the recent diversification of the tiger salamander complex. *Proceedings of the National Academy of Sciences 118:* e2014719118.

Feder, M. E. and J. F. Lynch. 1982. Effects of latitude, season, elevation, and microhabitat on field body temperatures of neotropical and temperate zone salamanders. *Ecology 63:* 1657–1664.

Flemming, A. F. 1994. Male and female reproductive cycles of the viviparous lizard, *Mabuya capensis* (Sauria: Scincidae) from South Africa. *Journal of Herpetology 28:* 334.

Frost, D. R. (ed.). 2024. Amphibian Species of the World: An Online Reference. Version 5.4 (March 2024). Electronic Database accessible at http: //research.amnh.org/vz/herpetology/amphibia/American Museum of Natural History, New York, USA. Captured on 10 March 2024.

Gómez Franco, W. G., G. R. Smith, and J. A. Lemos-Espinal. 2022. The effects of livestock, proximity to trees, and aquatic characteristics on the abundance of *Ambystoma altamirani* within a stream. *Journal of Herpetology 56:* 56–59.

Heredia-Bobadilla, R. L. and A. Sunny. 2021. Análisis de la categoría de riesgo de los ajolotes de arroyos de alta Montaña (Caudata: *Ambystoma*). *Acta Zoológica Mexicana (nueva serie) 37:* 1–19.

Heredia-Bobadilla, R.-L., O. Monroy-Vilchis, M. M. Zarco-González, D. Martínez-Gómez, G. D. Mendoza-Martínez, and A. Sunny. 2016. Genetic structure and diversity in an isolated population of an endemic mole salamander (*Ambystoma rivulare* Taylor, 1940) of central Mexico. *Genetica 144:* 689–698.

Heredia-Bobadilla, R.-L., O. Monroy-Vilchis, M. M. Zarco-González, D. Martínez-Gómez, G. D. Mendoza-Martínez, and A. Sunny. 2017. Genetic variability and structure of an isolated population of *Ambystoma altamirani,* a mole salamander that lives in the mountains of one of the largest urban areas in the world. *Journal of Genetics 96:* 873–883.

Hernández-Luría, J., O. Méndez-Méndez, R. Sánchez-Sánchez, G. R. Smith, and J. A. Lemos-Espinal. 2023. Observations of two invertebrate parasites on *Ambystoma altamirani* (Caudata: Ambystomatidae) from the Sierra de las Cruces, Mexico. *Phyllomedusa 22:* 37–42.

Kuramoto, M. 1975. Embryonic temperature adaptation in development rate of frogs. *Physiological Zoology 48:* 360–366.

Legorreta-Balbuena, G., G. Gutierrez-Ospina, I. V. Fierro, and G. Parra-Olea. 2014. *Ambystoma rivulare*. Reproduction. *Herpetological Review 45:* 107–108.

Lemos-Espinal, J. and R. E. Ballinger. 1994. *Rhyacosiredon leorae.* Size. *Herpetological Review 25:* 22.

Lemos-Espinal, J. A. and G. R. Smith. 2020. A conservation checklist of the amphibians and reptiles of the State of Mexico, Mexico with comparisons with adjoining states. *ZooKeys 953:* 137–159.

Lemos-Espinal, J. A., G. R. Smith, R. E. Ballinger and R. Ramírez-Bautista. 1999. Status of protected endemic salamanders (*Ambystoma*: Ambystomatidae: Caudata) in the Transvolcanic Belt of México. *British Herpetological Society Bulletin 68:* 1–4.

Lemos-Espinal, J. A., G. R. Smith, and G. A. Woolrich-Piña. 2015. Diet of larval *Ambystoma altamirani* from Llano de los Axolotes, Mexico. *Current Herpetology 34:* 75–79.

Lemos-Espinal, J. A., G. R. Smith, Á. H. Ruíz and R. M. Ayala. 2016. Stream use and population characteristics of the endangered salamander, *Ambystoma altamirani,* from the Arroyo Los Axolotes, state of Mexico, Mexico. *Southwestern Naturalist 61:* 28–32.

Lemos-Espinal, J. A., G. R. Smith, A. B. E. Zamora, G. Woolrich-Piña, and R. M. Ayala. 2017. Natural history of the critically endangered salamander *Ambystoma leorae* (Caudata: Ambystomatidae) from the Río Tonatzin, Mexico. *Phyllomedusa 16:* 3–11.

Light, L. E. and J. P. Bogart. 1989. Embryonic development and temperature tolerance in diploid and polyploid salamanders (genus *Ambystoma*). *American Midland Naturalist 122:* 401–407.

Martínez, A. I. M. 2007. Preferencia de plantas para la oviposición del axolote *Ambystoma mexicanum* en condiciones de laboratorio. Unpublished Undergraduate Thesis. Universidad Autónoma de México, Mexico.

Monroy-Vilchis, O., M. M. Zarco-González, H. Domínguez-Vega, and A. Sunny. 2015. *Ambystoma leorae* (Taylor, 1943). New records, natural history notes and threat status. *Herpetozoa 27:*166–168.

Morrison, C. and J.-M. Hero. 2003. Altitudinal variation in growth and development rates of tadpoles of *Litoria chloris* and *Litoria pearsoniana* in southeast Queensland, Australia. *Journal of Herpetology 37:* 59–64.

Parker, G. A. and M. Begon. 1986. Optimal egg size and clutch size: effects of environment and maternal phenotype. *American Naturalist 128:* 573–592.

Pepin, P. 1991. Effect of temperature and size on development, mortality, and survival rates of the pelagic early life history stages of marine fish. *Canadian Journal of Fisheries and Aquatic Sciences 48:* 503–518.

Sánchez-Manjarrez, D. 2017. Dimorfismo sexual en *Ambystoma rivulare* (Caudata: Ambystomatidae). Unpublished Undergraduate Thesis. Universidad Autónoma del Estado de México, Mexico.

Sánchez-Manjarrez, D., J. F. Méndez-Sánchez, J. R. Flores-Santín, J. L. Rheubert, and O. Hernández-Gallegos. 2022. Limb deformities in *Ambystoma rivulare* (Caudata: Ambystomatidae), a microendemic and threatened Mexican Salamander. *Herpetological Conservation and Biology 17:* 442–450.

Sánchez-Sánchez, R., O. Méndez-Méndez, G. R. Smith, R. Montoya- Ayala, G. Woolrich-Piña, and J. A. Lemos-Espinal. 2022. Field observations of *Ambystoma altamirani* at near-freezing conditions in the Sierra de las Cruces, Mexico. *Phyllomedusa 21:* 67–69.

SEMARNAT. 2018. *Programa de Acción para la Conservación de las Especies* Ambystoma *spp.* First Edition. Mexico. SEMARNAT/CONANP. 78 pp.

Sorci, G., J. Clobert, and S. Belichon. 1996. Phenotypic plasticity of growth and survival in the common lizard *Lacerta vivipara. Journal of Animal Ecology 65:* 781.

Stebbins, R. C. and S. M. McGinnis. 2018. *Peterson Field Guide to Western Reptiles Amphibians.* Fourth Edition. Boston and Massachusetts. Houghton Mifflin. 576 pp.

Sunny, A., O. Monroy-Vilchis, C. Reyna-Valencia, and M. M. Zarco-González. 2014. Microhabitat types promote the genetic structure of a micro-endemic and critically endangered mole salamander (*Ambystoma leorae*) of central Mexico. *PLoS ONE 9:* e103595.

Taylor, E. H. 1940. A new *Rhyacosiredon* (Caudata) from western México. *Herpetologica 1:* 171–176.

Villarreal-Hernández, V., G. R. Smith, R. M. Ayala, and J. A. Lemos-Espinal. 2020. The relationship between body and substrate color for *Ambystoma altamirani* (Caudata: Ambystomatidae) from the Arroyo los Axolotes, Mexico. *Phyllomedusa 19:* 243–251.

Woolrich-Piña, G., G. R. Smith, J. A. Lemos-Espinal, A. E. Zamora, and R. M. Ayala. 2017. Observed localities for three endangered, endemic Mexican ambystomatids (*Ambystoma altamirani, A. leorae,* and *A. rivulare*) from central Mexico. *Herpetological Bulletin 139:* 12–15.

Editor: Jaime Bertoluci