**Liolaemus lonquimayensis** (Squamata: Liolaemidae), a new lizard species for Chile without precloacal pores

**Liolaemus lonquimayensis** (Squamata: Liolaemidae), una nueva especie de lagartija para Chile, sin poros precoacales

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**ABSTRACT**

This study describes a new species of lizard of the genus *Liolaemus* (*L. lonquimayensis*), without precloacal pores, associated with the lava flows of the Lonquimay volcano in the Región de la Araucanía, Chile. This species belongs to the *elongatus* clade, and is a sister lineage of *L. elongatus*. *L. lonquimayensis* differs from its sister species, *L. elongatus*, in the total absence of precloacal pores, smaller maximum SVL (69.7 mm in *L. lonquimayensis* and 91.0 mm in *L. elongatus*), greater number of midline scales (88 in *L. lonquimayensis* and 77 in *L. elongatus*), body color light grey to black in *L. lonquimayensis* and light to dark brown in *L. elongatus*. *L. lonquimayensis* has an black occipital band formed by small black lines arranged horizontally flanked by two lighter dorsal longitudinal bands, more separated towards the tail, forming incomplete rings. Finally, a combination of genetic, meristic and morphological characters distinguishes *L. lonquimayensis* from the other species of the *elongatus* clade.

**KEYWORDS**: *Elongatus* clade, *Liolaemus* nov. sp, Lonquimay volcano, taxonomy.

**INTRODUCTION**

Species are habitually used as the units of analysis in biogeography, ecology, macroevolution and biological conservation (Sites and Marshall, 2004). The current delimitation of species uses an integrative approach with multiple complementary methods, especially in taxonomic groups with complex evolutionary histories (Torres-Pérez et al. 2009; Padial et al. 2010). Mitochondrial DNA (mtDNA) has proven to be very useful to define species in vertebrate groups (Avise 2004). However, methods based on traditional morphology continue to be used, providing multiple advantages when molecular methods cannot be applied or provide insufficient evidence (Hillis & Wiens 2000, Wiens 2004).

The genus *Liolaemus* is the second most diversified group of lizards in the planet after *Anolis*, with 244 described species (Uetz & Hošek 2014). Its distribution is very wide, including Argentina, Bolivia, Brazil, Chile, Paraguay, Peru and Uruguay, from sea level to high-altitude Andean environments (Pincheira-Donoso et al. 2008). It is precisely
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the diversity of environments that this genus inhabits that has modeled its adaptive radiation; it includes interesting variations in development, genetic and morphological patterns as well as in its rate of molecular evolution (Torres-Pérez et al. 2009).

The majority of the species of the genus *Liolaemus* produce pheromones involved in intraspecific communication by means of structures called precloacal pores, present mainly in males (Escobar et al. 2001; Labra et al. 2002). However, within the genus *Liolaemus* there are some groups that have species without these structures (the *capillitas*, chillanensis, elongatus, kriegi and pictus* groups) (Abdala et al. 2010, Lobo et al. 2010, Esquerré et al. 2013). In this study we describe a new species of the genus *Liolaemus* without precloacal pores, associated with the lava flows of Volcán Lonquimay.

**MATERIALS AND METHODS**

Four specimens were collected by hand, sacrificed by a pericardiac injection of sodium pentothal in the lava flows of the Volcán Lonquimay in the locality of Malalcahuello (34°23′S, 71°37′W), between the communities of Curacautín and Lonquimay (Fig. 1). Each individual was measured with a digital caliper to the nearest 0.1 mm, the sex was determined. We also examined the scale pattern ans scale terminology following Ortiz (1981). Description of color in life are based on noted taken in the field and color photographs. All specimens were deposited in the herpetological collection of the Museo de Zoología de la Universidad de Concepción (MZUC). We obtained a sample of liver tissue for molecular study and fixed in 95% ethanol using the Wizard SV Genomic extraction kit (Promega). Protocols for DNA extraction mtDNA cytochrome b primer description PCR, and sequencing procedures follow Victoriano et al. (2008). PCR products were purified and sequenced at Macrogen, Inc. (www.macrogen.com). The sequences obtained were edited in BIOEDIT v.7.0.9.0 (Hall 1999) and aligned in CLUSTAL X v.2.0 (Larkin et al. 2007). See Appendix I for specimens used for molecular analyses. The phylogenetic relations between the studied species were estimated by Bayesian inference with the program MRBAYES v3.1.2 (Huelsenbeck and Ronquist 2001), based on the evolutionary model TIM2+I+G previously selected by the JMODELTEST v0.1.1 program (Posada 2008). We used the Markov Chain Monte Carlo (MCMC) method to assure that trees were sampled proportionally to their probability of occurrence under the chosen evolutionary model. The analysis was performed using two independent runs of $2 \times 10^7$ generations each, sampling each chain every 1000 generations. The first 25% of the 20000 trees was discarded because the zone of stationary convergence had not been reached. The remaining trees were used to produce a consensus tree by the majority rule, which was graphed in the program MEGA v4.0 (Tamura et al. 2007). We used an *a posteriori* of probability of $P \geq 0.95$ as evidence of significant support for a given clade (Huelsenbeck & Ronquist 2001).

![FIGURE 1. Site where *Liolaemus lonquimayensis* was collected. Volcán Lonquimay, Reserva Nacional Malalcahuello-Nalcas, between the Comunities of Curacautín and Lonquimay.](image1)

**FIGURE 1.** Site where *Liolaemus lonquimayensis* was collected. Volcán Lonquimay, Reserva Nacional Malalcahuello-Nalcas, between the Comunities of Curacautín and Lonquimay.

**FIGURA 1.** Sitio donde fue recolectado *Liolaemus lonquimayensis*. Volcán Lonquimay, Reserva Nacional Malalcahuello-Nalcas, entre las comunas de Curacautín y Lonquimay.
RESULTS

Liolaemus longimayensis sp. nov.

Type material. Holotype: MZUC-40365, adult male collected on the northern slopes of Volcán Lonquimay (38°23′16″S/71°37′42″W; elevation 1777 m) on 6 February 2013 by G. Escobar, J. Santibáñez, X. Fuentealba and F. Escobar. Allotype: MZUC-40366, adult female. Same collection data as the holotype. Paratypes: MZUC-40367, adult female; MZUC-40368 juvenile female. Same collection data as the holotype (Fig. 3).

Diagnosis. Liolaemus longimayensis sp. nov. is a member of the elongatus group, characterized by moderate body size; body elongated but robust. Scales small, subimbricate and not ending in a spine. Tail ringed, tending to circular in transverse section, length reaching 1.5 times SVL. Absence of sexual dimorphism and dichromatism. Wide dark band in the inferior flanks from the axilla to the ingle, absent or similar to the wide vertebral band. Abdominal melanism infrequent (some species have ventral scales of a tenuous gray color). Gular folds absent (some species have hemigular folds). The species is found in riparian and saxicolous habitats (Espinoza et al. 2000; Cei and Videla 2003). L. longimayensis and L. thermae is distinguished from almost all species of the elongatus clade by the total absence of precloacal pores in males. (i. e., L. chillanensis, and L. elongatus).

Description of holotype. Adult male 69.74 mm (SVL), tail 111.6 mm. Axila-groin distances 33.0 mm. Head slightly longer (16.41 mm) than wide (12.33 mm); height 8.38 mm. Mouth width 11.11 mm. Length of anterior extremity, Meatus ovalate, height 3.19 mm. Transverse neck folds slightly wider than the head. Anterior extremities extended forward reach the tip of the mouth; length 23.23 mm. Posterior extremities extended forward reach the border of the meatus; length 43.02 mm. Foot length 20.6 mm. Base of the tail very widened; length 1.60 times the SVL. Precloacal pores absent.

Rostral scale hexagonal, 2.86 times taller than wide; in contact with six scales that include the anterior internasal scales. Small quadrangular scale between anterior and posterior internasal scales. Nasal scales hexagonal; nostrils located in the posterior part of both scales. 3-3 prenasal scales; 4-5 postnasal scales. 2-2 cantal scales separate 6-7 loreal scales from contact with the superior shields of the head. Preocular scale pentagonal, in its posterior part with an elongated subocular scale and 6-6 supralabial scales. 8-8 scales between last supralabial scale and inferior margin of

Figure 2. Liolaemus longimayensis, paratype adult female (MZUC-40367).

Figura 2. Liolaemus longimayensis, paratipo hembra adulta (MZUC-40367).
meatus. 6-6 small ear scales. 14-14 temporal scales. 8-8 supraciliar scales compressed dorsoventrally. 7-7 larger supraocular scales and 29-29 smaller, surrounded by 14-14 circumorbital scales. Interparietal scale hexagonal, elongated towards the posterior extreme of the head and with a prominent pineal eye in the middle of this scale. Two large parietal scales behind the interparietal, separated posteriorly by a small occipital scale. Two pairs of frontoparietal scales, anterior more elongated than the posterior. Two frontal scales (anterior divided longitudinally), followed by two quadrangular prefrontal scales, 16 cephalic shields, three major shields in front of the prefrontals and in contact with four scales (1-1 infralabial and 1-1 postmental); 6-6 infralabial scales. 5-5 prefrontal scales. First pair of postmental scales large, separated posteriorly by two gular scales. Gular scales rounded, smooth and imbricated, more elongated towards the mental scale. Transverse folds of the neck and axillar region have small granular scales. Dorsal and ventral scales almost the same size; however, dorsal scales are subtriangular, keeled and imbricate while ventral scales are rounded and smooth. Keels less prominent toward the sides of the body, disappearing in the flanks. 80 scales from the parietals to the beginning of the tail and 84 in body midline. Dorsal scales of forelimb triangular, smooth and imbricate; slightly keeled in the forearm. Dorsum of the hand with rounded, smooth and imbricate scales. Ventral forearm scales rounded in the anterior zone and granular in the posterior zone, while forearm scales are subtriangular and imbricate. Fourth digit of hand with 24-24 transversal lamellae with three keels in each. Dorsal scales of femoral and tibial region subtriangular, slightly keeled and imbricate. Dorsal scales of the foot subtriangular, smooth and imbricate. Ventral scales of the femoral region subtriangular in the longitudinal anterior zone, rounded in the midzone and granular in the posterior zone. Ventral scales of the tibia subtriangular, smooth and imbricate. Palm of the foot with subtriangular, slightly keeled and imbricate scales. Fourth toe of hind foot with 30-30 transverse lamellae with three keels in each. Dorsal scales of tail triangular, keeled and imbricate.

Design and color. Dorsal background color light gray. Juveniles and adults with a black occipital band formed by small black lines arranged horizontally, more separated towards the tail, forming incomplete rings. Parietal and flank bands of the background color. Black horizontal lines over the temporal bands almost touch the horizontal lines of the occipital band. Dorsal zone of extremities with black vertical lines and light gray points. Ventral color lighter than dorsum, with random small black blotches. Adult females
have a black abdominal blotch. Sexual dichromatism absent. Variation in paratypes. Size variation is given in Table 1. In meristic characters females differ from males (holotype) in: absence of quadrangular scale between anterior and posterior internasal scales. 2/3 prenasal scales, 3/4 posnasal scales. 4/6 loreal scales. 1/2 postocular scales. 5/7 supralabial scales. 6/8 scales between last supralabial and inferior of meatus. 4/5 small preauricular scales. 9/10 temporal scales. Seven supraciliar scales. Six large supraocular scales, 20/25 small supraocular scales and 11/12 circumbital scales. 5/6 infralabial scales. 89-90 scales around midline. 25-27 lamellae in fourth digit of hand and 31/34 lamellae in fourth digit of foot.

**TABLE 1. Morphometric variation in the type series.** Characters: (SVL) snout-vent length, (AIL) axilla-ingle length, (HL) head length, (BL) mouth length, (MHW) maximum head width, (HH) head height, (LAE) length of anterior extremity, (LPE) length of posterior extremity (FL) foot length, (LT) tail length. The dash indicates that the tail was amputated.

| SEXO/STATE | MZUC 40365 | MZUC 40366 | MZUC 40367 | MZUC 40368 |
|-----------|------------|------------|------------|------------|
| SVL       | 69.7       | 69.4       | 68.3       | 45.5       |
| AIL       | 33.0       | 35.7       | 36.1       | 20.8       |
| HL        | 16.4       | 14.6       | 14.9       | 10.6       |
| BL        | 11.1       | 9.5        | 10.0       | 6.8        |
| MHW       | 12.3       | 10.7       | 11.0       | 7.5        |
| HH        | 8.3        | 8.2        | 8.4        | 5.1        |
| LAE       | 23.2       | 22.5       | 22.9       | 16.5       |
| LPE       | 43.0       | 40.7       | 40.1       | 29.4       |
| FL        | 20.6       | 20.0       | 20.4       | 14.6       |
| LT        | 111.6      | 92.2       | −          | 77.2       |

**FIGURE 4. General view of the type locality of Liolaemus lonquimayensis.**

**FIGURA 4. Vista general de la localidad tipo de Liolaemus lonquimayensis.**
Etymology. The specific name *lonquimayensis* refers to the Lonquimay volcano, the type locality of this Andean species.

Distribution. *Liolaemus lonquimayensis* is only known from the type locality

Natural history. In its type locality, *L. lonquimayensis* is sympatric and synoptic with *L. cristiani*. Its habitat is flows of volcanic lava, with sparse vegetation and abundant basalt rocks (Fig. 4). Females are viviparous, giving birth to about two live offspring

The phylogenetic analyses used a matrix of 521 pb per sequence. Of these, 204 bases were variable and 171 were parsimony-informative. The consensus tree obtained located *Liolaemus lonquimayensis* within the *elongatus* clade. This species is a sister lineage to *L. elongatus*, with high a posteriori probability (PP = 1.00) (Fig. 2). The genetic distance between these species was 2.9%.

**DISCUSSION**

*L. lonquimayensis* differs from its sister species, *L. elongatus*, in the total absence of precloacal pores, smaller maximum SVL (69.7 mm in *L. lonquimayensis* and 91.0 mm in *L. elongatus*), greater number of midline scales (88 in *L. lonquimayensis* and 77 in *L. elongatus*), body color light grey to black in *L. lonquimayensis* and light to dark brown in *L. elongatus*. Also, *L. lonquimayensis* has well-differentiated rings in the tail, which are not clearly distinguished in *L. elongatus*. *L. chillanensis* is differentiated from *L. lonquimayensis* because the males of the former have precloacal pores. Its dorsal pattern is characterized by a wide vertical band with small longitudinal black lines, while the dorsal design of *L. lonquimayensis* is composed of irregularly fused horizontal lines flanked by two lighter dorsal longitudinal bands. Finally, although *L. thermarum* does not have precloacal pores like *L. lonquimayensis*, it is differentiated from the latter species by a greater maximum SVL (85 mm), a lower number of mid-line scales (mean 73), a vertebral band formed by small and irregular black spots that are sometimes fused, forming a solid black vertebral band, and does not have rings on the tail as *L. elongatus* and *L. chillanensis* have.

Evolutionary relations in the *elongatus* clade have been widely discussed since its description. Lobo et al. (2010) assigned the following species to the *elongatus* clade: *L. austromendocinus*, *L. elongatus*, *L. flavipiceus*, *L. gununakuna*, *L. parvus*, *L. petrophilus*, *L. punmahuida*, *L. thermarum* and *L. tregenzai*. They also included in the clade the “capillitas clade”, composed of *L. capillitas*, *L. dicktracyi*, *L. heliodermis*, *L. talampaya*, *L. tulkas* and *L. umbrifer*. However, the systematic proposal of these authors was not supported by a phylogenetic hypothesis. The “capillitas group”, along with the species *L. austromendocinus*, *L. gununakuna*, *L. parvus* and *L.
petrophilus are recovered within the petrophilus clade and not the elongatus clade, as was indicated by Morando et al. (2003), Avila et al. (2004) and Quinteros et al. (2008). We recovered the kriegi clade (L. buergeri + L. kriegi) as the sister lineage of the elongatus clade (Fig. 5). However, L. cristiani does not form part of this clade as Lobo et al. (2010) proposed; L. cristiani groups with L. villaricensis, another species without precloacal pores. For this reason, the chillanensis group (L. chillanensis + L. villaricensis) observed by Lobo (2005) and formally proposed by Lobo et al. (2010) is rejected. L. chillanensis belongs to the elongatus group (Torres-Pérez et al. 2009, Avila et al. 2010), and L. cristiani groups with L. villaricensis. Finally, it must be noted that the species without precloacal pores do not form a monophyletic group, rejecting the “neuquensis group” (L. coerules, L. cristiani, L. neuquensis and L. thermarum) of Cej & Videla (2003).

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**APPENDIX I. Specimens used for phylogenetic analyses**

*L. austromendocinus* LJAMM 2715, *L. buergeri*, LJAMM 2682, *L. capillitas* BYU 47100, *L. chillanensis* MZUC 28252, *L. cristiani* MZUC 40369, *L. cyanogaster* CPV 209, *L. dicktracyi* LJAMM 5750, *L. elongatus* BYU 47101, BYU 47092, MVZ 232399, *L. flavipiceus* LJAMM-CNP 7906, *L. gununakana* LJAMM 2440, *L. kriegi* MVZ 232273, *L. longquimayensis* MZUC 40365-40368, *L. neuquensis* FML 9465, *L. parvus* LJAMM-CNP 2711, *L. petrophilus* BYU 47097, *L. pictus* B17LPCPO58, *L. punmahuida* LJAMM-CNP 2626, *L. septentrionalis* E11VTLP 46, *L. talampaya*, LJAMM 2737, *L. theramurum* LJAMM-CNP 5792, *L. tulkas* LJAMM-CNP 4227, *L. umbrifer* LJAMM 5029 and *L. villaricensis* MZUC 28232.

Outgroup: *Phymaturus indistinctus* LJAMM 2124

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