

REVALIDATION OF *LATRODECTUS THORACICUS* NICOLET, 1849
(ARANEAE: THERIDIIDAE): BIOLOGICAL AND PHYLOGENETIC
ANTECEDENTS

*REVALIDACION DE LATRODECTUS THORACICUS NICOLET,
1849 (ARANEAE: THERIDIIDAE): ANTECEDENTES BIOLOGICOS Y
FILOGENETICOS*

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ABSTRACT

The number and identity of the species of black widow spiders of the genus *Latrodectus* that inhabit Chile remain unclear. Here we present results of taxonomic work based on morphologic and molecular characters of a large series of specimens collected throughout Chile, which aimed to assess Chilean *Latrodectus* diversity. Results show that all studied populations belong to a single species, for which the correct name is *Latrodectus thoracicus* Nicolet 1849. Therefore, we remove this taxon from the synonymy of *L. mactans* (Fabricius, 1775). In addition, we suggest that *L. mirabilis* (Holmberg, 1876) may be junior synonym of *L. thoracicus*. We recognize two species of black widows for continental Chile, *L. thoracicus* and *L. variegatus* Nicolet, 1849. We provided an emended diagnosis and comprehensive description of *L. thoracicus* as well as new locality records and novel data on its natural history. Given our results, we recommend the continued collecting specimens and more collection based taxonomic work as the only way of gathering a correct appreciation of *Latrodectus* diversity in southern South America.

KEYWORDS: Chile, species limits, taxonomy, Black widows.

RESUMEN

El número e identidad de las especies de viudas negras del género *Latrodectus* que habitan Chile no es claro. En este trabajo presentamos resultados de un estudio taxonómico basado en caracteres morfológicos y moleculares de una serie grande de especímenes colectados a lo largo de Chile, cuyo objetivo fue evaluar la diversidad de *Latrodectus* en Chile. Los resultados indican que las poblaciones estudiadas pertenecen a una misma especie, para la cual el nombre correcto es *Latrodectus thoracicus* Nicolet 1849. Por lo tanto removimos dicho taxón de la sinonimia de *L. mactans* (Fabricius, 1775). Además, sugerimos que *L. mirabilis* (Holmberg, 1876) puede constituir un sinónimo junior de *L. thoracicus*. Reconocemos dos especies de viudas negras para Chile continental, *thoracicus* y *L. variegatus* Nicolet, 1849. Proveemos una diagnosis enmendada y una descripción detallada de *L. thoracicus* así como nuevas localidades de registro y datos sobre su historia natural. A la luz de nuestros resultados, afirmamos que es imperioso continuar realizando colecta de especímenes y trabajo taxonómico basado en colecciones como la única forma de obtener una correcta apreciación de la diversidad de *Latrodectus* en el sur de América del Sur.

PALABRAS CLAVE: Chile, límites de especies, taxonomía, viudas negras.

INTRODUCTION

Latrodectus Walckenaer, 1805 is a diverse and cosmopolitan genus of theridiid spiders with 30 recognized species (Platnick 2009). It is considered a taxonomically complex genus as the status of several forms have not been properly evaluated and specific boundaries are not well defined or understood (Garb *et al.* 2004; Levi 1959, 1967a); therefore, in multiple cases, populations have been uncritically referred to as different taxa.

The members of this genus are of medical importance due to the potency of their venom, which contains a neurotoxin that produces a massive liberation of neurotransmitters. Among the proteins that cause the latrodectism one of the most important is the α -latrotoxin (Ori & Ikeda 1998; Grishin 1999; Orlova *et al.* 2000). Biochemical studies have been done to identify the proteins present in some Chilean *Latrodectus* specimens. The results have been coincident with those based on North American and European species (Romero *et al.* 1996, 2000). Medical and related literature has associated the Chilean cases of latrodectism with *Latrodectus mactans* (Fabricius, 1775) (Artaza *et al.* 1982, Artaza *et al.* 1984; Schenone & Correa 1985; Romero *et al.* 2003; Parodi *et al.* 2008) or with *L. curacaviensis* (Muller, 1776) (Faúndez 2009). However, the number and identity of the species of *Latrodectus* inhabiting Chile, is for the moment not clear (Canals *et al.* 2004); therefore it is not possible to determine which species of *Latrodectus* cause latrodectism in Chile.

The genus *Latrodectus* has a wide distribution in Chile, ranging from the deserts of the north (20°S) to the Patagonian steppes near the Magellan Strait (approximately 50°S). The number and identity of the species of *Latrodectus* currently inhabiting Chile are not well known. This is due to the complex taxonomic history of the diverse forms of *Latrodectus*, and to the lack of studies that have included Chilean specimens. The first records of *Latrodectus* in Chile were provided by Nicolet (1849), who described *Latrodectus variegatus* Nicolet 1849 and *Latrodectus thoracicus* Nicolet, 1849 with Isla Grande de Chiloé and Concepción as the type localities, respectively. At the same time, Nicolet (1849) mentioned a third form, *Latrodectus formidabilis* Walckenaer, 1837, from Chile; this taxon was later considered as a synonym of *L. mactans* by Gerschman & Schiapelli (1943)

and of *L. curacaviensis* by Levi (1959). However, Platnick (2009) points out that *L. mactans* is not present in Chile and lists three *Latrodectus* species for Chile: 1) *Latrodectus geometricus* C. L. Koch, 1841, with a worldwide distribution; 2) *Latrodectus curacaviensis* (Müller, 1776) found in the Antillas and South America, with the exception of Argentina; and 3) *L. variegatus* Nicolet, 1849 with a restricted distribution in Chile and adjacent zones of Argentinean and Chilean Patagonia (Nicolet 1849; Levi 1959, 1967a, 1967b; Abalos 1980). It should be noted that no revisionary work has included more than one Chilean specimen of *Latrodectus*. Levi (1959) included one specimen identified as *L. curacaviensis*. The phylogenetic analysis of Garb *et al.* (2004) included a single Chilean specimen that was not identified at the species level and for which no information on its geographic origin was provided.

The present study is based on the analysis of morphological and molecular characters, and has the aim of clarifying the taxonomic identity and the distribution of the species of black widow present in continental Chile.

MATERIALS AND METHODS

MATERIAL

We studied a total of 229 females, 8 males and 98 immature specimens from three Chilean collections: Museo de Zoología de la Universidad de Concepción (MZUC-UCCC), Museo Nacional de Historia Natural (MNHN), and Laboratorio de Entomología y Ecología, Universidad de La Serena (LEULS), and the African National Museum Bloemfontein (NMB). Additional spiders were collected by the authors using a manual technique (Upton 1991; Sorensen *et al.* 2002), from 33 localities from Arica and Parinacota Region in northern Chile to the Magallanes and Antártica Chilena Region in southern Chile; these specimens were deposited in the MZUC-UCCC.

MORPHOLOGICAL ANALYSIS

Morphological descriptions follow those of Lotz (1994) and Abalos (1980). Measurements in the descriptions are in millimetres. The following two coefficients were calculated: cephalothoracic coefficient (T) = cephalothorax length (without chelicerae) divided by cephalothorax wide; TT

coefficient (TT) = sum of the length of the patella and tibia on leg I divided by cephalothorax length (without chelicerae).

PHYLOGENETIC ANALYSIS

Design and sampling. Phylogenetic and genetic analyses were based on a fragment of the mitochondrial genome corresponding to the cytochrome oxidase subunit I (COI) gene. Analyses included sequences of nine Chilean specimens; eight of these were collected by us at the following two localities: 1) Casas Viejas, Comuna San Clemente, Región del Maule (35°27'40,2" S 71°29'16,8" W) (n=7) and 2) Lomas Los Huasos, Andacollo, Región de Coquimbo (30°13'49" S 71°5'9" W) (n=1)); the ninth Chilean haplotype, which lacks species determination and specific locality, was taken from GenBank (AY383074; Garb *et al.* 2004). The remaining sequences were downloaded from GenBank. Thus, our sampling includes 30 sequences of *Latrodectus* from 13 species (Table 1). As the monophyly of *Latrodectus* is well supported (Garb *et al.*, 2004) the outgroup was constituted by a single representative of the false widow spiders theridiid genus *Steatoda*.

Sequence acquisition. DNA was extracted using a commercial (Promega) kit from one or two legs. The rest of the specimens were preserved as vouchers. A fragment of the COI gene was amplified with primers LCOI 1498 and HCO 2198 (Folmer *et al.* 1994). Amplifications were done in three stages: 1) initial denaturation at 94 °C for 90 seconds (s); 2) 35 cycles of denaturation at 94 °C for 30 s, annealing at 55 °C for 40 s, and extension at 72 °C for 45 s; and 3) final extension at 72 °C for 10 min. Amplicons were sent to Macrogen Inc. Korea, for purification and sequencing. Obtained sequences were submitted to GenBank (accession numbers GU112098-105).

Sequence analysis. Sequence alignment was done with Clustal X (Thompson *et al.* 1997) using the default values for all alignment parameters. Observed percentage of sequence divergence was calculated with MEGA 4 (Tamura *et al.* 2007) ignoring those sites with missing data. Gene trees were inferred by Maximum Parsimony (Farris, 1983) using PAUP* (Swofford, 2000) with characters treated as unordered and with no weights. The search strategy consisted of 200 replicates of heuristic search with random addition of sequences and TBR branch swapping. Support for the nodes was

assessed by a Jackknife (JK) analysis consisting of 1000 pseudo-replicates with 2 replicates of random addition of sequence, TBR branch swapping, and 33% of character deletion. Given that an haplotype was shared among newly sequenced specimens, redundant sequences were excluded from the analysis (see Fig. 1).

RESULTS

Of 646 characters, 186 were variable and 143 were parsimony-informative. The phylogenetic analysis recovered 442 equally parsimonious trees (length = 458 steps; CI = 0.555; RI = 0.627), the consensus of which (Fig. 1) is moderately resolved. As in Garb *et al.* (2004) the basal dichotomy of the *Latrodectus* clade was the *geometricus* group (JK = 93), here represented by *L. geometricus* and *L. rhodesiensis*, and the *mactans* group (JK = 91), which contained the rest of the *Latrodectus* species included in our study. There was a well supported clade (JK = 91) that contained Chilean haplotypes gathered by us, haplotypes gathered by Garb *et al.* (2004) from an unidentified Chilean specimen, other specimens identified as *L. variegatus* from the Argentinean provinces of Río Negro and Santa Cruz, and a haplotype recovered from a specimen identified as of *L. mirabilis* from Buenos Aires province. Sister to this clade is a second haplotype of *L. mirabilis* also from Buenos Aires Province, Argentina, which on average diverges by 0.8% from the clade containing the Chilean haplotypes; the resulting clade is also well supported (JK = 92).

Morphological (see below) and phylogenetic analyses shown that all Chilean specimens analyzed belong to a single species. Comparisons with the descriptions and museum series of other taxa, allow us to conclude that the correct name to apply to the studied populations is *Latrodectus thoracicus* Nicolet, 1849, whose redescription follows.

Latrodectus thoracicus Nicolet, 1849
(Figs. 2-4)

Latrodectus thoracicus. Nicolet, 1849: 462.

Latrodectus formidabilis. Nicolet, 1849: 460, lam. 4, f. 10.

Latrodectus mactans. Gerschman & Schiapelli, 1943: 16.

Latrodectus curacaviensis. Levi, 1959: 39

TABLE I. List of specimens from where haplotypes of the COI gene used in the phylogenetic analysis were gathered. For those sequences generated here numbers of corresponding voucher specimens are provided (for locality data see Material Examined). Sequence marked with an asterisk was from an unidentified Chilean specimen and here referred to *L. thoracicus*.

TABLA I. Lista de especímenes a partir de los cuales se generaron los haplotipos del gen COI usados en el análisis filogenético. Para aquellas secuencias generadas en este estudio también se indica los correspondientes números de los especímenes vouchers (por datos de localidades ver Material Examined). El asterisco indica una secuencia generada por Garb *et al.* (2004) a partir de un espécimen chileno no identificado y que aquí es referido a *L. thoracicus*.

	species	specimen voucher	accession number	source
Ingroup				
1	<i>L. antheratus</i>		AY383048	Garb <i>et al.</i> 2004
2	<i>L. antheratus</i>		AY383047	Garb <i>et al.</i> 2004
3	<i>L. corallinus</i>		AY383062	Garb <i>et al.</i> 2004
4	<i>L. corallinus</i>		AY383061	Garb <i>et al.</i> 2004
5	<i>L. diaguaita</i>		AY383063	Garb <i>et al.</i> 2004
6	<i>L. diaguaita</i>		AY383064	Garb <i>et al.</i> 2004
7	<i>L. geometricus</i>		AY383066	Garb <i>et al.</i> 2004
8	<i>L. hasselti</i>		EF121035	Vink <i>et al.</i> 2008
9	<i>L. hesperus</i>		AY383071	Garb <i>et al.</i> 2004
10	<i>L. katipo</i>		EF121016	Vink <i>et al.</i> 2008
11	<i>L. mactans</i>		AY383072	Garb <i>et al.</i> 2004
12	<i>L. menavodi</i>		AY383075	Garb <i>et al.</i> 2004
13	<i>L. mirabilis</i>		AY383076	Garb <i>et al.</i> 2004
14	<i>L. mirabilis</i>		AY383077	Garb <i>et al.</i> 2004
15	<i>L. pallidus</i>		AY383055	Garb <i>et al.</i> 2004
16	<i>L. rhodesiensis</i>		AY383079	Garb <i>et al.</i> 2004
17	<i>L. thoracicus</i>		AY383074*	Garb <i>et al.</i> 2004
18	<i>L. thoracicus</i>	LT0204Cv01	GU112098	this study
19	<i>L. thoracicus</i>	LT0105Cv01	GU112099	this study
20	<i>L. thoracicus</i>	LT0105Cv02	GU112100	this study
21	<i>L. thoracicus</i>	LT0105Cv03	GU112101	this study
22	<i>L. thoracicus</i>	LT0105Cv05	GU112102	this study
23	<i>L. thoracicus</i>	LT0108Cv01	GU112103	this study
24	<i>L. thoracicus</i>	LT0108Cv02	GU112104	this study
25	<i>L. thoracicus</i>	LT0903An01	GU112105	this study
26	<i>L. tredecimguttatus</i>		AY383080	Garb <i>et al.</i> 2004
27	<i>L. variegatus</i>		AY383084	Garb <i>et al.</i> 2004
28	<i>L. variegatus</i>		AY383083	Garb <i>et al.</i> 2004
29	<i>L. variolus</i>		AY383082	Garb <i>et al.</i> 2004
Outgroup				
30	<i>Steatoda grossa</i>		AY383086	Garb <i>et al.</i> 2004

HOLOTYPE

A female from Provincia de Concepcion, Chile. The type is lost.

EMENDED DIAGNOSES

Opening of the epigynum oval to suboval, with anterior and posterior lips similar in length, and anterior margin often with a small lobulated median pointed process (Fig. 3a). Connecting ducts of the spermatheca with three loops in dorsal view; the second and third ones located at the side of the spermatheca and rotated over themselves in a single plane; the last loop outline almost an entire circle (Fig. 3c-3d). Embolus of the male palpus with three coils (Fig. 4a); the conductor placed in an angle larger than 90°, stout and strong in the distal third (Fig. 4c); radix lanceolate (Fig. 4d).

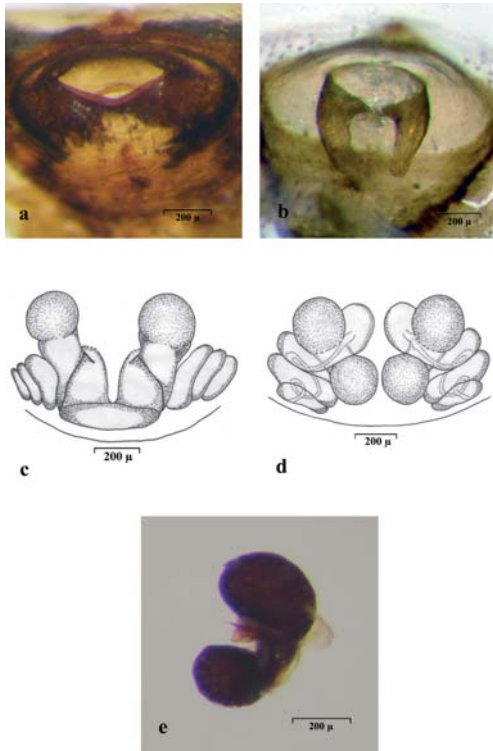


FIGURE 3: *Latrodectus thoracicus*: a) ventral view of the epiginium, b) dorsal view of the epiginium, c) ventral view of the internal genitalia, d) dorsal view of the internal genitalia, and e) lateral view of the left spermatheca.

FIGURA 3: *Latrodectus thoracicus*: a) vista ventral del epiginio; b) vista dorsal del epiginio, c) vista ventral de la genitalia interna, d) vista dorsal de la genitalia interna, y e) vista ventral de espermateca izquierda.

DESCRIPTION:

Female: Cephalothorax, legs, and abdomen are shiny black (Fig. 2b). Abdomen with dorsal red or orange markings that are diverse in shape and arrangement (Fig. 2c-2d); ventrally with a trapezoid mark with a median cut between the spinnerets and epigastral furrow; the shape of this mark is slightly variable, but is generally the classic hourglass shape (Fig. 2e). Pair of circular spots present on each side of the lateral anterior and lateral posterior spinnerets (Fig. 2f). The pattern of these spots varies according to age and geography (Fig. 2c-2d, 4f). The abdominal

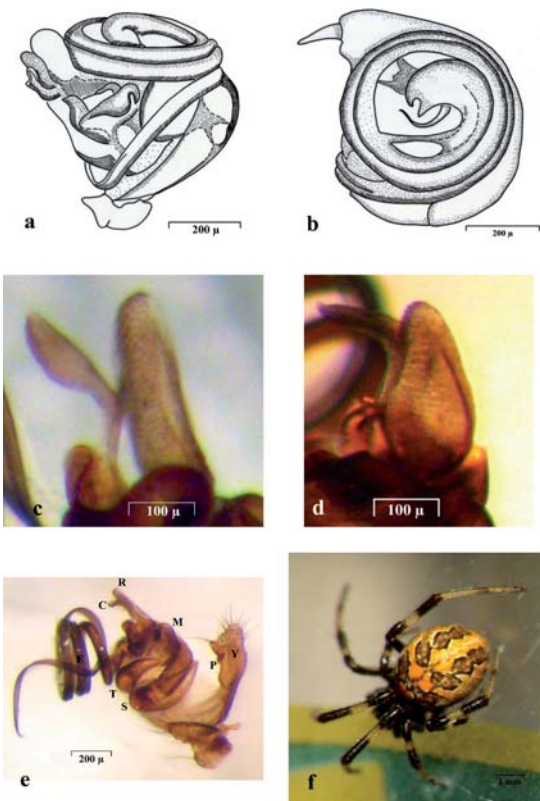


FIGURE 4: *Latrodectus thoracicus*: a) ventral view of the left palpus, b) ectal view of the left palpus, c) right palpus conductor, d) right palpus radix, e) ventral view of the right palpus, and f) immature specimen. Y = cymbium; P = paracymbial hook; S = subtegulum; T = tegulum; M = median apophysis; E = embolus; R = radix; C = conductor.

FIGURA 4: *Latrodectus thoracicus*: a) vista ventral del palpo izquierdo, b) vista ectal del palpo izquierdo; c) conductor del palpo derecho, d) radix del palpo derecho, e) vista ventral del palpo derecho, y f) espécimen juvenil. Y, Cimbio; P, Gancho del cimbio; S, subtegulo; T, tegulo; M, apófisis media; E, embolo; R, radix; C, conductor.

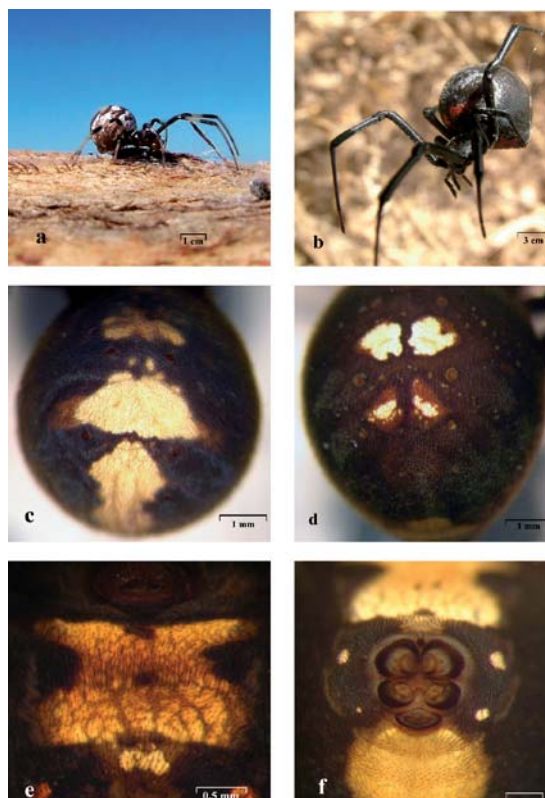


FIGURE 2: *Latrodectus thoracicus*: a) adult male, b-f) adult female, c-d) variations of dorsal pattern coloration, e) ventral pattern, and f) pattern around spinnerets.

FIGURA 2: *Latrodectus thoracicus*: a) macho adulto, b-f), hembra adulta, c-d) variaciones de patrones de manchas dorsales, e) patrón ventral, y f) manchas alrededor de las hilanderas.

dorsum with fine, curved setae of different lengths. Epigynum subglobose, rugose and projecting; with an opening oval to suboval, wider than longer, with sclerotized lips and the anterior margin often with a small median pointed process as a lobule, and both lips similar in length (Fig. 3a-3b). Internal genitalia with two dumb-bell shaped spermathecae (Fig. 3e), V-shaped but with their bases separated; the connecting ducts with three loops, the second and third ones located at the side of the spermatheca and rotated over themselves in a single plane, while the last loop outline almost an entire circle (Fig. 3c-3d). Measurements of a sample of $n=80$ are as follows: total length (excluding legs): 11.29 (10.8-13.1); cephalothorax length: 3.4 (1.3-5.8); width (at widest part): 3.0 (1.0-4.2); T coefficient: 1.12 (0.9-1.72); length tibia I: 3.8 (0.5-5.7); length patella I: 1.6 (0.6-

5.33); TT coefficient: 1.59 (0.56-2.03).

Male: smaller than the female. Cephalothorax dark brown, wider than longer. Abdomen off-white, with four dark dorsal bands; two of them are irregular and located in the antero-medial part and project to the sides; the other two bands are elongated and directed to the posterior end. Legs longer than in the female; colour dark brown with creamy white stripes (Fig. 2a). Male palp and bulb typical for the genus; embolous with three coils (Fig. 4a, 4b, 4e); the conductor is placed in an angle larger than 90° , stout and strong in the distal third (Fig. 4c); radix lanceolate (Fig. 4d). Measurements as follows ($n=8$): total length 2.27 (2.08-2.66); cephalothorax length: 0.94 (0.84-1.03); width: 0.96 (0.82-1.07); T coefficient: 0.98 (0.95-1.02); length tibia I: 181 (1.24-2.18); length patella I: 0.5 (0.39 - 0.62); TT coefficient: 2.43 (2.12-2.81).

Immatures: Around 55 days after emergence, individuals show basic characters of the genus (serrate setae on metatarsus IV, evident colulus with two setae, lateral eyes apart, and lack of teeth of the cheliceral margin). The coloration is characteristic, the cephalothorax and legs brown-orange, and dark brown between femur and patella. Dorsum of the abdomen with a series of rhomboidal dark brown spots (Fig. 4f). Venter with four circular and yellowish spots located around the spinnerets, and with a yellowish outline of the adult trapezoid spot located between the epigastral furrow and the spinnerets. Later the cephalothorax and the abdomen become black; abdomen with red spots or bands different in shape and variable arrangements.

Eggs: Spherical, yellowish, length between 0.82 and 0.94 (mean= 0.89). Mean number of eggs on each egg sac is 536 ± 48 ($n= 52$).

Egg-sac ($n = 52$): spheroid or sometimes pear-shaped, with a mean diameter of 9.76 (7.11-12.87); soft papery texture without surface ornamentations. When fresh, silk thread is present on the surface and is soft.

COMPARISONS

Females of *L. thoracicus* differ from those of the other *Latrodectus* species mentioned for Chile, *L. variegatus*, *L. curacaviensis*, and *L. geometricus* (Platnick 2009), by the arrangement of the spermathecae, the number of loops of the genitalia

ducts, and the arrangement of the dorsal spots of the abdomen. *Latrodectus geometricus* has parallel spermathecae while those of *L. thoracicus* are placed in a V-formation. *Latrodectus variegatus* and *L. curacaviensis* (Abalos 1980) have spermathecal ducts with two loops, and *L. thoracicus* has three loops. *Latrodectus curacaviensis* has also a black abdomen with an elongated central band, surrounded by three pairs of bands placed obliquely (*vide* McCrone & Levi 1964; Abalos 1980). *Latrodectus thoracicus*, on the other hand, has a black abdomen but with spots or bands of different shapes and variable arranged.

Males of *L. thoracicus* differ from those of *L. variegatus*, *L. geometricus*, and *L. curacaviensis* by the number of loops of the embolus and the shape of the conductor. The embolus of *L. variegatus* has two loops, that of *L. geometricus* has four, while that of *L. thoracicus* has three loops. In *L. curacaviensis* the distal third of the conductor is more or less cylindrical and in *L. thoracicus* is dilated.

DISTRIBUTION

This species is found throughout Chile. It ranges from Arica y Parinacota Region in the north (20°S) to Barranca Negra in Magallanes Region in the southern extreme of the continent (50°S) and adjacent areas of Argentinean Patagonia. It is abundant in the central and southern parts of Chile.

MATERIAL EXAMINED

Región de Arica y Parinacota (XV): 1 ♀ Sapó Huirra, Camino a Azapa. 21-Nov-1965, Tolar col. (MNHN Cat 774); Región de Tarapacá (I): 1 ♀ La Tirana. Jun-2001, J. Ardiles col. (MZUC-UCCC); Región de Antofagasta (II): 1 ♀ San Pedro de Atacama. 08-Feb-2009, M. A. Aguilera col. (MZUC-UCCC); 1 ♀ Calama. 12-Sep-1986, Ibarra col. (MZUC-UCCC); Región de Atacama (III): 21 ♀ - 4 ♂ Parc. Las Bombas, Camarones sur, Vallenar. 11-Feb-2004, M. A. Aguilera col. (MZUC-UCCC). 1 ♀ Algarrobal. 16-Sep-1984, Loyola col. (MZUC-UCCC); Región de Coquimbo (IV): Prov. del Choapa: 2 ♀ Los Molles. Oct-2003, Agosto y Espinoza col. (MZUC-UCCC); Prov. del Elqui: 2 ♀ Andacollo. Sep-2003, P. Augusto col. (MZUC-UCCC); 1 ♀ Cerro la Coipa, Andacollo. 17-Aug-2003, P. Augusto col. (MZUC-UCCC); 4 ♀ Lomas Los Huasos, Andacollo. Sep-2003, P. Augusto col. (MZUC-UCCC); 1 ♀ Lomas Los Huasos, Andacollo. Sep-2003, P. Augusto col. LT0903An01 (MZUC-UCCC); 1 ♀ Cerro El

Curque, Andacollo. 23-Jul-2003, P. Augusto col. (MZUC-UCCC); 1 ♀ Playa blanca, Chungungo. 27-Feb-2005, J. Pizarro col. (MZUC-UCCC); 1 ♀ Quebrada porotito, 35 km N La Serena. 05-Jun-2004, D. Iriart col. (MZUC-UCCC); 1 ♀ Quebrada porotito, 35 km N La Serena. 16-Apr-2004, J. Pizarro col. (MZUC-UCCC); 1 ♀ Quebrada porotito, 35 km N La Serena. Jun-2004, P. Augusto col. (MZUC-UCCC); 2 ♀ Juan Soldado, N de La Serena. 05-Jun-2004. L. Espinoza col. (MZUC-UCCC); 3 ♀ Juan Soldado, N de La Serena. 05-Jun-2004. J. Pizarro col. (MZUC-UCCC); 2 ♀ Punta de Teatinos N La Serena. Sep-2003. L. Espinoza col. (MZUC-UCCC); 1 ♀ Totoralillo 15 km S Coquimbo. 01-May-2004. L. Espinoza col. (MZUC-UCCC); 1 ♀ Vicuña. 05-Nov-2006. E. Cisterna col.; 42 ♀ Vicuña. 27-Dec-2007. E. Cisterna col. (MZUC-UCCC). Región del Maule (VII): Prov. Talca: 34 ♀ Parc. 62, Casas Viejas, Com. San Clemente. 3-Feb-2004, M. A. Aguilera col. (MZUC-UCCC); 1 ♀ Parc. 62, Casas Viejas, Com. San Clemente. 3-Feb-2004, M. A. Aguilera col. LT0204Cv01 (MZUC-UCCC); 38 ♀ - 2 ♂ Parc. 62, Casas Viejas, Com. San Clemente. 5-Jan-2005, M. A. Aguilera col. (MZUC-UCCC); 5 ♀ Parc 62, Casas Viejas, Com. San Clemente. 5-Jan-2005, M. A. Aguilera col. LT0105Cv01, LT0105Cv02, LT0105Cv03, LT0105Cv04, LT0105Cv05 (MZUC-UCCC); 14 ♀ - 2 ♂ Parc. 62, Casas Viejas, Com. San Clemente. 20-Jan-2008, M. A. Aguilera col. 2 ♀ Parc. 62, Casas Viejas, Com. San Clemente. 20-Jan-2008, M. A. Aguilera col. LT0108Cv01, LT0108Cv02 (MZUC-UCCC). Prov. Linares: 3 ♀ Aeródromo, Linares. 15-Mar-2009. C. Pérez col. (MZUC-UCCC). Región del Bío - Bío (VIII): Prov. de Ñuble: 1 ♀ San Fabián de Alicó. 28-Mar-2009. M. A. Aguilera col. (MZUC-UCCC). Prov. del Bío - Bío: 6 ♀ Los Angeles. 18-Jan-1968, F. Haemmerli col. (MZUC-UCCC); 1 ♀ Los Angeles. 27-Feb-2004. Y. Vera col. (MZUC-UCCC); 1 ♀ Charrúa 10 km de Monteáguila. 13-Feb-2009. P. Oyarzún col. (MZUC-UCCC); 1 ♀ Quilaco 7 km E Quilaco. 11-Mar-2006. P. Arias col. (MZUC-UCCC); 1 ♀ Charrúa, 10 km de Monteáguila. 13-Feb-2009, Oyarzún col. (MZUC-UCCC); 1 ♀ VII Región. Feb-2008. E. Hernández col. (MZUC-UCCC). Región de La Araucanía (IX): Prov. Malleco: 3 ♀ km 8 entre Angol y Renaico. 23-Dec-1967, T. Cekalovic col. (MZUC-UCCC); 1 ♀ Malleco-Victoria. Feb-1984, Anónimo (MNHN Cat 784). Prov. Cautín: 4 ♀ Temuco. Dec-1983, Anónimo

(MNHN Cat 869); 1 ♀ Temuco, 40 km E por Huichahue. Prov. Cautín. Feb-1984, M. Vallejos col. (MNHN Cat 893); 11 ♀ Quino, Temuco. Feb-2007, F. Romero col. (MZUC-UCCC); 1 ♀ Los riscos a 5 km de Pucón. 14-dic-1984, G. Weigert col. (MZUC-UCCC); 1 ♀ Río Huachitivo. 12-Feb-81. T. Cekalovic col. (MZUC-UCCC). Región de Los Lagos (X): Prov. Llanquihue 4 ♀ Calafquén. 20-Feb-2002, A. Larrain col. (MZUC-UCCC); Región de Magallanes y La Antártica Chilena (XII): Prov. Magallanes: 1 ♀ Molco. 22-Feb-1983, T. Cekalovic col. (MZUC-UCCC); 1 ♀ Barranca negra. 25-Jan-1955, T. Cekalovic col. (MZUC-UCCC); 2 ♀ Barranca negra. 25-Dec-55, T. Cekalovic col. (MZUC-UCCC); 1 ♀ Cerro Guido. 20-Jan-66. T. Cekalovic col. (MZUC-UCCC).

NATURAL HISTORY

In the Central Valley of Chile *L. thoracicus* can be found in sunny areas with or without low herbaceous ground cover. Specimens shelter in holes in the ground, under pieces of wood and rocks or at the base of small bushes. It possible to find specimens associated with agricultural plantations such as those of oat, corn, alfalfa, and vineyards, where they are located close to the ground, under bales, among leaves and trunks of vineyards, blackberries or other bushes.

The webs of *L. thoracicus* are irregular and are placed on the ground or on the vegetation. It is possible to distinguish some specific areas in the web's structure: refuge zone, where the spider stays most of the time and places the egg-sacs; waste zone, where the spider leaves the food rests; capture zones, which correspond to all web places with the exception of those just mentioned. Another zone can be seen in the web where undigested preys are kept, wrapped with silk. Webs placed on the ground are more or less square and can be from few centimetres to up 0.5 meters wide. However, webs built on vegetation can extended longer than 2 meters from bottom to top and intermingle with the vegetation.

The most common prey of *L. thoracicus* are locusts, crickets, tenebrionids, scarabaeids, isopods, grasshoppers, and also other spiders. Occasionally *L. thoracicus* feeds on wasps.

Specimens can be found throughout most of the year, but the population fluctuates during the year. In the 2008 austral summer, two males were collected from a female's web during a search of leaves and

trunks in a vineyard at San Clemente, Región del Maule. One of the males was in the web while the other was mating with the female. The timing of egg laying varies according to latitude; at 28°S it occurs in the first weeks of December, and at 35°S in the second week of January. Each female produce five egg-sacs.

DISCUSSION

Latrodectus thoracicus was described by Nicolet (1849) on the basis of one specimen collected near Concepción, central Chile. It was considered as a junior synonym of *L. mactans* by Gertchman & Schiapelli (1943) and later of *Latrodectus curacaviensis* by Levi (1959). However, in these last two papers neither type nor topotypical material was examined. The characters mentioned by Nicolet (1849) in his description of *L. thoracicus* match those of immature specimens reared in this study. Therefore we suggest that Nicolet's description was based on an immature specimen and that the specimens here studied belong to *L. thoracicus*. Currently, after Gerschman & Schiapelli (1943) *L. thoracicus* is considered as a junior synonym of *L. mactans*; and as such is listed (spelled as *L. thoracicus*) in Platnick's (2009) catalogue, even when he does not include Chile in the distribution of *L. mactans*. Our assessment of patterns of morphological variation and the results of the phylogenetic analysis clearly indicate that *L. thoracicus* differs from both *L. mactans* and *L. curacaviensis*. Similarly, *L. thoracicus* morphologically differs from *L. variegatus*, another Chilean species described from Chiloe Island. Therefore, we consider *L. thoracicus* is a valid species.

Nicolet (1849) also mentioned *L. formidabilis* from Chile, but based on the information provided in his short description, we believe this record may represent *L. thoracicus*.

There is low genetic diversity of COI in populations of *L. thoracicus*; haplotypes recovered from Chilean populations diverge on average by only 0.5 %; this value is in line with those found by Vink *et al.* (2008) for species from New Zealand. The sample of *L. thoracicus* from San Clemente shows minimal haplotypic diversity. Four of seven specimens from this locality share the same haplotype; pairwise comparisons give an average value for this sample of 0.2 %. Remarkably, the

most abundant haplotype found in San Clemente was also recovered from Argentinean specimens identified by Garb *et al.* (2004) as belonging to *L. miriabilis* and *L. variegatus* from Buenos Aires, Río Negro, and Santa Cruz provinces. This unexpected similarity may be due to the short sequence lengths (430 base pairs) obtained by Garb *et al.* (2004) from the Argentinean specimens and/or because species have retained an ancestral genetic variant (Pamilo & Nei 1988). However, this finding questions the distinction of these nominal forms or at least the correct determination of the specimens from where these haplotypes were gathered. It is possible that *L. miriabilis* is a junior synonym of *L. thoracicus*. We did not propose any formal change to the taxonomic status of *L. miriabilis* at this stage, because we have not had the opportunity to examine the specimens sequences by Garb *et al.* (2004) to check their correct identity. *Latrodectus thoracicus* is morphologically distinguishable from *L. variegatus* as described by Nicolet (1859). Therefore, we suggest that the specimens identified by Garb *et al.* (2004) as *L. variegatus* from Argentina belong in fact to *L. thoracicus*.

Two species of *Latrodectus* inhabit continental Chile. One species, *L. variegatus*, seems to be restricted to Chiloe Island. The second species *L. thoracicus*, is widely distributed in the country. We suggest that future studies should include the analysis of type material as well as of newly collected specimens from type localities as the only way of assigning the correct taxonomic names to mitochondrial haplotypes and morphotypes. Whatever the results of these studies might be, it is safe to assume that the current taxonomic scenario does not correctly reflect the alpha diversity of the genus *Latrodectus* and/or that species' geographic ranges are poorly delimited. Given the medical relevance of this genus, it would be valuable to conduct further field work and collection based taxonomic work in order to gain a correct appreciation of *Latrodectus* diversity and geographical distribution.

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REFERENCES

- ABALOS, J.W. 1980. Las arañas del género *Latrodectus* en la Argentina. *Obra Centenaria del Museo de La Plata* 6: 29-51.
- ARTAZA O., FUENTES J. & SCHINDLER, R. 1982. Latrodectismo: evaluación clínico-terapéutica de 89 casos. *Revista Médica de Chile* 110:1101-1105.
- ARTAZA O., FUENTES J., GÓMEZ P. & MORRIS R. 1984. Latrodectismo (II): evaluación clínico-terapéutica de 78 casos. *Parasitología al Día* 8:45-49.
- CANALS, M., CASANUEVA, M.E. & AGUILERA M.A. 2004. ¿Cuáles son las especies de arañas peligrosas en Chile? *Revista Médica de Chile* 132(6): 773-6.
- FARRIS, J.S. 1983. The logical basis for phylogenetic analysis. *In: Platnick, I., Funk, V.A. (Eds.), Advances in Cladistics. Proceedings of the second meeting. Willi Henning Society* Columbia University Press, New York Vol. 2: 1-36.
- FAÚNDEZ, E.I. 2009. Arañas (Arachnida: Araneae) peligrosas de la Región de Magallanes. *Anales Instituto Patagonia* 37(1):127-131.
- FOLMER, O., BLACK, M., HOEH, W., LUTZ, R. & VRIJENHOEK, R., 1994. DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. *Molecular Marine Biology and Biotechnology* 3(5):294-299.
- GARB, J. E., GONZÁLEZ, A. & GILLESPIE, R.G. 2004. The black widow spider genus *Latrodectus* (Araneae: Theridiidae): phylogeny, biogeography and invasion history. *Molecular Phylogenetics and Evolution* 31(3):1127-1142.
- GERSCHMAN, B.S. & SCHIAPELLI R.D. 1943. Revisión del género *Latrodectus* Walckenaer 1805. *In* Sampayo, R., *Latrodectus mactans* y latrodectismo. Buenos Aires, 2(Syst.):1-23.
- GRISHIN, E. 1999. Polypeptide neurotoxins from spider venoms. *European Journal of Biochemistry* 264(2):276-280.
- LEVI, H.W. 1959. The spider genus *Latrodectus* (Araneae, Theridiidae). *Transactions of the American Microscopical Society* 78:7-43.
- LEVI, H.W. 1967a. Cosmopolitan and pantropical species of theridiid spiders (Araneae: Theridiidae). *Pacific Insects* 9:175-186.
- LEVI, H.W. 1967b. The Theridiid spider fauna of Chile. *Bulletin of the Museum of Comparative Zoology* 136:1-20.

- LOTZ, L.N. 1994. Revision of the genus *Latrodectus* (Araneae: Theridiidae) in Africa. Navorsinge van die Nasionale Museum Bloemfontein. 10(1):1-60
- NICOLET, A.C. 1849. Arácnidos. In Gay, C. (ed.), Historia Física y Política de Chile. Zoología 3: 319-543.
- ORI, M. & IKEDA, H. 1998. Spider Venoms and Spider Toxins. Journal of Toxicology. Toxin reviews 17(3): 405-426.
- ORLOVA E.V., RAHMAN A., GOWEN B., VOLYNSKI K.E., ASHTON A.C., MANSER C, VAN HEEL, M. & USHKARYOV, Y.A. 2000. Structure of α -latrotoxin oligomers reveals that divalent cation-dependent tetramers form membrane pores. Nature Structural & Molecular Biology (7): 48-53.
- PAMILO, P. & NEL, M. 1988. Relationships between gene trees and species trees. Molecular Biology and Evolution 5(5):568-583.
- PARODI, J., ROMERO, F., MILEDI, R. & MARTÍNEZ, A. 2008. Some effects of the venom of the Chilean spider *Latrodectus mactans* on endogenous ion-currents of *Xenopus laevis* oocytes. Biochemical and Biophysical Research Communications 375(4):571-575.
- PLATNICK, N.I. 2009. The World Spider Catalog. Version 10.0. American Museum of Natural History. URL:<http://research.amnh.org/entomology/spiders/catalog81-87/index.html>. Accessed: August 15, 2009.
- ROMERO, F., ALTIERI, E., QUIÑEHUAL, C. & CAYUQUEO, A. 2000. Actividad contráctil del músculo papilar cardiaco y conducto deferente de rata inducida por veneno de la araña *Latrodectus mactans* de Chile. Gayana 64(2):161-170.
- ROMERO, F., ALTIERI, E., URRUTIA, M. & JARA, J. 2003. Venom of *Latrodectus mactans* from Chile (Araneae, Theridiidae): effect on smooth muscle. Revista de Biología Tropical 51(2):305-312.
- ROMERO, F., SALVATICI, R., SCHULZ, C., CABEZAS, M., STANDEN, D. & ALTIERI, E. 1996. Efecto del veneno de *Latrodectus mactans* sobre la respuesta contráctil del músculo papilar de rata. Revista Chilena de Cardiología 15(3):112.
- SCHENONE, H. & CORREA, L. 1985. Algunos conocimientos prácticos sobre la biología de la araña *Latrodectus mactans* y el síndrome de latrodectismo en Chile. Boletín Chileno de Parasitología 40(1-2):18-23.
- SORENSEN, L., CODDINGTON, J.A. & SCHARFF, N. 2002. Inventorying and estimating subcanopy spiders diversity using semiquantitative sampling methods in an Afriomontane forest. Environmental Entomology 31(2):319-330.
- SWOFFORD, D.L. 2000. PAUP*. Phylogenetic Analysis using Parsimony (* and other methods). Sinauer Associates, Sunderland, Massachusetts.
- TAMURA, K., DUDLEY, J., NEL, M. & KUMAR, S. 2007. MEGA 4: Molecular Evolutionary Genetics Analysis (MEGA) software version 4.0. Molecular Biology and Evolution 24(8):1596-1599.
- THOMPSON, J.D., GIBSON, T.J., PLEWNIAK, F., JEANMOUGIN, F. & HIGGINS, D.G., 1997. The CLUSTAL X windows interface: flexible strategies for multiple sequence alignment aided by quality analysis tools. Nucleic Acids Research 25(24): 4876-4882.
- UPTON, M.S. 1991. Methods for collecting, preserving, and studying insects and allied forms. The Australian Entomological Society, Miscellaneous Publication N°3. 4th edition, Brisbane, Australia. 86 pp.
- VINK, C.J., SIRVID, JAGOBA, MALUMBRES-OLARTE, J., GRIFFITHS, J.W., PAQUIN, P., & PATERSON, A.M. 2008. Species status and conservation issues of New Zealand's endemic *Latrodectus* spider species (Araneae: Theridiidae). Invertebrate Systematics 22(6):589-604.

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