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A new species of *Psectrascelis* (Coleoptera: Tenebrionidae: Pimeliinae) from the coastal dunes of the Atacama Desert, Chile

SEBASTIÁN LARREA-MEZA[1], PABLO PINTO[2], GONZALO A. COLLADO[3] & ÁLVARO ZÚÑIGA-REINOSO[4, 5]

[1] *Laboratorio de Ecología e Impactos Ambientales, Escuela de Biología Marina, Facultad de Ciencias del Mar y de Recursos Naturales, Universidad de Valparaíso, Avenida Borgoño 16344, Reñaca, Viña del Mar, Chile*
sebalarrea@gmail.com; https://orcid.org/0000-0001-8066-7515
[2] *Escuela de Salud, Campus Rodelillo, Universidad de Viña del Mar, Viña del Mar, Chile.*
gaiseriku@gmail.com; https://orcid.org/0000-0002-0247-0976
[3] *Departamento de Ciencias Básicas, Facultad de Ciencias, Universidad del Bío-Bío, Avenida Andrés Bello 720, Chillán, Chile.*
gcollado@ubiobio.cl; https://orcid.org/0000-0001-9076-4255
[4] *Institut für Zoologie, Universität zu Köln, Zülpicher Str. 47b, 50674, Köln, Germany.*
azunigar@uni-koeln.de; https://orcid.org/0000-0002-7949-081X
[5] *Corresponding author.*

Abstract

Psectrascelis is one of the most specious genus of darkling beetles of South America, comprising 82 species that inhabit arid and semiarid environments. The species are distributed mainly in Argentina and Chile. In general, the taxonomy of several species in this group has been difficult to approach. An example is *Psectrascelis pilosa* Solier and *Psectrascelis conjugens* Fairmaire, species morphologically similar which presents a rather poor original description and mislabelled. These species are distributed sympatrically in coastal terraces at the northern side of Huasco river, northern Chile. We collected several specimens of *Psectrascelis* on the southern side of Huasco river which are morphologically similar to *P. pilosa*. Morphological and genetic analysis support the description of the species *Psectrascelis ammophila* sp. nov., which is more closely related to *Psectrascelis elongata* and *Psectrascelis pilipes specularis* than other morphologically similar species (*Psectrascelis pilosa* and *Psectrascelis conjugens*). The importance of the Huasco river as barrier and the evolutionary convergence is discussed to explain the crypsis and the phylogenetic pattern found.

Key words: Biogeographical barrier, cryptic species, convergence, phylogeny, Huasco river, taxonomy, Nycteliini

Introduction

The genus *Psectrascelis* Solier, 1836 is a large monophyletic group of flightless darkling beetles in the tribe Nycteliini (Flores 2000). It is also one of the most diverse groups of Tenebrionidae in South America, with 82 species (Peña 1985, Flores 1997, 2007, Cid-Arcos & Campodónico 2017). *Psectrascelis* is mainly distributed in Argentina and Chile, but is also present in Peru, Bolivia and Paraguay (Flores 1997). Chile is the country that represents the greatest diversity of the genus, with 42 species (Peña 1985, Zúñiga-Reinoso & Flores 2013, Cid-Arcos & Campodónico 2017). Most species inhabiting different arid and semiarid environments and they can be found in highlands of Andes and Costa cordilleras or in lowlands of Atacama and Patagonia (Peña 1985, Vidal & Guerrero 2007, Zúñiga-Reinoso & Flores 2013). Biology of these darkling beetles is still rather unknown, and only some comment about its low vagility have been recorded, because most of the species are active during the day or night walking around their hiding places (Peña 1985, Flores 1997). Moreover, the larvae develop on sandy substrates with good aeration (Vidal & Guerrero 2007) and the adult inhabit in the same spot where the larvae are developing (Peña 1985). Therefore, the distribution of these beetles is in small patches with the microhabitat requirements for its biological cycle.

In general, the taxonomy of *Psectrascelis* has been difficult and challenging. In the last revision of the group, Peña (1985) left several species with arbitrary taxonomic arrangements. However, this can be explained given the

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characteristics of the group, among which protrude the great number of species, the huge distribution range, lost types material, some poor original description and imprecise type localities. For instance, the cases of *Psectrascelis pilosa* Solier, 1851 and *Psectrascelis conjugens* Fairmaire, 1876, whose original descriptions are incomplete, type material lost and/or mislabelled material. Peña (1985) after reviewing the holotype, fixed the distribution of *P. pilosa* on the coast of the Atacama region in Chile, from Chañaral (- 26.30 S) to the northern side of the Huasco river (-28.42 S). Additionally, for similar reasons than above, Peña (1985), after reviewing the holotype of *P. conjugens*, assigned Puerto Viejo, Copiapó river mouth as type locality, which is within the distributional range of *P. pilosa*. In this area, both species are constrained in a thin band of coastal dunes where adults and larvae cohabit (Zúñiga-Reinoso pers. obs.). Fairmaire (1876) mentioned that *P. conjugens* and *P. pilosa* are morphologically very similar but *P. conjugens* has a more compacted and rounded elytra. Additionally, another remarkable difference between both species is the subparallel wrinkles on the pronotum present in *P. conjugens* (Peña 1985). Recently we collected several specimens of *Psectrascelis* on the southern side of Huasco river which are very similar to *P. pilosa*, but our morphological and genetic analyses support the hypothesis of a new taxon.

Materials and Methods

Sampling and morphological analysis. All material was collected by hand under the sand in coastal dunes in the locality of Aguada de Tongoy, about 14 km south of the Huasco city, northern Chile, on the southern side of Huasco river (Fig. 1). Additionally, to determinate which morpho corresponds to *P. pilosa*, we got high resolution photos of the paratype material of this species from the National Museum of the Natural History of Paris to compare with our material. Besides, we got photos of the specimen of *P. conjugens* analysed in Peña (1985), which was labelled as "*cum typus comparatum*". This material was used as an indirect reference to the holotype because the Holotype was not found in the National Museum of the Natural History of Paris (Antoine Mantilleri com. pers.).

For the description of external morphology, we partially followed Flores (2007). Body length was measured dorsally along the midline from the apex of the mandibles to the elytral apex. For the description of the external genitalia we followed Flores (1996) for males and Kaminski et al. (2020) for females. In each case, the genitalia were immersed in a solution of hot water and KOH for 5-10 minutes for cleaning. To establish differences between species regarding external morphology and male and female genitalia of the new species we compared our candidate species with those species morphologically most similar (i.e. *P. pilosa*). All the morphological structures were revised under stereo microscope Zeiss Stemi 508 (ZEISS, Germany) and scanning electron microscope Hitachi SU3500 (Hitachi High-Tech, Japan).

The specimens reviewed were deposited in the following collections: Museo Nacional de Historia Natural, Santiago, Chile (MNNC); Museo de Zoología, Universidad de Concepción, Concepción, Chile (MZUC); Instituto Argentino de Investigaciones de Zonas Áridas, Mendoza, Argentina (IADIZA); Instituto de Entomología, Universidad Metropolitana de Ciencias de las Educación, Santiago, Chile (IEUMCE); Museo de Entomología de la Universidad de Chile, Santiago, Chile (MEUC); Sebastián Larrea Private Collection, Valparaíso, Chile (SLPC); Pablo Pinto Private Collection, Valparaíso, Chile (PPPC); Andrés Ramírez Private Collection, Atlanta, USA (ARPC); Marcelo Guerrero, Private Collection, Santiago, Chile (MGGC); Pedro Vidal Private Collection, Santiago, Chile (PVGH); Sergio Roitman, Private Collection, Santiago, Chile (SRRC); Manuel Diéguez, Private Collection, Santiago, Chile (VMDC).

Molecular analysis. Sequences of the mitochondrial gene cytochrome oxidase I (COI) of three specimens of the new taxon from Aguada de Tongoy (voucher code: PsecATON1 to PsecATON3) were analysed. Sequences were compared with morphologically similar species (*P. pilosa*: Tres Playas, Atacama Region and *P.* aff. *conjugens*: Caleta Totoral, Atacama Region), sympatrically distributed species (*Psectrascelis elongata* Solier, 1851) and closer distributed species (*Psectrascelis pilipes specularis* Peña, 1974: Punta de Choros, Coquimbo Region) (see Fig. 1). DNA was extracted from the thoracic muscles using EZNA® Insect DNA Kit (Omega Bio-Tek, Inc.). Amplification protocol for partial fragments of COI gene were performed following Zúñiga-Reinoso & Méndez (2018). The DNA sequence of each individual was edited and aligned in BIOEDIT version 7.0.5.3 (Hall 1999) and, subsequently, manually checked.

For the phylogenetic analyses we first selected the best model of sequence evolution (TIM2+G) using the Akaike Information Criterion (AIC) in the program jModelTest 0.1.1 (Posada 2008). A Bayesian inference analysis was performed using the program Mr. Bayes 3. 2. 6 (Huelsenbeck & Ronquist 2001) implemented in the server

CIPRES Science Gateway 3.3 (Miller et al. 2010). The analysis was run for 10 million generations, sampling every 1000 trees using *Psectrascelis* (*Cerostena*) *impressicollis* Germain, 1855 as outgroup. The initial 25 % of the resulting trees were discarded as burn-in. Once convergence of the four independent runs was confirmed by the average standard deviation of split frequencies and the potential scale reduction factor, results from the runs were combined to obtain a total of 30,004 trees. We constructed a consensus tree by a 50 % majority rule; the consistency of the nodes was determined by posterior probabilities.

Results

Phylogenetic analysis. The matrix has a length of 825 bp and contains 83 polymorphic sites and 65 parsimony informative characters. Three indels were detected for COI in the species *P. pilosa* and *P.* aff. *conjugens* at positions 516-518 (further information in Table 1). The Bayesian tree shows two clades (pp= 1), one of them is highly supported (pp= 0.99) containing the species *P. conjugens* and *P. pilosa* as sister. The second one is poorly supported (pp= 0.66) and contain the subspecies *P. p. specularis* as sister to the clade formed by *P. ammophila* and *P. elongata* (pp= 1) (Fig. 2).

Table 1. Collection localities of the Psectrascelis specimens used in this study. Genbank accession numbers are listed. Lat and Long, decimal georeferences.

Species/subspecies	Voucher	Locality	Lat	Long	COI Length	Genebank
P. impresicollis	PimpFAR1	Farellones	-33.34	-70.30	822	MT873534
P. ammophila	PsecATON1	Aguada de Tongoy	-28.57	-71.29	822	MT873535
P. ammophila	PsecATON2	Aguada de Tongoy	-28.57	-71.29	822	MT873536
P. ammophila	PsecATON3	Aguada de Tongoy	-28.57	-71.29	822	MT873537
P. elongata	PeloATON1	Aguada de Tongoy	-28.57	-71.29	822	MT873532
P. aff. *conjugens*	PconjB19_1	Caleta Totoral	-27.82	-71.08	538	MT873538
P. pilosa	PpiloB16_1	Tres Playas	-28.43	-71.18	783	MT873539
P. pilosa	PpiloB16_2	Tres Playas	-28.43	-71.18	783	MT873540
P. pilipes specularis	PspeCHOR2_1	Punta de Choros	-29.29	-71.35	822	MT873533

Psectrascelis ammophila Larrea-Meza & Zúñiga-Reinoso, sp. nov.

(Fig. 1, 2, 3, 4)

Type locality. Chile, Atacama Region, Huasco Province, Aguada de Tongoy (-28.57 S, -71.29 W).

Type material. *Holotype*. Chile, Atacama Region, Huasco Province, Aguada de Tongoy (-28.57 S, -71.29 W). 16-X-2017. Leg. S. Larrea (MNNC). *Paratypes*: All paratypes with the same locality as the holotype. 17 ♀♀/9 ♂♂. Leg. S. Larrea. (10♀♀/5♂♂ [SLPC], 3♀♀/2♂♂[MNNC], 2♀♀/1♂ [IADIZA], 2♀♀/1♂ [MZUC]). 5♀♀/ 1♂. Leg. P. Pinto (1♂/1♀ [PPPC], 3♀♀ [MNNC], 1♀ [MZUC]). 8♀♀/ 2♂♂. Leg. A. Ramírez (2♂♂/2♀♀ [ARPC], 2♀♀ [MNNC], 2♀♀ [MZUC], 2♀♀ [SLPC]). 20-IX-2016. 3 ♂ ♂. Leg. M. Guerrero (2 ♂ ♂ [MGGC], 1 ♂ [PVGH]). 6 ♂ ♂/ 4 ♀ ♀. Leg. S. Roitman (2 ♂ ♂ y 2 ♀ ♀ [SRRC], 1 ♂ y 1 ♀ [MNNC], 1 ♂/ 1 ♀[IEUMCE], 1 ♂ [MEUC], 1 ♂ [VMDC]). 20-IX.2017. 3 ♂ ♂/ 4 ♀ ♀. Leg. M. Guerrero (1 ♂/1♀ [MGGC], 1 ♂/1♀ [PVGH], 1 ♂/1 ♀ [MNNC], 1 ♀ [MEUC]. 16.X.2017. 1♂/1 ♀. Leg. M. Guerrero. (1 ♀ [MGGC]), 1 ♂ [MNNC]).

Other material examined: Data exact as labelled. 1♀ Peñablanca, S. Huasco, Atacama, 9-Julio-58, Coll: L. E. Peña. [PVGH]. 1 ♂. Q. Tongoy, Freirina (S), Atacama, 17 Oct- 90, Coll: L.E. Peña. [PVGH].

Description. Body length 17.5 mm, width 9.4 mm. Body dark brown, convex and cover by grey large pilosity, dense and decumbent (Fig. 3A). Labrum emarginate and lateral margins curved; with shallowed punctures and scattered, short pilosity and sparse in the central portion being larger and dense in the lateral margin. Ventral view with abundant setae. Clypeus anteriorly sinuated, with thick punctures and scattered. Short pilosity, dense and erected covering the lateral margin, forming a tuft of hairs directed forward. Clypeal disc with scattered punctures and short and sparse pilosity. Clypeal suture not visible. Front convex and punctured. Mentum convex with the anterior

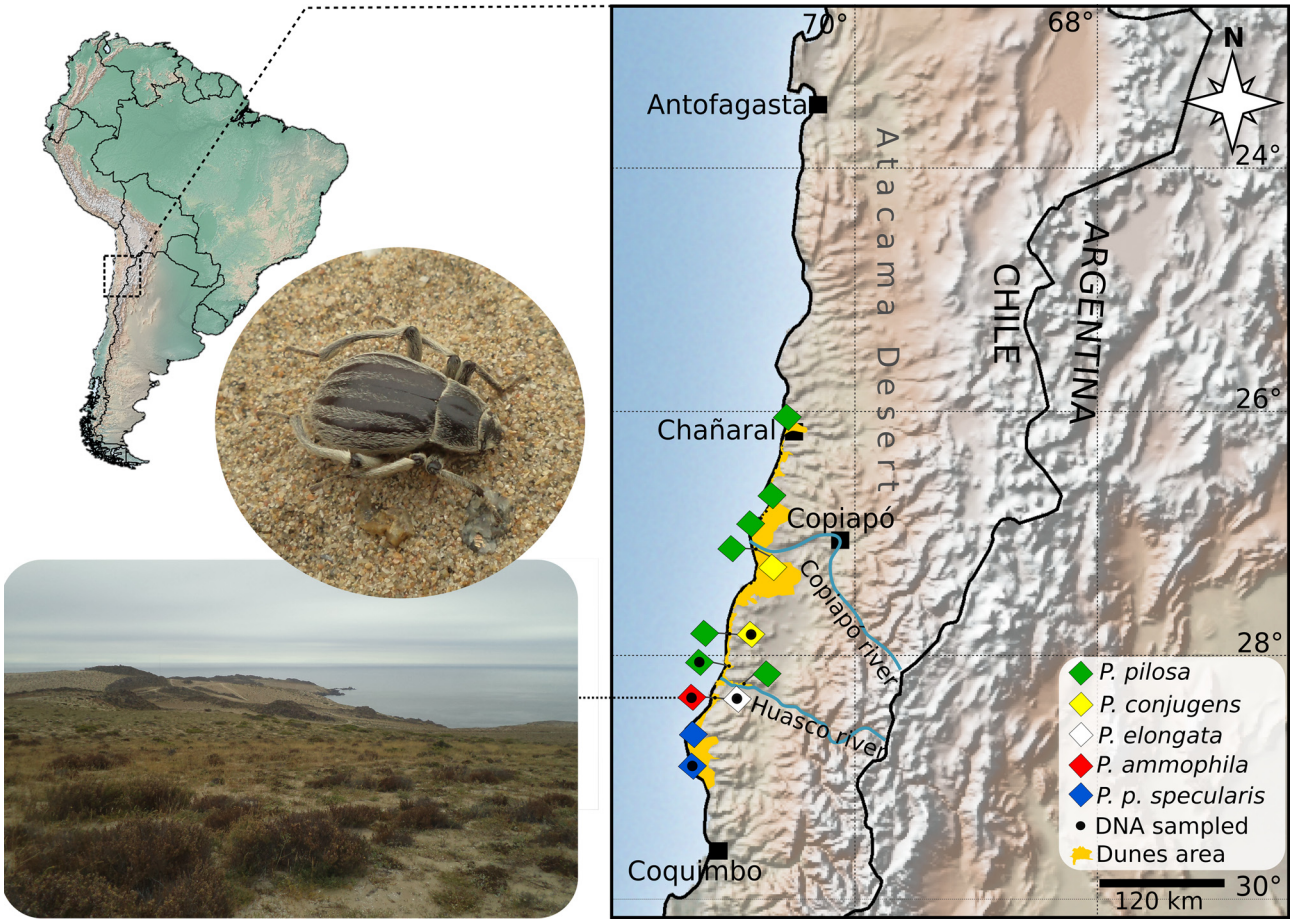


FIGURE 1. Overview of the distribution of *Psectrascelis* species included in this study. Rhombs with a black circle are the species sampled and analysed in this study. The yellow areas are the dunes zones in the study area. Most important branches of the Huasco and Copiapó basin are marked with light blue lines. Coastal habitat and a *Psectrascelis ammophila* sp. nov. specimen *in situ* in Aguada de Tongoy are shown.

margin notched, punctured and erected pilosity. Gena slightly wrinkle, with abundant pilosity mainly under the eyes. Antennae brownish, surpassing the pronotum margin by the last three segments. Segment I and II covered by erected and thick setae. From segment III to XI cover mainly by abundant, thin and curved setae, but some thick and large setae are scattered. Segment I rounded, segment II smaller than I, segment III larger than I and II together. Last segment lenticular with apex sharped. Pronotum trapezoid and convex, 1,68x wider than longer: widest in the base (5.9 mm) and anteriorly narrowest (4.0 mm). Anterior margin conspicuous, angulated, and emarginate with short and abundant setae directed forward running under and along the margin. Central area of anterior margin broad, narrowing towards sides. Punctures on the surface are less abundant and smaller in the disc and dense and bigger in both anterior and posterior margins. Pilosity concentrated in the lateral margins. Pronotum surface with longitudinal slight wrinkles. Pronotal disc raised with a notorious longitudinal cleft along the midline (Fig. 3A). Posterior margin bisinuated with short setae under margin running along the margin, shortening to almost disappear in the middle zone. Proepisternum wrinkled and punctured with dense pilosity and anterior margin raised. Place close to the posterior margin and coxa, strongly globose, glabrous and slightly grooved. Prosternum convex, wrinkled, punctured, and hairy, with anterior margin raised. Prosternal apophysis oval, hairy and thick punctures declining towards the apex (Fig. 3B). Mesosternum convex, strongly wrinkled, and punctured. Abundant pilosity, but close to the prosternal apophysis smooth and glabrous. Metasternum slightly convex with anterior margin strongly wrinkled. Central portion slightly grooved and posterior margin with pilosity (Fig. 3B). Elytra globose and rounded, laterally very curved. Brownish with abundant pilosity concentrated in the lateral margin (Fig. 3A). Thick and fine punctuation and two pairs of carinas slightly raised (Fig. 3A). Zone between carinas subconcave with dense punctures and dense pilosity. Elytral suture slightly raised (Fig. 3A), with fine punctures and glabrous. Wider in the central portion. Pseudopleurum punctured and dense pilosity. Epipleuron with sparse punctures and pilosity. Abdomen shiny black, con-

vex, with thick and fine shallowed punctures (Fig. 3B). Ventrite I wrinkled and sparse decumbent pilosity. Ventrite II with weak longitudinal wrinkles, with few hairs barely notorious. Ventrite III and IV with scarce hair in the posterior margin and some setae barely notoriuos. Ventrite V hairy, densely punctured and with abundant erected setae. Legs black or brown with tibiae and tarsi brownish. Coxae strongly punctured, hairy and with thin setae. Trochanter hairy, with thin setae. Femora hairy except by the ventral face, where the pilosity is less dense. Tibiae with a line of spine along the structure. Protibiae dorsally glabrous and strongly punctured. All tibiae with two terminal longer spines. Metatibiae wider in the posterior third arched, the ventral face a small velvety patch (Fig. 3C). Tarsi with abundant setae. Pro and mesotarsi with five segments. Metatarsi with 4 segments. Male genitalia (Fig. 4A), lateral styles of tegmen curved, narrowed towards apex. Apex with abundant long setae without tufts. Basal margin triangular. Basal lamina of tegmen shorter than ls, sub-straight with last third narrowing to the base, basal margin straight.

Female. The females are in general externally quite similar to the males, but 1.12x larger, elytra more raised and wider than the males (Fig 3D and E). Antennae in female exceed the pronotum by a single segment. Tibiae straight without the velvety patch (Fig. 3F). Females have a brownish central protuberance in the abdominal sternites II, III, and IV. Genitalia in ventral view (Fig. 4B), lobes of coxite 2 and 3 with sinuated and subparallel edges. Second lobes of coxite + valvifer (v+cl 2) are longer than wide, rough anterior margin and long sparse hairs. Third lobes of coxite (cl 3), wider in the base, completely wrinkled and abundant and disorderly long setae. A tuft of long hair (sensory setae: ss) in apical position close to the base of the fourth lobes of coxite (cl 4). Strongly sclerotized cl 4, short with a shallow indentation.

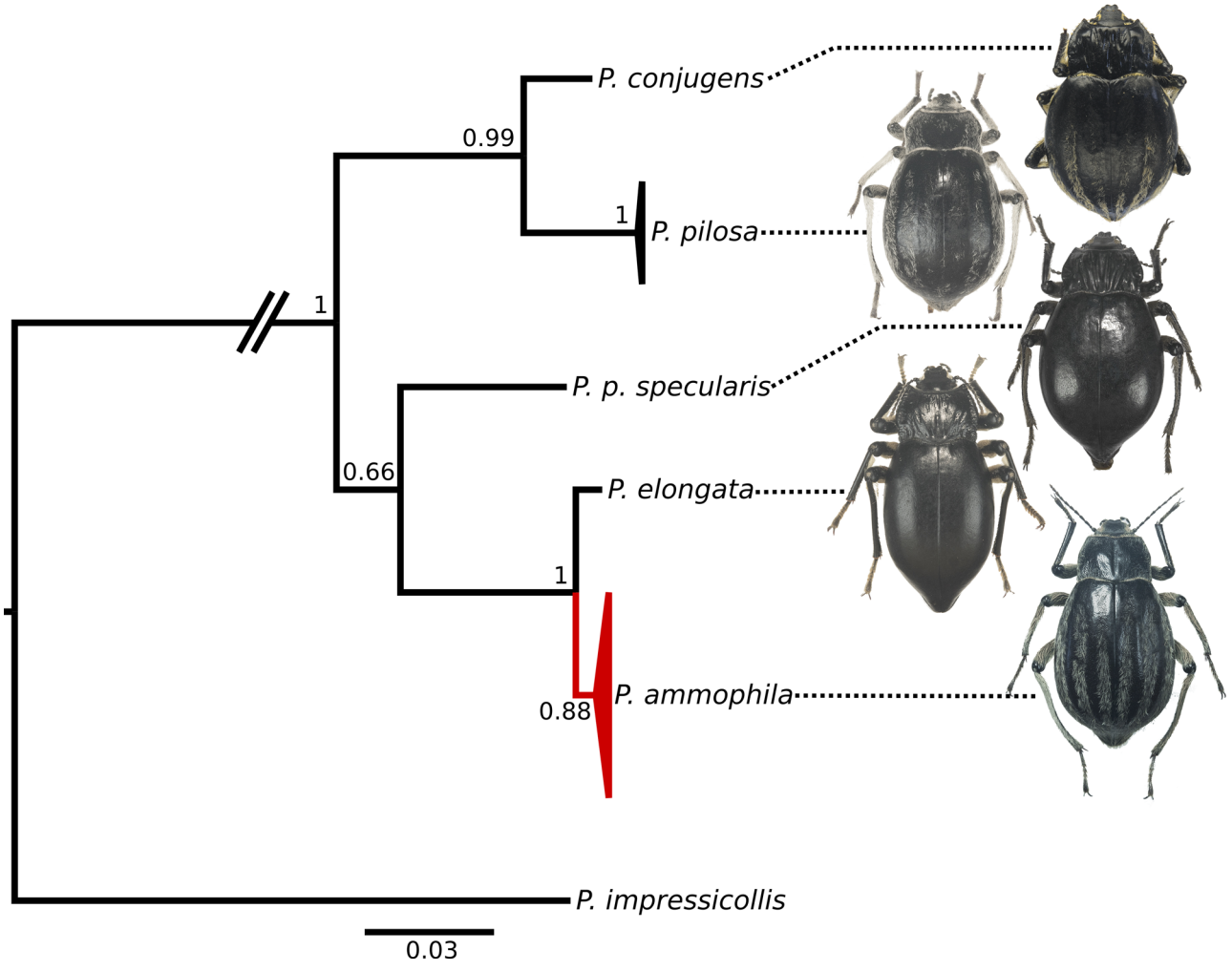


FIGURE 2. Bayesian phylogenetic tree based on COI gene. Numbers on the nodes correspond to the posterior probabilities. Front the tree are dorsal photos of species included in this study (ingroup).

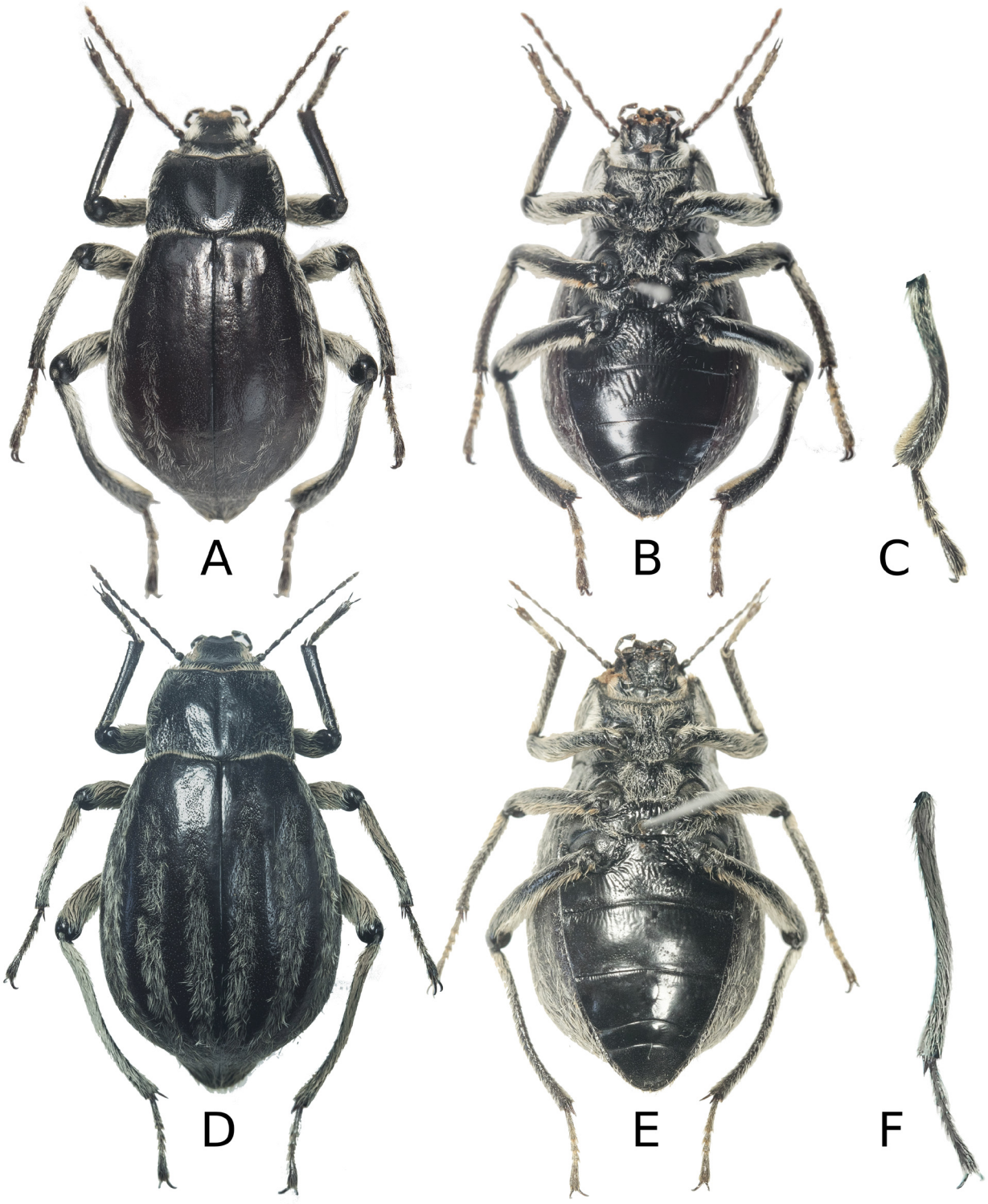


FIGURE 3. Dorsal and ventral view of the habitus of *Psectrascelis ammophila*. A-B: Male. D-E: Female. C-F: Tibiae detail of male and female respectively.

Diagnosis. The diagnostic traits of *P. ammophila* **sp. nov.** will be diagnosed in comparison with *P. pilosa* because they are morphologically very similar, although the conducted phylogenetic analysis did not render them as sister species (Fig. 4). For this, the traits of *P. pilosa* will be given in parentheses when correspond. In *P. ammophila* **sp. nov.** the pronotum (length: 3.8 mm; wide: 5.4 mm) is 1.08x larger and 1.1x narrower than *P. pilosa* (length: 3.5 mm, wide: 6 mm). Wider in a base (wider in the middle). Pronotum trapezoid (subrectangular). Elytra globose

and rounded (longer and elliptical). Body colour brown to dark brown (dark grey-black). Apex of the lateral styles without tufts (with tufts of long setae, Fig 4C). Lobes of coxite with sinuated and subparallel edges (straight and narrowed towards apex, Fig 4D). Third lobes of coxite with disorderly long setae (orderly long setae, Fig 4D). Short cl 4 with a shallow indentation (longer and smooth cl 4, Fig, 4D).

Intraspecific variation. Prosternal apophysis with different designs and mesoventrite convex or concave. Subparallel slight wrinkles on the pronotum can be notorious, weak or even absent. Average body length of male is 17.1 mm (n= 13, sd ±0.7) with a range between 15.6 and 17.9 mm. The average body length of the female is 19.3 mm (n=29, sd ±0.8) with a range between 18 and 21.1 mm.

Sexual dimorphism. Females 1.12x larger than the males (Fig. 3). Antennae in female exceed the pronotum by a single segment while the antennae in males are wider and exceed the pronotum by three segments. Male tibiae arched and with a velvety patch (Fig. 3C). Females have a brownish central protuberance in the abdominal sternites II, III, and IV.

Distribution and habitat. Chile, Atacama Region, Huasco Province. The area is located in the Coquimbean Province of the Central Chile sub-region (Morrone 2015), district of Intermediate Desert (*sensu* Peña 1966). In its currently known and very small range, *P. ammophila* **sp. nov.** inhabits the coastal terraces on the southern side of Huasco river, with bushes and borrowed in clear sand dunes at an altitude of about 100 m (Fig. 1).

Etymology. The specific name is from the ancient Greek which means sand lover (*Ammos* (ἄμμος), mean "sand", and *phila* (φιλος), mean "love") because the specimens during the day are buried under the sand of the dunes where they inhabit.

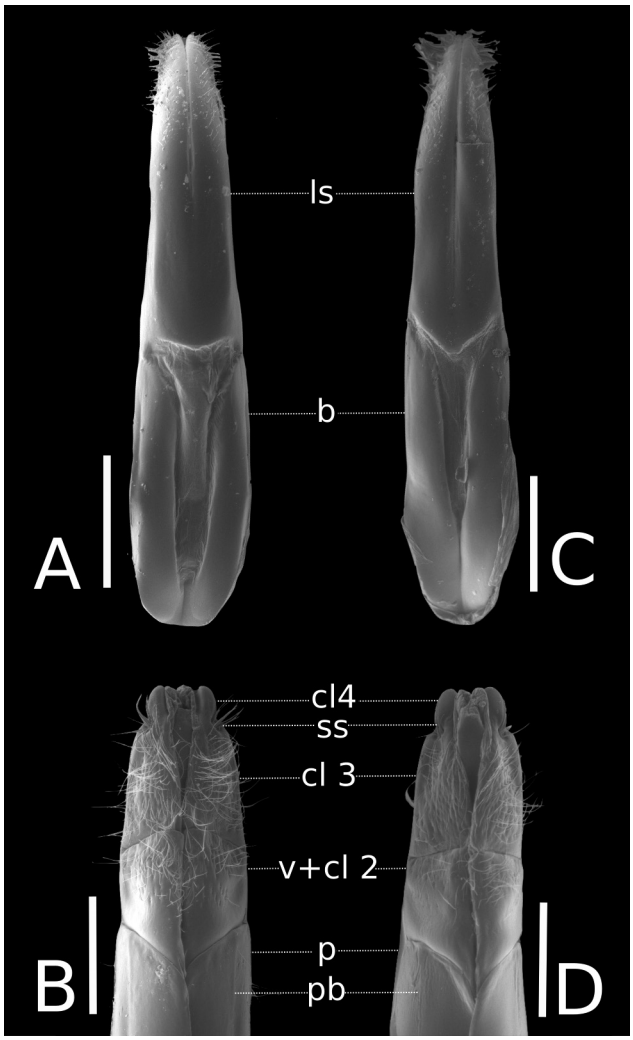


FIGURE 4. Ventral view comparison of aedeagus and ovipositor of *Psectrascelis ammophila* **sp. nov.** (A, B) with *Psectrascelis pilosa* (C, D). ls, lateral style; b, basal lamina; pb, paraproct baculi; cl2–cl4, coxite plates 2–4; p, paraproct; ss, sensory setae; v, valvifer. Scale bar: 1 mm.



FIGURE 5. Dorsal view of the Neotype female of *Psectrascelis conjugens* and the original labels.

Discussion

In this study, *P. ammophila* **sp. nov.** is proposed as a new species in the genus *Psectrascelis* based on traditional characters (external morphology and male genitalia). Additionally, we included for the first time the female genitalia and a genetic marker as evidence to support this hypothesis. In this case, male genitalia provided useful characters for species delimitation as has been widely reported for other Nycteliini (e.g. *Psectrascelis*: Flores 2007, *Nyctelia*: Zúñiga-Reinoso & Jerez 2012, *Callyntra*: Zúñiga-Reinoso et al. 2017, *Gyriosomus*: Zúñiga-Reinoso et al. 2019). Moreover, this is the first study that includes the description of the female genitalia for a species of *Psectrascelis,* which also provided scarce characters to discriminate *P. ammophila* **sp. nov.** from other similar species of the genus (e.g. *P. pilosa*). The species *P. ammophila* **sp. nov.**, *P. pilosa* and *P. conjugens* inhabit in dunes of coastal terraces of the Atacama Desert and they share almost the same external morphology. Surprisingly, the ovipositor shape of these species is quite similar as well, likely because the shape of this structure is broadly correlated with the type of substrate (Tschinkel & Doyen 1980). Similarity of ovipositor according to the soil between no-sister species was reported for *Gyriosomus* as well (Zúñiga-Reinoso et al. 2019). Commonly, the female terminalia has been considered very useful for higher levels of classification (e.g. Iwan & Kamiński 2016, Kamiński et al. 2020) and low variation intra-genus is expected, although has been also useful at the species level in other Nycteliini (Zúñiga-Reinoso et al. 2018, 2019). On the other hand, the distribution range of these species is apparently continuing, only interrupted by the Huasco river (see Fig. 1). *Psectrascelis ammophila* **sp. nov.** is distributed in the southern side, while *P. pilosa* and *P. conjugens* are sympatrically distributed in the northern side of the Huasco river (Peña 1985). Even when *P. ammophila* is morphologically quite similar to *P. pilosa* and *P. conjugens* and closer distributed but allopatrically, the obtained topology of the phylogenetic tree indicates that *P. ammophila* **sp. nov.** do not share a most recent com-

mon ancestor (MRCA) with the other two species. *Psectrascelis ammophila* **sp. nov.** is recovered as closer to the other two species (*P. p. specularis* and *P. elongata*), being molecularly closest to *P. elongata*. This means that the Huasco river was a barrier for older divergences on the tree, representing a biogeographical brake. Additionally, the similarity of these *Psectrascelis* could be explained by convergence. The evolutionary parallelism or convergence has been explained for crypsis found in other Nycteliini (Zúñiga-Reinoso & Méndez 2018), which is also apparently common in other genera of the tribe (see Zúñiga-Reinoso et al. 2019 for *Gyriosomus,* Zúñiga-Reinoso & Méndez 2018 for *Callyntra*). The low node support value on the clade composed by *P. p. specularis*, *P. elongata* and *P. ammophila* **sp. nov.** is expected because the species included in our study is only a small sample of this speciose genus, which is considerably diverse in this region (see Peña 1985). Therefore, several lineages in this clade should be missing, but even so, the branching pattern shows two main lineages completely independent.

Other minor findings of our study are that we confirm the observation of Peña (1985) who assigned all the specimens from the northern side of Huasco river to *P. pilosa*. Additionally, *P. conjugens* was recorded only by one specimen in Puerto Viejo (Peña 1985) and only recently this species was recorded again with material assigned as *P. conjugens* from Caleta Totoral (R. Honour com. pers. and here). Therefore, in this study we would be confirming its distribution into the distribution range of *P. pilosa* as Peña (1985) did but in a new locality. Additionally, due to the type material of *P. conjugens* has not been found (Antoine Mantilleri com. pers.), we designated the specimen collected by Peña as a Neotype, which was compared with the Holotype and is currently deposited in PVGH (further information in Fig. 5). With these confirmed findings 43 species of *Psectrascelis* are recorded for Chile (83 in total) of which 24 inhabit in the Coquimbean Biogeographical Province (Morrone 2015). Apparently, most of these darkling beetles are strongly linked to blooming desert phenomena. Our and other findings are linked to the last flowering desert in 2017, which allowed to discover and rediscover other darkling beetles as well, apparently highly depending on strong ENSO years (e.g. Guerrero & Vidal 2018, Guerrero & Diéguez 2018, Zúñiga-Reinoso et al. 2019).

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