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Unravelling the phylogenetic complexities of *Ischyropsalis* (Opiliones: Dyspnoi: Ischyropsalididae) in the North Iberian Peninsula, with the description of three new species

Ricardo LÓPEZ-ALONSO^{1,*^}, Esteban PASCUAL-PARRA^{2,^}, Lucía LABRADA³,
 Carlos G. LUQUE⁴, Eduardo CIRES⁵,
 Andrés ARIAS⁶ & Claudia GONZÁLEZ-TORAL⁷

^{1,2,5,6} Department of Organisms and Systems Biology, University of Oviedo,
 c/ Catedrático Rodrigo Uría s/n 33071 Oviedo, Spain.

³ Department of Biology and Geology, I.E.S. Avelina Valladares, 36680 A Estrada,
 Pontevedra, Galicia, Spain.

⁴ Gestión del Patrimonio Bioespeleológico, Impress GroupTM Consulting S.L., P.O. Box 879,
 39080 Santander, Cantabria, Spain.

⁵ Institute of Natural Resources and Territorial Planning (INDUROT), Campus de Mieres,
 c/ Gonzalo Gutiérrez Quirós s/n, 33600 Mieres, Spain.

⁷ Royal Botanic Gardens, Kew, Richmond, TW9 3AE, UK.

Corresponding author: lopezalonsord@gmail.com

² Email: estebanfrusciante@gmail.com

³ Email: lucia.labrada@edu.xunta.gal

⁴ E-mail: carlos.g.luque@impress-group.com

⁵ E-mail: cireseduardo@uniovi.es

⁶ Email: ariasandres@uniovi.es

⁷ Email: C.gonzalez_toral@kew.org

[^]Equal contribution

Abstract. *Ischyropsalis* C.L. Koch, 1839 is a genus of harvestmen inhabiting both terrestrial surface habitats and caves in central and southern Europe. The accepted number of species is controversial due to feature similarities and their strong sexual dimorphism. The northern part of the Iberian Peninsula harbours 16 accepted species, seven of which are epigeal, while the other nine exclusively live in caves. Some of these species have not been included in the only phylogenetic study conducted on this genus, so their taxonomic status has not yet been evaluated within a phylogenetic framework. In this context, we aim to unravel the phylogenetic relationships among the taxa within this genus, from the northern part of the Iberian Peninsula, by conducting morphological and phylogenetic analyses based on mitochondrial (COI) and nuclear (EF1- α) molecular markers. Our results revealed three previously undescribed species from karstic caves in Cantabria and Asturias, herein described as *I. damiani* sp. nov., *I. impressa* Luque &

Labrada sp. nov. and *I. aguerana* Luque & Labrada sp. nov. and a new designation, *nodifera*-group, for those Iberian species related with *I. nodifera*.

Keywords. *Ischyropsalis impressa*, *Ischyropsalis aguerana*, *Ischyropsalis damiani*, taxonomy, SEM ultrastructure, phylogenetics.

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Introduction

Opiliones Sundevall, 1833, commonly known as harvestmen, are a monophyletic order of arachnids formed by four suborders, 70 families, 1691 genera and more than 6700 described species (summarized in Kury *et al.* 2024). Dyspnoi Hansen & Sørensen, 1904, one of the major clades, includes the monogeneric subfamily Ischyropsalidinae Simon, 1879 (Schönhofer 2013). Ischyropsalidinae is a monotypic and morphologically distinct west Palearctic subfamily, being its genus *Ischyropsalis* C.L. Koch, 1839, restricted to the central and southern European mountain regions (i.e., the Alps, Carpathians, Apennines and the northern part of the Iberian Peninsula) (Schönhofer *et al.* 2015). The members of this genus, characterised by large bodies and chelicerae as well as a distinct penis morphology, have been reported to be difficult to find due to their habitat preferences, which in some cases include caves (Luque & Labrada 2012; Schönhofer 2013; Schönhofer *et al.* 2015).

Recent morphological and molecular studies revealed that the Basque-Cantabrian Mountains (northern Iberian Peninsula), the Alps and the Pyrenees are centres of endemism for this genus (Schönhofer 2013; Schönhofer *et al.* 2015). The genus *Ischyropsalis* was established with *Ischyropsalis kollari* C.L. Koch, 1839, as the type species by subsequent designation (Thorell 1876: 467). Its definition was later modified and expanded with the addition of further species. At present, the genus includes 24 valid species and was revised by Schönhofer (2013). At the time of his study, Martens (1969b) could not examine the few species known from the Cantabrian Mountains, which are very closely related and strictly allopatric. Consequently, parts of his treatment of this fauna remained hypothetical. These points were later corrected by the works of E. Dresco, C. Prieto and C.G. Luque (see Luque & Labrada 2012; Schönhofer 2013, and references therein).

The taxonomy of this genus is based on a combination of the male cheliceral apophysis, including the glandular area and the penis morphology (Martens 1969a, 1969b; Prieto 1990; Luque 1991; Luque & Labrada 2012; Schönhofer *et al.* 2015). The number of species has varied during the decades due to morphological similarities, which could be convergent in the case of the genitalia according to Martens (1969b), and the strong sexual dimorphism displayed by the species of *Ischyropsalis* (Martens 1978; Schmidt *et al.* 2024). Thus, several authors have described many species (e.g., C.F. Roewer in 1950 described 30 new species), some of which have been found to be synonymous (e.g., Roewer 1950; Helvesen & Martens 1972) (see Schönhofer 2013 for specifications). In this context, Martens (1969b) and Dresco (1970), the former focusing mainly on alpine species and the latter on Iberian species, made some of the most significant contributions based on morphology to the taxonomy of this genus, although their evolutionary and biogeographic hypotheses seldom coincided (Schönhofer *et al.* 2015). In addition, the phylogenetic study of Schönhofer *et al.* (2015) focused on most of the 24 valid species recognised by Schönhofer (2013), using mitochondrial and nuclear molecular markers (*Cytochrome Oxidase Subunit I* (COI), *Elongation Factor 1 Alpha* (EF1- α) and *28S ribosomal RNA* (28S rRNA)) and showed that the morphological characters employed in current systematics were not lineage-associated. This result provided further support for the observations of Martens (1969b). Schönhofer *et al.* (2015) recovered

three major phylogenetic lineages (defined as species groups) within *Ischyropsalis*: 1) the monospecific *I. hellwigii* group — *I. hellwigii* subsp. *hellwigii* (Panzer, 1793) and *I. hellwigii* subsp. *lucantei* (Simon, 1879) —; 2) the *I. manicata* group formed by species occurring in the western Alps, the Apennines and the Carpathians (*I. carli* Lessert, 1905, *I. alpinula* Martens, 1978, *I. manicata* Koch, 1869 and *I. adamii* Canestrini, 1873); 3) the Alpine-Iberian group, formed by the Alpine clade (*I. dentipalpis* Dresco, 1959, *I. kollari* Koch, 1839, *I. lithoclasica* Martens & Schönhofer, 2010, *I. muellneri* Harmann, 1898, *I. ravasini* Hadzi, 1942 and *I. strandi* Kratochvil, 1936), and 4) the Iberian clade (*I. luteipes* Simon, 1872, *I. pyrenaea* Simon, 1872, *I. dispar* Simon, 1872, *I. magdalenae* Simon, 1881, *I. navarrensis* Roewer, 1950, *I. nodifera* Simon, 1879 and *I. robusta* Simon, 1872). However, Schönhofer *et al.* (2015) demonstrated that morphology-based classifications show little congruence with molecular phylogeny. Most of the traditionally used diagnostic characters are randomly distributed across the phylogeny, suggesting extensive homoplasy and limiting their value for inferring evolutionary relationships within the genus (Martens 1969b; Prieto 1990; Luque 1991; Luque & Labrada 2012; Schönhofer & Martens 2015). For example, the size, position and shape of the glandular fields, often associated with knob-like apophyses on their dorsal and/or distal margins, are species-specific features and constitute primary diagnostic characters for species delineation and identification. However, these traits do not consistently reflect deeper phylogenetic relationships and therefore may be misleading when used to infer higher-level affinities (Martens 1969b; Schönhofer *et al.* 2015). Although comprehensive, the study of Schönhofer *et al.* (2015) did not include in their molecular analysis the Iberian taxa *I. hispanica* Roewer, 1953, *I. petiginosa* Simon, 1913, *I. cantabrica* Luque & Labrada, 2012, *I. noltei* Dresco, 1972 and *I. gigantea* Dresco, 1968.

Schönhofer (2013) included 12 taxa occurring in the Iberian Peninsula. Six show an epigeal habitat preference (*I. hellwigii* subsp. *lucantei*, *I. luteipes*, *I. nodifera*, *I. petiginosa*, *I. robusta* and *I. hispanica*), while six are hypogean (*I. pyrenaea*, *I. navarrensis*, *I. dispar*, *I. magdalenae*, *I. cantabrica* and *I. gigantea*) (Luque & Labrada 2012). The seven Iberian species molecularly studied by Schönhofer *et al.* (2015) (*I. luteipes*, *I. pyrenaea*, *I. dispar*, *I. magdalenae*, *I. navarrensis*, *I. nodifera* and *I. robusta*) were recovered as part of an ‘Iberian clade’, which comprised two inner clades, the ‘Pyrenean’ and ‘Cantabrian’ clades, and whose members are characterised by extraordinarily long and slender chelicerae. Within this group, the differences in size and morphology of both chelicerae and the bristle field on the basal cheliceral article, located on the distomedial side, are used to differentiate between species (Schönhofer *et al.* 2015).

Some of the Iberian species not included in previous phylogenetic studies occur in the karst areas of the Cantabrian Mountain Range, located in the northern Iberian Peninsula (Sendra 2023). Many of these karstic areas are of special importance for the Cantabrian and Asturian biodiversity, as they host a highly specialized and endemic cave-dwelling fauna, with some taxa presenting unique, unusual or even bizarre morphological, behavioural or ecological adaptations (Labrada *et al.* 2010, in press; Angus *et al.* 2012; Luque & Labrada 2012, 2017; Salgado *et al.* 2012; Faille *et al.* 2021; Sendra 2023; Fresneda *et al.* 2024). In these karstic areas, two endemic troglobiont species of *Ischyropsalis* occur: the Cantabrian species *I. gigantea* and *I. cantabrica* as well as the new species described herein. Moreover, they are located within a latitudinal belt (ca 42° to 46° N in Europe), harbouring a high biodiversity of cave-dwelling terrestrial fauna (‘mid-latitude biodiversity ridge’) resulting from historical, mostly climatic, conditions (Culver *et al.* 2006). On the other hand, the species status of some other Iberian species occurring remains controversial, such as that of the epigeal *I. hispanica*, which was synonymized with *I. nodifera* by Martens (1969b), while Prieto (1990) restored this taxon as a valid species. In contrast, *I. petiginosa*, *I. gigantea* and *I. cantabrica*, though clearly distinguishable morphologically and was proposed that could be grouped into the ‘Cantabrian clade’ based on their distribution and morphological traits (Luque 1991; Luque & Labrada 2012), still lack molecular validation to confirm their phylogenetic independence (Schönhofer *et al.* 2015).

The taxonomic complexity of the genus has led to the recognition of different species groups within *Ischyropsalis*. The *dispar*-group was first formally proposed by Luque & Labrada (2012) after the description of *Ischyropsalis cantabrica*, based on a set of shared morphological characters that differentiated several Iberian taxa from the remaining species of the genus. Subsequent molecular analyses confirmed that this group represents a distinct evolutionary lineage associated with the Vasco-Cantabrian region and shaped by geographic isolation and cave colonization processes (Schönhofer *et al.* 2015).

In this context, the phylogenetic position and taxonomic status of at least three classically recognised species from the Cantabrian lineage of the Iberian clade (*I. hispanica*, *I. petiginosa* and *I. nodifera* sensu Schönhofer *et al.* 2015) have not been molecularly validated and three putative new species have been discovered in an area of high *Ischyropsalis* diversity. We aim to clarify the specific status of these taxa and describe three new species from caves in the northern Iberian Peninsula using morphological and molecular approaches.

Material and methods

Study area

The Cantabrian Range has an irregular but extensive area (approximately 20 000 km²) of carbonate outcrops of Palaeozoic (pre-Carboniferous and Carboniferous) limestones and Mesozoic (Jurassic and Cretaceous) limestones and dolomites, with a large number of caves and therefore potential cavernicolous habitats (Sendra 2023). For instance, in the provinces of Cantabria (5321 km²) and Asturias (10 604 km²) alone, accounting for 3.14% of the territory of Spain, approximately 20 000 caves have been documented (as visualized at the Spanish Caving Federation (FEE), and Cantabrian (FCE) and Asturian (FESPA) Caving Federations websites). Twenty-seven hydrogeological units have been identified by the Geological Survey of Spain in the Cantabrian region (García de la Noceda *et al.* 1999). In order to maximise the range of potential cavernicolous habitats, we have sampled three karstic areas that differed in terms of age of the rock formation, number of caves, altitude, distance from present sea-shore and geographic longitude. Fifteen caves were selected for this study in the Cantabrian region, divided into two areas: from East to West, selected hydrogeological units were Cantabria Province (Figs 1–2) and partly Somiedo-Trubia-Pravia in Asturias Province (Fig. 3). The cave entrances are referenced with coordinates expressed in decimal degrees.

Collection permits, techniques, material preservation, and repositories

The specimens were collected using pitfall traps and direct capture. They were preserved either in 75% ethanol (EtOH) or in absolute EtOH and deposited in the following institutions and collections:

- BOS = (subcollection of Opiliones: BOS-OPI, Spanish acronym), Department of Organisms and Systems Biology at Oviedo University, Asturias, Spain
- CGL = private collection of C.G. Luque, Cantabria, Spain
- CJM = collection of J. Martens, currently deposited in the Senckenberg Museum Frankfurt (SMF), Germany
- CRBA = collection of M. Rambla, Animal Biodiversity Resource Centre at Barcelona University, Spain
- MNCN = National Museum of Natural Sciences, Madrid, Spain

Field collecting permits were issued to C.G. Luque and L. Labrada by the Regional Government of Cantabria (Spain). The species used in the phylogenetic analyses were collected from their type localities.

Morphological analyses

Specimens for Scanning Electron Microscopy (SEM) were air-dried, mounted on aluminium stubs, metalized with gold (16 nm of deposit) in an Argon atmosphere with bio-rad SC-515 by the ‘Sputter coating’ method and imaged with acceleration velocities of 15 kv. Initially, some of the specimens were dehydrated in an ethanol series reaching 100% to be dried by CO₂ critical point in a pressure chamber. Stubs were examined and micrographs provided under high vacuum in a Philips XL-20 SEM at the National Museum of Natural Sciences (Spain) and in a JEOL JSM-6610LV SEM at the University of Oviedo. Chelicerae and genitalia of male and female specimens were excised, mounted on slides with clove oil, and illustrated using a Carl Zeiss IV binocular microscope with a mounted camera lucida. Description and abbreviations format follows Luque & Labrada (2012). Morphological nomenclature follows Martens (1969b) for male internal genitalia and shape of the chelicera apophyses, like effectively used in previous works (Luque & Labrada 2012; Schönhofer 2013). The final versions of the drawings

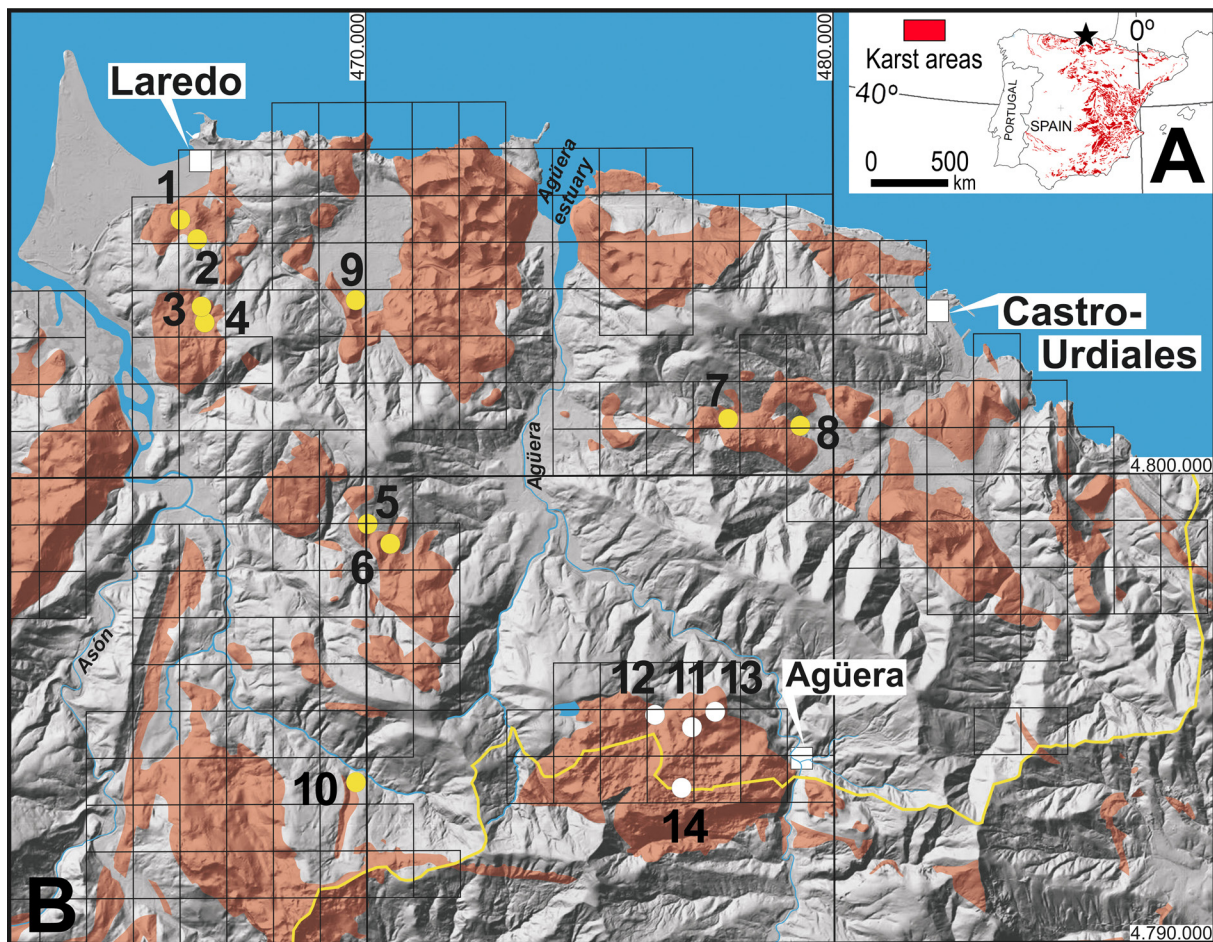


Fig. 1. Geographic distribution of *Ischyropsalis impressa* Luque & Labrada sp. nov. (yellow dots) and *I. aguerana* Luque & Labrada sp. nov. (white dots) in northeastern Cantabrian Mountains. **A.** Location of study area and map of the Karst in Spain at 1 : 1 000 000-scale where karstified carbonate rocks emerge. **B.** LiDAR visualisation of the study area showing Cantabria–Biscay border (solid yellow line). The orange-brown area represents the range of limestone outcrops from the Urgonian formation (of Aptian age, Early Cretaceous) at 1 : 50 000-scale. Location of the caves surveyed: **1.** La Baja. **2.** Hoyo de Peñaflor. **3.** Perelada. **4.** Hoyo Molino. **5.** Hoyo Menor-Tocinos. **6.** Hoyo Menor-Rejuyo. **7.** Hoyo de la Cubilla. **8.** La Lastrilla. **9.** Covacho del Cogorio, **10.** Hoyo de las Rebolligas. **11.** La Pasada. **12.** Hoyo de las Fuentes. **13.** La Tabla II. **14.** Gueriza.

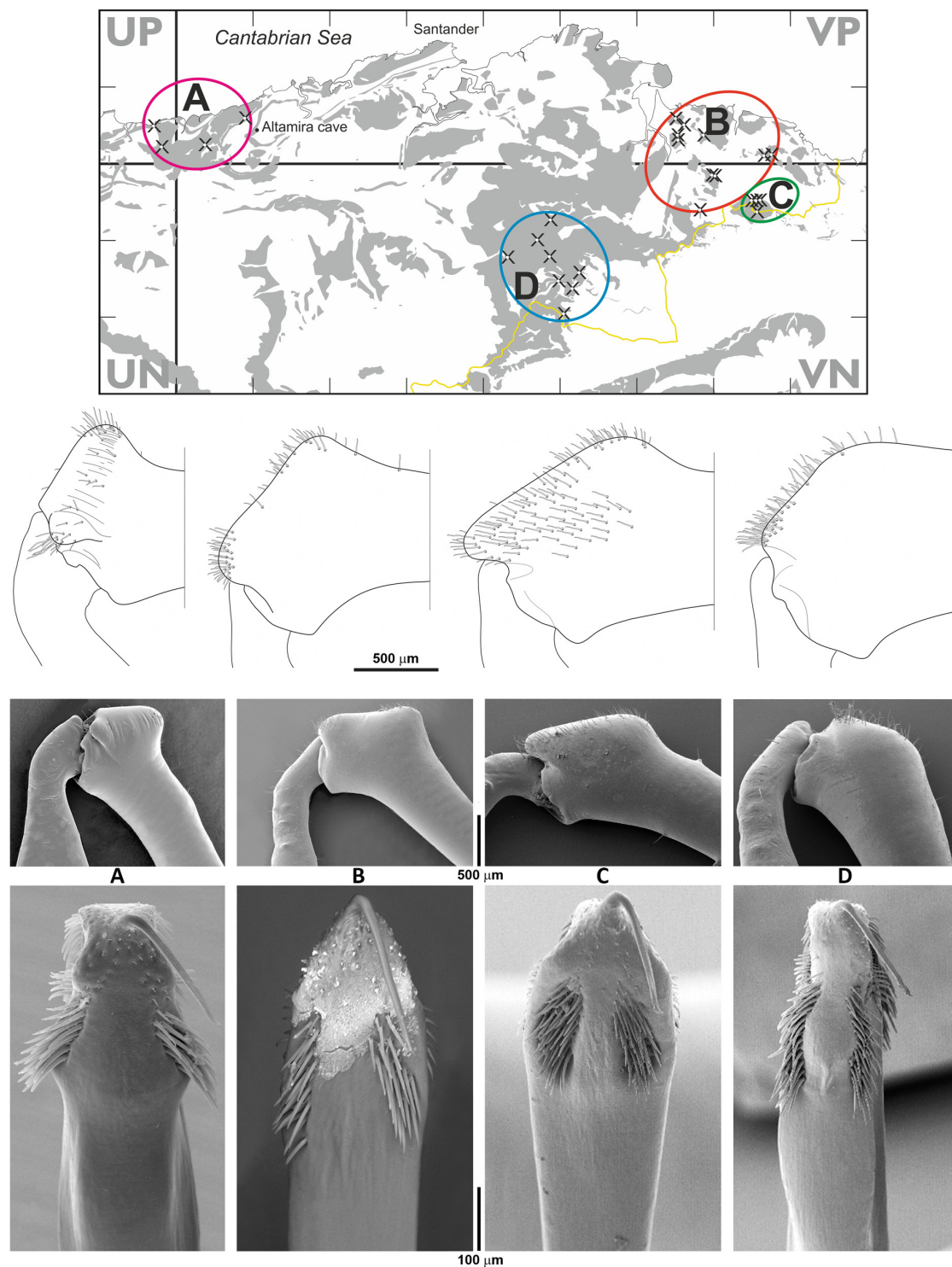


Fig. 2. Distribution of the four troglobiont species of *Ischyropsalis* C.L. Koch, 1839 in Cantabria and geological map at 1 : 50 000-scale the outcrops of the rock systems (in gray) which contain limestone (solid yellow line is the Cantabria–Burgos–Biscay border). **A.** *Ischyropsalis cantabrica* Luque & Labrada, 2012. **B.** *I. impressa* Luque & Labrada sp. nov. **C.** *I. aguerana* Luque & Labrada sp. nov. previously published as *Ischyropsalis* aff. *dispar* Simon, 1872 (Luque & Labrada 2012: figs 54–55). **D.** *I. gigantea* Dresco, 1968. Male cheliceral apophysis on distal end of basal article with basal part of second article of right chelicera (medial view) and distal part of penis (dorsal view) are shown.

were prepared using Adobe Photoshop software ver. 7.0. All measurements are in millimetres (mm) with a precision of 0.01 mm and were taken employing a micrometer disc and by using the same optical devices as above. We give the average value followed by the minimum and maximum range in brackets.

DNA extraction and amplification

DNA was extracted from the legs of the male specimens (including individuals from species type localities) following the instructions of the DNeasy Blood and Tissue kit (Qiagen, Hilden, Germany) as done in previously studies such as Schönhofer *et al.* (2015). The obtained DNA was stored at -20°C. The nuclear marker *Elongation Factor 1 Alpha* (EF1- α) was amplified using the pair of primers OP2BSAB and OPRC4 and the PCR amplification protocol described by Hedin *et al.* (2010). The *Cytochrome Oxidase Subunit I* (COI) mitochondrial marker was amplified by using the primers C1-J-1718SPIDERA and C1-N2776SPIDER and the PCR protocol described in Vink *et al.* (2005). For those samples with fragmented DNA, the internal set of primers of Leray *et al.* (2013), mlCOIintF and mlCOIintR, and the universal primers of Folmer *et al.* (1994), HCO2198 and LCO1490, were used, following the PCR conditions described by Leray *et al.* (2013). The PCR products were visualized in agarose gel. The ones with the expected size (see Table 1 for details) were sent to Macrogen (Macrogen, Madrid, Spain) for Sanger sequencing.

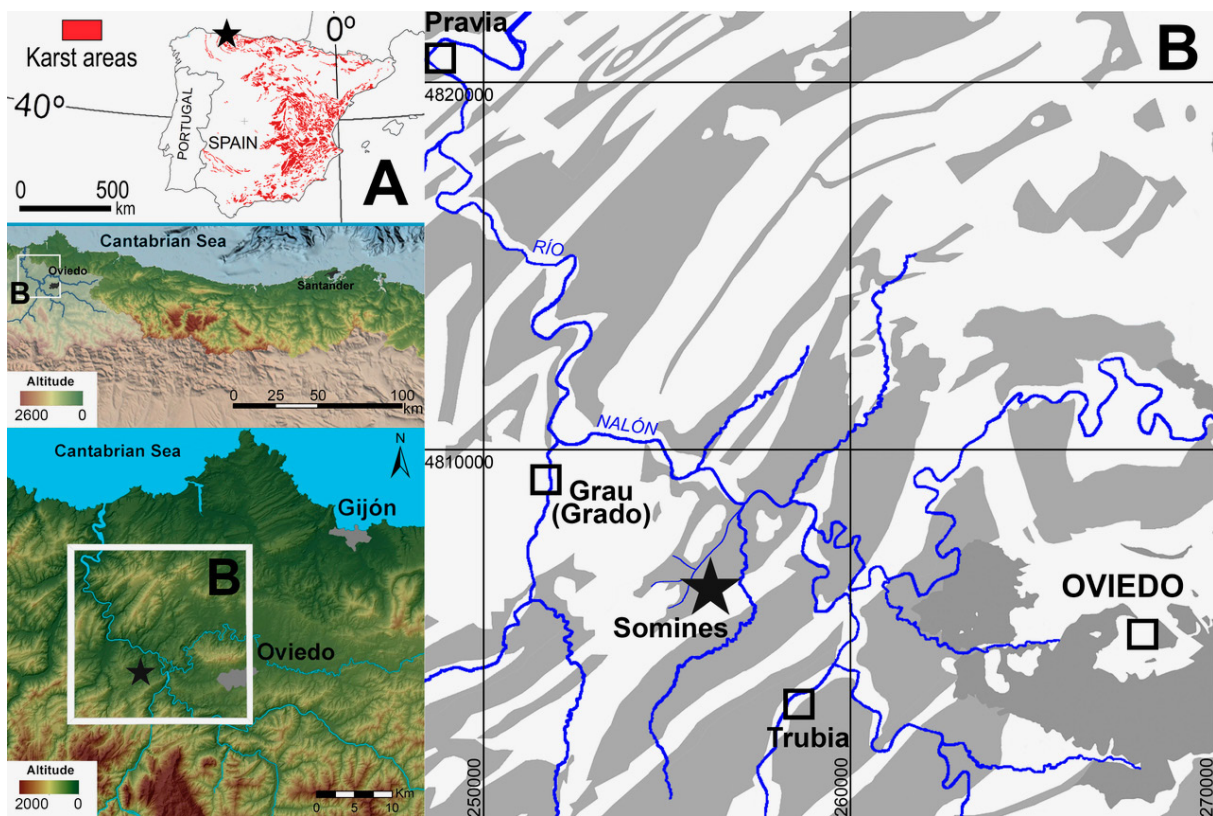


Fig. 3. Type location of *Ischyropsalis damiani* sp. nov. in Asturias region (indicated by star). **A.** Location of study area and map of the Karst in Spain at 1 : 1 000 000-scale where karstified carbonate rocks emerge (karst areas distribution is based on Geological Survey of Spain; IGME, Spanish acronym). **B.** Map of northern Iberia showing the location of the middle Nalón basin (Base DEM: (©) IGN-CNIG), the location of Elvira Cave and areas of Oviedo with bedrock in which karst processes occur (the range of limestone outcrops of Carboniferous age is represented in grey, at a scale of 1 : 50 000).

Phylogenetic analyses

The obtained sequences were reviewed and edited using Geneious Prime ver. 2023.2.1 software (Kearse *et al.* 2012). When the lower peaks were a third part of the total length of a higher peak and occurred in the reverse and forward sequences, double peaks were considered as ‘valid’.

The available sequences in GenBank for species of the genus were downloaded and included in our data matrices, along with outgroup used in Schönhofer *et al.* (2015). We built two matrices, one with the concatenation of the two genes and another one separating exon and intron (see [Supp. file 1](#)). The alignments were performed by the Multiple Sequence Comparison by Long-Expectation algorithm (MUSCLE) as implemented in Jalview ver. 2.11.1.3 (Edgar 2004; Waterhouse *et al.* 2009). To determine the best-fitting substitution, the Akaike Information Criterion (AIC) implemented in jModelTest ver. 2.1.10 was used (Akaike 1974; Darriba *et al.* 2012). The General Time Reversible model with gamma distribution (GTR+G) (Tavaré 1986; Yang 1994) was the best-fitting model for both dataset markers. Additionally, separate EF1- α datasets were generated for combined exon–intron sequences, (following Schönhofer *et al.* (2015) methodology) and independent phylogenetic analyses were performed for each dataset (COI α EF1- α exon and intron, COI α EF1- α exon only). The resulting topologies were compared with those obtained from the full dataset and are provided in [Supp. file 2](#).

Two different methods were used to determine the phylogenetic relationships within *Ischyropsalis*: Maximum Likelihood (ML) as implemented in the IQtree ver. 1.6.12 webserver (Nguyen *et al.* 2015; Trifinopoulos *et al.* 2016) and Bayesian Inference (BI) as implemented in Mr Bayes ver. 3.2 (Ronquist *et al.* 2012). The ML trees were built by creating an initial Neighbour-Joining (NJ) tree and using the Nearest-Neighbour Interchange (NNI) method for branch rearrangement (Robinson 1971; Moore *et al.* 1973). Branch support was estimated by 10 000 ultrafast bootstrap replications (BT) (Minh *et al.* 2013; Chernomor *et al.* 2016; Hoang *et al.* 2017). Two parallel runs of Markov Chain Monte Carlo (MCMC), consisting of 1 cold and 3 hot chains each, were used for the BI analyses, which ran for 10 000 000 generations and had a burning fraction of 0.25 (Hastings 1970; Altekar *et al.* 2004). The Posterior Probability (PP) was used to estimate the statistical branch support in the BI analyses (Metropolis *et al.* 1953; Hastings 1970). The Effective Sample Size (ESS) and a visual inspection of the trace plots were used to assess the convergence of the BI chains using Tracer ver. 1.7. (Rambaut *et al.* 2018). In all the analyses the ESS values were over 5000. Whenever the statistical branch support was below 50%, tree branches were collapsed.

Results

Taxonomy

Class Arachnida Cuvier, 1812
Order Opiliones Sundevall, 1833
Suborder Dyspnoi Hansen & Sørensen, 1904
Family Ischyropsalididae Simon, 1879
Subfamily Ischyropsalidinae Simon, 1879

Genus *Ischyropsalis* C.L. Koch, 1839

For logonymy: see Schönhofer (2013) and Kury *et al.* (2024).

Diagnosis (emended from Rozwalka *et al.* 2012 and Schönhofer 2013)

Defined by peculiarities of the penis glans morphology, the enlarged chelicerae with glandular fields, located distally on the basal article in males of most species and supported by molecular phylogenetic evidence. Body egg-shaped, uniformly black or brownish-black and greyish or brownish (occasionally

white). Mostly lacking dorsal armature, smooth dorsum, often glossy, mostly with scutum laminatum or parvum (rarely scutum intermedium); in few species opisthosomal areas raised to small, pointed bumps. Two or more spine-like metapeltidial structures, arranged in a transversal row. Ocularium without spine; eyes separated by distinct furrow. Chelicerae massive and enlarged, distinctly longer than the body (can be up to ca one and half to twice body length). In most species, male chelicerae are spineless or have only short piliferous tubercles; female chelicerae possess spines only. Male chelicerae with dorsal or dorso-medial secretion-extruding field indicated by bristle-field in the distal end of basal article. In most species glandular fields of males associated with knob-like apophyses. Long and thin palps. Legs of medium to long length, robust and unarmed. Club-shaped penis with simple truncus, basal two-third portion with a single intrinsic muscle. Short muscular tendon, the base of truncus divided into two short root-like structures. Glans conical to inflated. The sclerotised ventral plate is short, from rhombic to bilobed at the base. Sclerotised area of glans with several defined regions exhibiting dense fields of very thick, long and curved backward-pointing setae. Glans gradually tapering into a long stylus, which is simple and pointed, bent at the base but without contortion or additional structures. Female with very short ovipositor, not segmented, with few setae in the apical area, receptaculum seminis with numerous (4–6, rarely 10) finger-like ampullae, basally surrounded with funnel-shaped structure.

Ischyropsalis impressa Luque & Labrada sp. nov.

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Figs 1–2, 4–6, 15G

Diagnosis

A species of the westernmost *dispar*-group (see Luque & Labrada 2012) with short and slender chelicerae. The species of this group can be told apart by morphological characters and by the allopatric range. Males are discernible by the chelicerae, which show a weak distal bulge on the basal article (Figs 4D, 5B–D), as well as a mace-shaped distal end with an extended bristle area (Figs 5B, 7), and by their distinct penis morphology (Figs 5H, 6B–D). Females can be distinguished by the presence of a relatively short, or sometimes absent, dorsal spine at the distal end of the basal article, together with several large dorsal and ventral spines on the basal chelicerar article (Figs 4C, 5E–G, see arrows).

Etymology

The specific epithet is a Latin adjective in the feminine gender to agree with the generic name. It is a wordplay derived from ‘Impress’, a Santander-based consulting firm where the taxonomic studies were performed.

Type material

Holotype

SPAIN – **Cantabria Province** • ♂; Laredo, La Baja Cave; 43.39957° N, 3.42064° W; 50 m a.s.l.; 22 Aug. 2004; L. Labrada and C.G. Luque leg.; MNCN 20.02/17329.

Allotype

SPAIN – **Cantabria Province** • ♀; same collection data as for holotype; MNCN 20.02/17332.

Other paratypes (only adult specimens)

SPAIN – **Cantabria Province** • 3 ♂♂, 2 ♀♀, in five vials; same collection data as for holotype; MNCN 20.02/12858, 20.02/17330, 20.02/17331, 20.02/17334, 20.02/17335.

Additional material studied

SPAIN – **Cantabria Province** • 1 ♂, 1 ♀; Laredo; La Baja Cave; 12 Aug. 1985; CGL • 5 ♂♂, 4 ♀♀; same collection data as for preceding; 28 Mar. 1992; CGL • 3 ♂♂, 3 ♀♀; same collection data as for preceding; 22 Aug. 2004; CGL • 3 ♂♂, 5 ♀♀; same collection data as for preceding; 28 Jul. 2006; CGL • 1 ♀ juv.; Liendo, Iseca Nueva, Covacho del Cogorio Spring Cave; 43.38423° N, 3.37303° W; 22 m a.s.l.; Aug. 1985; CGL • 1 ♀; Limpias, Seña, Perelada Cave; 43.38281° N, 3.41501° W; 180 m a.s.l.; 26 Aug. 1988; CGL • 1 ♂ juv.; Limpias, Seña, Hoyo de Peñaflor Cave; 43.39613° N, 3.41477° W; 95 m a.s.l.; 26 Dec. 1992; CGL • 1 ♂, 1 ♀; Limpias, Seña, Hoyo Molino Cave; 43.37996° N, 3.41347° W; 175 m a.s.l.; 31 Apr. 1988; CGL • 1 ♀; same collection data as for preceding; 43.37996° N, 3.41347° W; 175 m a.s.l.; 31 Apr. 1993; CGL • 1 ♀; Laredo, La Baja Cave; 28 Mar. 1991; C.G. Luque leg.; CJM 3756 • 1 ♂, 1 ♀; same collection data as for preceding; CRBA 97705 • 1 ♀; Guriezo, Hoyo Menor-Rejuyo Cave; 43.33922° N, 3.36356° W; 270 m a.s.l.; 6 Mar. 1988; C.G. Luque leg.; CJM 3757 • 1 ♀; Castro-Urdiales, Sámano, La Lastrilla Cave; 43.36131° N, 3.25478° W; 60 m a.s.l.; 26 Jun. 1988; C.G. Luque leg.; CJM 3758 • 6 ♀♀; Limpias, Hoyo Molino Cave; 3 Sep. 2010; L. Labrada and C.G. Luque leg.; CGL • 1 ♂; Guriezo, Hoyo Menor-Rejuyo Cave; 9 Dec. 2010; L. Labrada and C.G. Luque leg.; CGL • 1 ♀; Guriezo, Hoyo Menor-Tocinos Cave; 43.34123° N, 3.36783° W; 290 m a.s.l.; 31 Mar. 2013; L. Labrada and C.G. Luque leg.; CGL • 1 ♀; Castro-Urdiales, Hoyo de la Cubilla Cave; 43.36319° N, 3.27513° W; 170 m a.s.l.; 31 Mar. 2013; L. Labrada and C.G. Luque leg.; CGL • 1 ♂; Rasines, Hoyo de las Reboligas Cave; 43.29214° N, 3.37477° W; 180 m a.s.l.; 9 Aug. 2013; C.G. Luque leg.; CGL.

Two other adult females were captured by the same collectors as the holotype, one of them was used for DNA extraction and sequencing with voucher number BOS-OPI-00002. Extracted specimens and DNA aliquots are deposited in the Departments of Zoology at Oviedo University.

Remarks

The material originally collected between 1985 and 1992 by Luque (1991, 1992) from La Baja, Hoyo de Peñaflor, Hoyo Molino and Perelada caves, incorrectly identified as *I. dispar*, as well as the material collected in the Hoyo Menor-Rejuyo and La Lastrilla caves, incorrectly identified as *I. magdalenae*, are here reassigned to *I. impressa* sp. nov.

Description

Measurements (mm) were taken for the holotype, allotype and paratypes (4 ♂♂, 3 ♀♀). Body length: holotype 5.04, ♂♂ paratypes 5.04–5.48, allotype 5.56, ♀♀ paratypes 5.56–5.97; length of basal chelicerae segment (paratypes in parentheses): holotype 4.76 (4.76–5.10), allotype 5.65 (5.65–5.72). Females show a wider range in body length, probably due to different stages of gravidity but their chelicerae are less variable in size than in male. Total length of leg II: 42.78 (42.78–44.32), 42.80 (42.80–44.21). Length of the leg II segments: femur: 8.88 (8.88–9.05), 9.41 (9.41–9.75); patella: 1.37 (1.37–1.42), 1.51 (1.51–1.57); tibia: 8.44 (8.44–8.98), 8.49 (8.49–8.86); metatarsus: 13.61 (13.61–13.98), 13.34 (13.34–13.73); tarsus: 10.48 (10.48–10.89), 10.05 (10.05–10.30). The measurements of the male (holotype) and female (in parentheses) femora from the first to the fourth pair of legs are 6.49 (6.91), 8.88 (9.41), 5.38 (5.61), 7.13 (7.42).

Male

PROSOMA. Low prosoma, height at the eye area about 0.85 of its length. Fine and densely roughened, brown to black, slightly ascending from the border of the second thoracic tergite, and this tergite is ornamented with a transversal row of bristles with ten or more denticles of different sizes. Eye mound weakly developed. Eyes widely separated and with small lenses (Figs 4C–D, 5A).

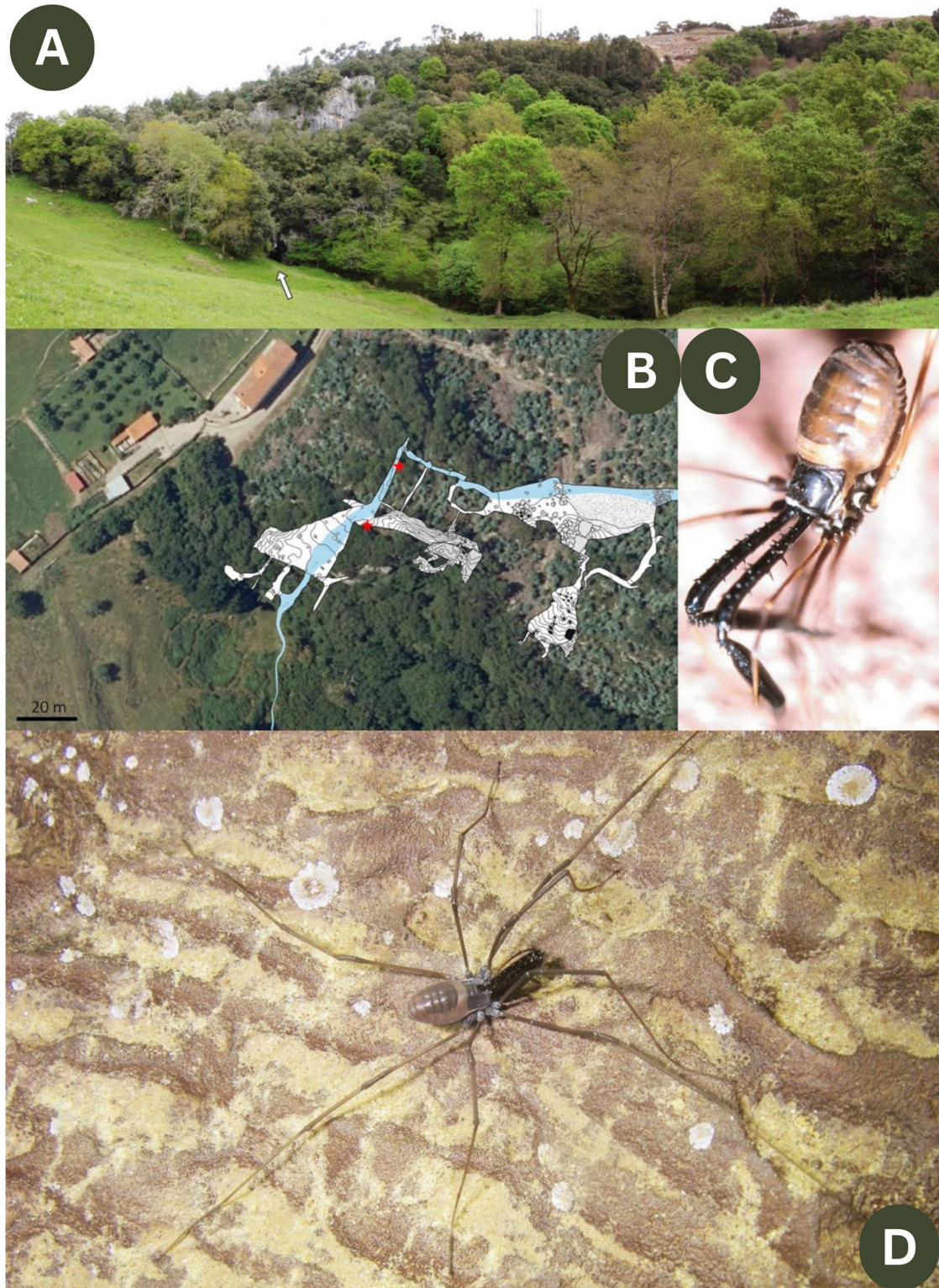


Fig. 4. Type locality and habitus of *Ischyropsalis impressa* Luque & Labrada sp. nov. **A.** Entrance to La Baja Cave (arrow). **B.** Digital orthophoto with projection of the plan view of the studied cave (surveyed August 2004–2006 by C.G. Luque and J. Ruiz-Cobo; see Ruiz-Cobo *et al.* 2008) and sampling sites of the new species (stars). **C.** Female in situ (La Baja Cave, CGL 269). **D.** Female in situ (Covacho del Cogorio Spring Cave, CGL 270). Photo collage by C.G. Luque. The final image courtesy of E. Peradalta.

OPISTHOSOMA. Sclerotised brownish areas, finely granulated and surrounded by transversal grooves among shallow tergites. Each tergite with transverse rows of inconspicuous small tubercles, each one carrying a thick and short hair. Males and females with scutum parvum (Fig. 4C–D).

CHELICERAE. Sexually dimorph. Short and slender, brown to black (can be up to ca twice body length). Male basal article gracile and smooth (Figs 5C–D, 15G) with only a few short hairs, which widens

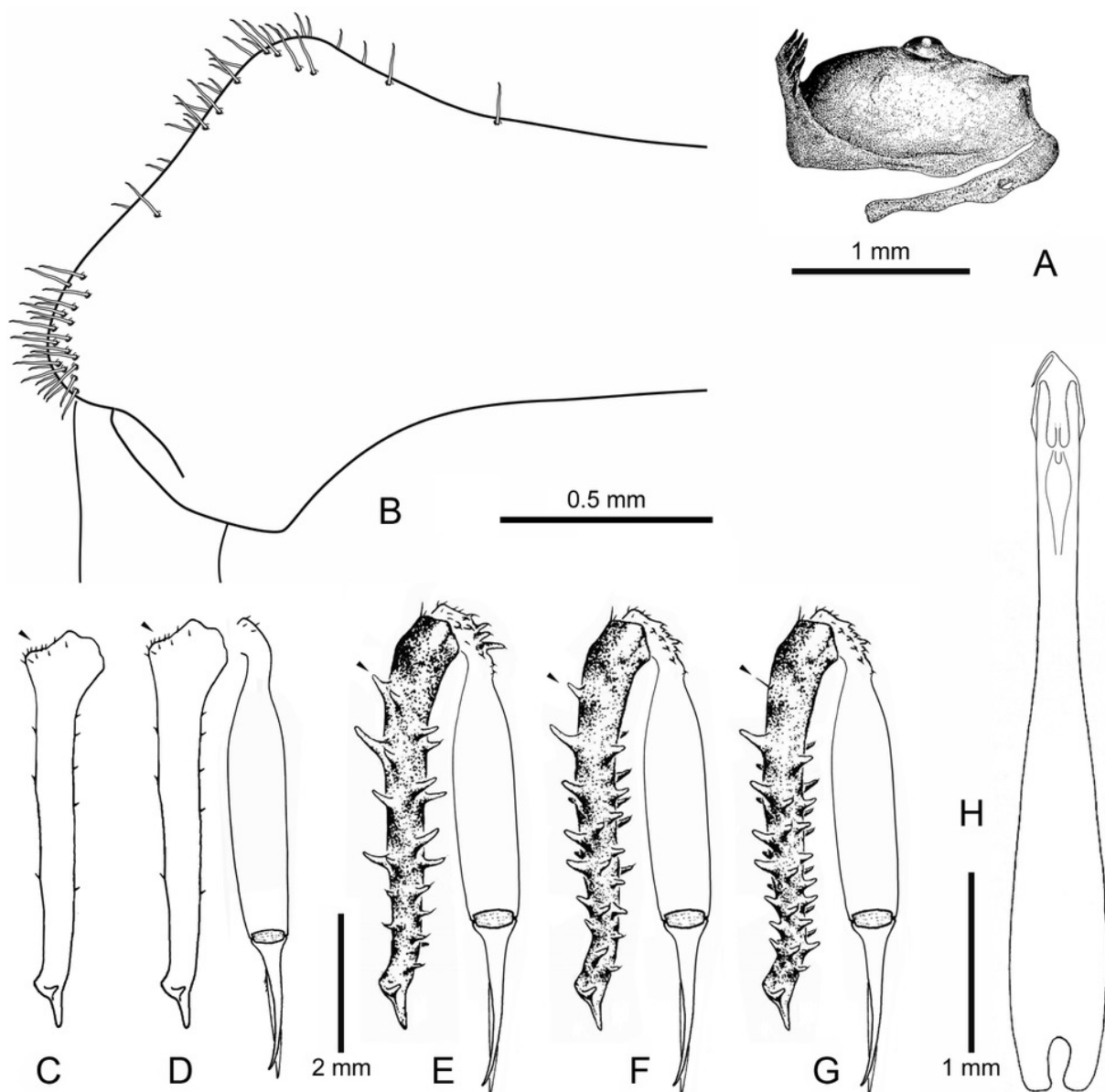


Fig. 5. *Ischyropsalis impressa* Luque & Labrada sp. nov. **A–B, H.** Holotype, ♂, from La Baja Cave (MNCN 20.02/17329). **A.** Prosoma, lateral view. **B.** Distal part of chelicerae (medial view), apophyses and cheliceral secretion-extruding-field indicated by bristle-field. **C.** Paratype, ♂, from Rasines, Hoyo de las Rebolligas Cave (MNCN 20.02/17334), chelicera. **D.** Paratype, ♂, from Limpias, Hoyo Molino Cave (MNCN 20.02/17335), chelicera. **E.** Allotype, ♀, from Laredo, La Baja Cave (MNCN 20.02/17332), chelicera. **F.** Paratype, ♀, from Guriezo, Hoyo Menor-Rejuyo Cave (MNCN 20.02/17331), chelicera. **G.** Paratype, ♀, from Castro-Urdiales, Hoyo de la Cubilla Cave (MNCN 20.02/17334), chelicera. **H.** Total view of ventral penis. Arrows indicate diagnostic characters mentioned in the text.

distally to a weakly pronounced bulge (Fig. 5C–D). The distal end of the basal article is slightly and ventrally angled, dorsally extended to a rounded apophysis (with a small glandular hairy area; Figs 5B, 6A; see Luque & Labrada 2012: fig. 38) and sloping flat to the insertion site of the distal article (medial view). Distal article is almost smooth, with only a few short hairs and rounded protuberances, bristle-like, usually short and blunt, set into a basal socket (see Luque & Labrada 2012: fig. 34).

PENIS. Total length of the penis 4.0 mm. Penis truncus penis (Fig. 5H) from base to below glans gradually narrowing. Slightly widening below glans, not dorsally bulged (dorso-lateral and dorsal views, Fig. 6B–D) but a rather straight prolongation of the remainder of the truncus. Glans with conical apex gradually narrowing into a remarkably long stylus. Glans are separated into two sclerotised areas: dorsally with numerous setae arranged into two bands with 21 setae each (Fig. 6C). It ventrally displays short and narrow sclerite, almost parallel-sided. The basal end is divided into two lobes, each bearing long and abundant but loosely distributed bristles, which become absent towards the distal part of the glans or are reduced to only minute setae (Fig. 6D).

LEGS. Light brown, slender, medium length and all segments round in cross section. Metatarsus and tarsus in lighter colour. Base of femora whitish to yellowish, just as in *I. cantabrica* (see Luque & Labrada 2012: figs 18–24). Smooth femora without sculpture elements (spines or tubercles), only with microtrichia and setae. Trochanter and coxa with several types of sculpture elements: granules, tubercles, cones and long macrotrichia, which arise from a basal socket.

Female

CHELICERAE. Basal article more robust and armed with numerous long spines of different sizes over its whole length (7–8 longer spines on dorsal surface, 5–6 ventro-medially, and 6–8 ventro-laterally; Figs 4C–D, 5E–G).

OVIPOSITOR. Bilobulate, with the ventral apex covered with few setae. The seminal receptacle has four short finger-like ampullae. In this respect, it does not differ from other species of *Ischyropsalis* in the region (see Dresco 1968: fig. 19; Martens 1969b: figs 66, 68).

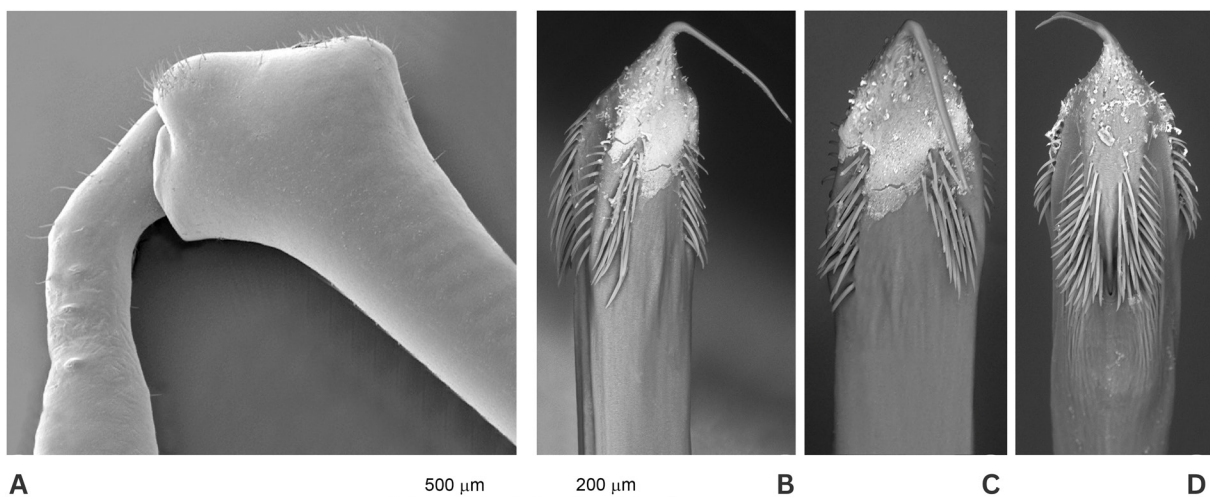


Fig. 6. *Ischyropsalis impressa* Luque & Labrada sp. nov. from La Baja Cave (Luque & Labrada 2012: figs 38–41). **A.** Paratype, ♂ (MNCN 20.02/12858), distal end of basal article with basal part of second article of chelicera, medial view. **B–D.** Paratype, ♂ (MNCN 20.02/17331), distal part of penis with glans and stylus. **B.** Dorso-lateral view. **C.** Dorsal view. **D.** Ventral view.

Variability

Although species within the *dispar*-group may exhibit some degree of intraspecific variation, such as a more developed female spination in larger specimens (Martens 1969b; Luque & Labrada 2012), the examined material of *I. impressa* sp. nov. shows limited morphological variability. While the body is sometimes depigmented but scutum parvum uniformly dark brown. Only the population located at the eastern end (e.g., in La Lastrilla Limestone Cave system) is characterized by a body and legs with reduced pigmentation and smaller eyes.

Distribution

Endemic to the northeastern coastal region of Cantabria. Restricted to a series of isolated karst areas and cave systems in the north foothills of the Cantabrian range. This area is formed by an Aptian Cretaceous limestone rock-type, called Urgonian Limestone, with a radius of less than 15 km² (Figs 1–2, 15G).

Ecology

The present records suggest that *I. impressa* sp. nov. is a troglobiont species. It has only been recorded in a deep cave environment which is inhabited due to its cool, moist and dark microclimate. All karst areas and cave systems (Hoyo de Peñaflo, Hoyo Molino, Hoyo Menor, Hoyo de la Cubilla, La Lastrilla, La Baja, Covacho del Cogorio, etc.) are found with direct access to a subterranean river (the unsaturated or vadose zone of underground).

Ischyropsalis aguerana Luque & Labrada sp. nov.

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Figs 1, 7–9, 15D

Diagnosis

A species of the westernmost *dispar*-group (see Luque & Labrada 2012) with long and thick chelicerae. The species of this group can be told apart by morphological characters and by allopatric range. Males are discernible by large distal apophyses on the basal cheliceral article, with an extended bristle area (Figs 8B–8C, 9A), and their distinct penis morphology (Figs 8E, 9B–9D). Females are discernible by the presence of five large dorsal spines on the basal chelicera article (Figs 7C, 8D, see arrows).

Etymology

The specific epithet ‘*aguerana*’ is a feminine adjective derived from Agüera, the locality where the species was found.

Type material

Holotype

SPAIN – **Cantabria Province** • ♂; Guriezo, La Pasada Cave; 43.30194° N, 3.28484° W; 440 m a.s.l.; 25 Apr. 1993; C.G. Luque leg.; MNCN 20.02/17333.

Allotype

SPAIN – **Cantabria Province** • ♀; same collection data as for holotype; 22 Apr. 1993; C.G. Luque leg.; MNCN 20.02/17336.

Other paratypes (only adult specimens)

SPAIN – **Cantabria Province** • 1 ♂; same collection data as for holotype; MNCN 20.02/12859 • 4 ♂♂, 3 ♀♀, in seven vials; Guriezo, Hoyo de las Fuentes Cave; 43.30421° N, 3.29391° W; 320 m a.s.l.; Aug. 1985; C.G. Luque leg.; misidentified as *Ischyropsalis dispar* by María Rambla; CRBA 97563 to 97567, 97569, 97570.

Additional material studied

SPAIN – **Cantabria Province** • 1 ♂, 1 ♀; Guriezo, Hoyo de las Fuentes Cave; 43.30421° N, 3.29391° W; 320 m a.s.l.; 3 Aug. 2006; CGL • 1 ♀; Guriezo, Gueriza Cave [also is known as Agueriza or Agüeriza]; 43.29012° N, 3.28729° W; 590 m a.s.l.; Oct, 1985; CGL • 1 ♀; Guriezo, La Tabla II Cave; 43.30494° N, 3.27872° W; 435 m a.s.l.; 25 Apr. 1993; CGL • 1 ♀, used for DNA extraction and sequencing; Guriezo, La Pasada Cave; Aug. 2006; C.G. Luque leg.; voucher number BOS-OPI-00001. The extracted specimen and DNA aliquots deposited in the Department of Organisms and Systems Biology at Oviedo University.

Remarks

The material originally collected in August 1985 by C.G. Luque (unpubl. material) from Hoyo de las Fuentes Cave, identified (on the label) as *I. dispar* by M. Rambla (also the material collected in April 1993 from La Pasada Cave), is here reassigned to *I. aguerana* sp. nov.

Description

Measurements (mm) were taken for the holotype, allotype and paratypes (6 ♂♂, 4 ♀♀). Body length: holotype 5.14, ♂♂ paratypes 5.14–5.42, allotype 5.68, ♀♀ paratypes 5.68–5.75; length of basal cheliceral segment (paratypes in parentheses): holotype 5.30 (5.30–5.42), allotype 5.56 (5.56–5.85). Females show a wider range in body length, probably due to different stages of gravidity but their chelicerae are less variable in size than in male. Total length of leg II: 42.81 (42.81–43.67), 43.77 (43.77–44.87). Length of leg II segments: femur: 8.87 (8.87–9.03), 9.2 (9.2–9.37); patella: 1.37 (1.37–1.41), 1.52 (1.52–1.57); tibia: 8.39 (8.39–8.49), 8.59 (8.59–8.95); metatarsus: 13.65 (13.65–14.0), 13.81 (13.81–14.13); tarsus: 10.53 (10.53–10.74), 10.65 (10.65–10.85). The measurements of the male (holotype) and female (in parentheses) femora from the first to the fourth pair of legs are 5.81 (6.25), 8.87 (9.2), 5.2 (5.63), 6.9 (7.65).

Male

PROSOMA. Low prosoma, height at the eye area about 0.85 of its length. Fine and densely roughened, brown to black, slightly ascent from the border of the second thoracic tergite, and this tergite is ornamented with a transversal row of bristles with ten or more denticles of different sizes. Eye mound weakly developed. Eyes widely separated and with small lenses.

OPISTHOSOMA. Brownish sclerotised areas, finely granulated, surrounded by transversal grooves among shallow tergites. Each tergite with transverse rows of inconspicuous small tubercles, each one carrying a thick and short hair. Males and females with scutum parvum (Fig. 7C).

CHELICERAE. Sexually dimorph. In general, long and slender, brown to black (can be up to ca twice body length). Almost smooth male basal article, with a longitudinal row of 6–7 short piliferous tubercles, ventrally, and 2–3 short piliferous tubercles, dorsally. Male basal cheliceral article widens distally to a weakly pronounced bulge (Fig. 9A). The cheliceral apophysis is large, triangular and pointed with a rounded edge (lateral view), which harbours a large glandular and hairy area (Figs 2C, 5B, 9A; see Luque & Labrada 2012: fig 54), bristle-like, short and blunt, set into a basal socket (see Luque & Labrada 2012: fig. 34). The distal article with numerous and large piliferous tubercles rounded in shape at the dorsal knee, and some short hairs at the base of the movable finger.

PENIS. Total length of the penis 4.5 mm. Penis truncus (Fig. 8E) from base to below glans gradually narrowing. Below glans slightly widening, not dorsally bulged (dorso-lateral and lateral views, Fig. 9B–D) but a rather straight prolongation of the remainder of the truncus. Glans with conical apex gradually narrowing into a remarkably long stylus. Glans separated into two sclerotised areas: dorsally with numerous setae arranged into two bands with 35 setae each (Fig. 9B), and it ventrally displays a long and narrow sclerite, almost parallel-sided. Basal part is divided into two lobes with long, abundant and dense bristles,

thicker than in other parts of the glans, missing towards the distal part of glans or minute setae only (Fig. 9C). Differences between *I. dispar* and *I. aguerana* sp. nov. penis morphology are shown in Fig. 9E–H.

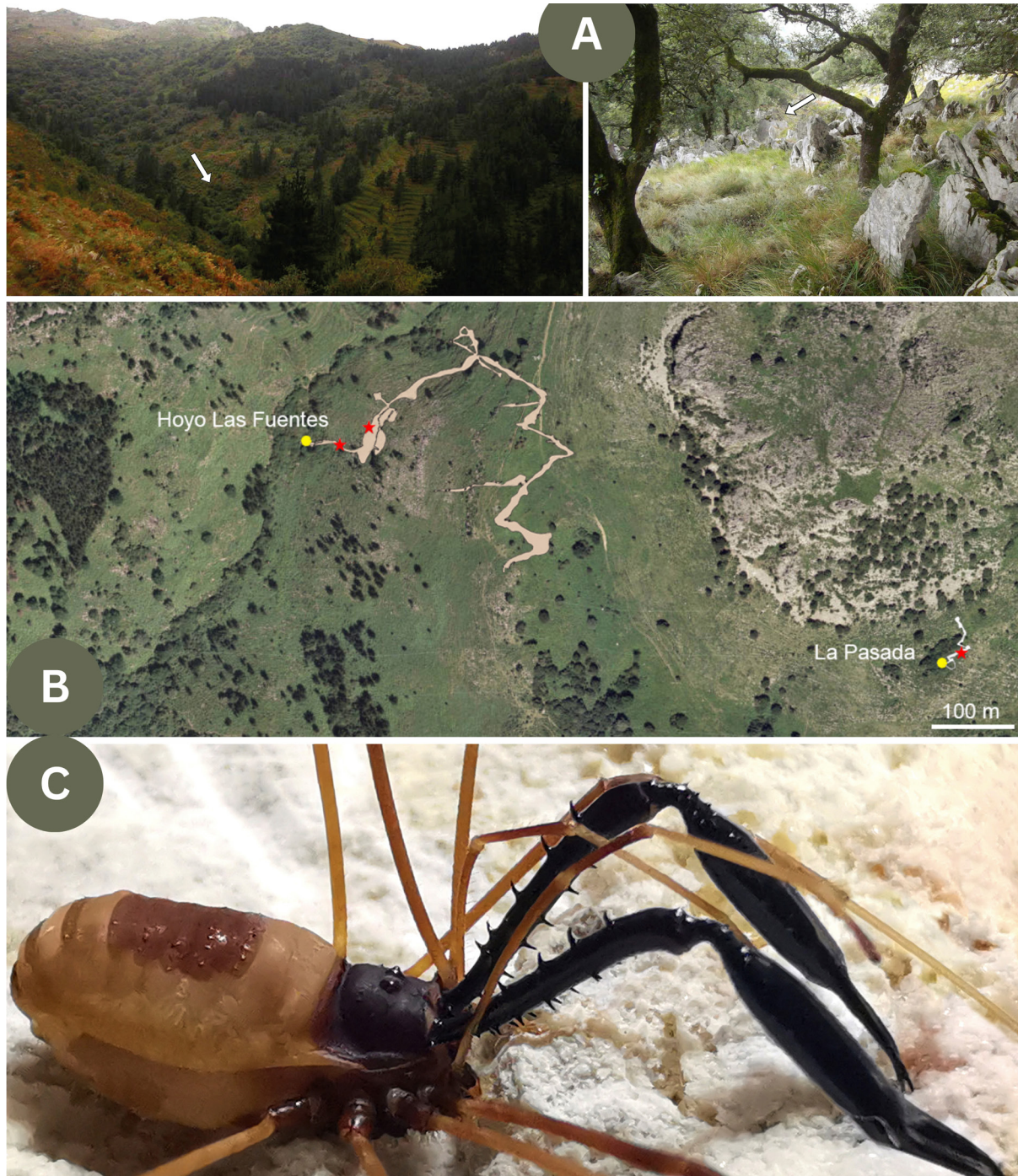


Fig. 7. Type locality and habitus of *Ischyropsalis aguerana* Luque & Labrada sp. nov. **A.** Entrance of the caves of Hoyo de las Fuentes (left) and La Pasada (right). **B.** Digital orthophoto with projection of the plan view of the studied cave (surveyed August 2004–2006 by C.G. Luque, E. Muñoz and A. Subiñas), the sampling locations of the new species (stars) and cave entrances (yellow dots). **C.** Live female from La Pasada Cave (CGL 469). Collage of photographs by C.G. Luque.

LEGS. Light brown, slender, medium length and all segments round in cross-section. Metatarsus and tarsus lighter in colour. Base of femora whitish to yellowish, and smooth femora without sculpture elements (spines or tubercles) only with microtrichia and setae. Trochanter and coxa with several types of sculpture elements: granules, tubercles and cones, and long macrotrichia, which arise from a basal socket.

Female

CHELICERAE. Female basal article more robust and armed with numerous and long spines of different sizes across its entire length (5–6 longer spines on dorsal surface, 6–7 ventro-medially, and 4–5 ventro-laterally; Figs 7C, 8D).

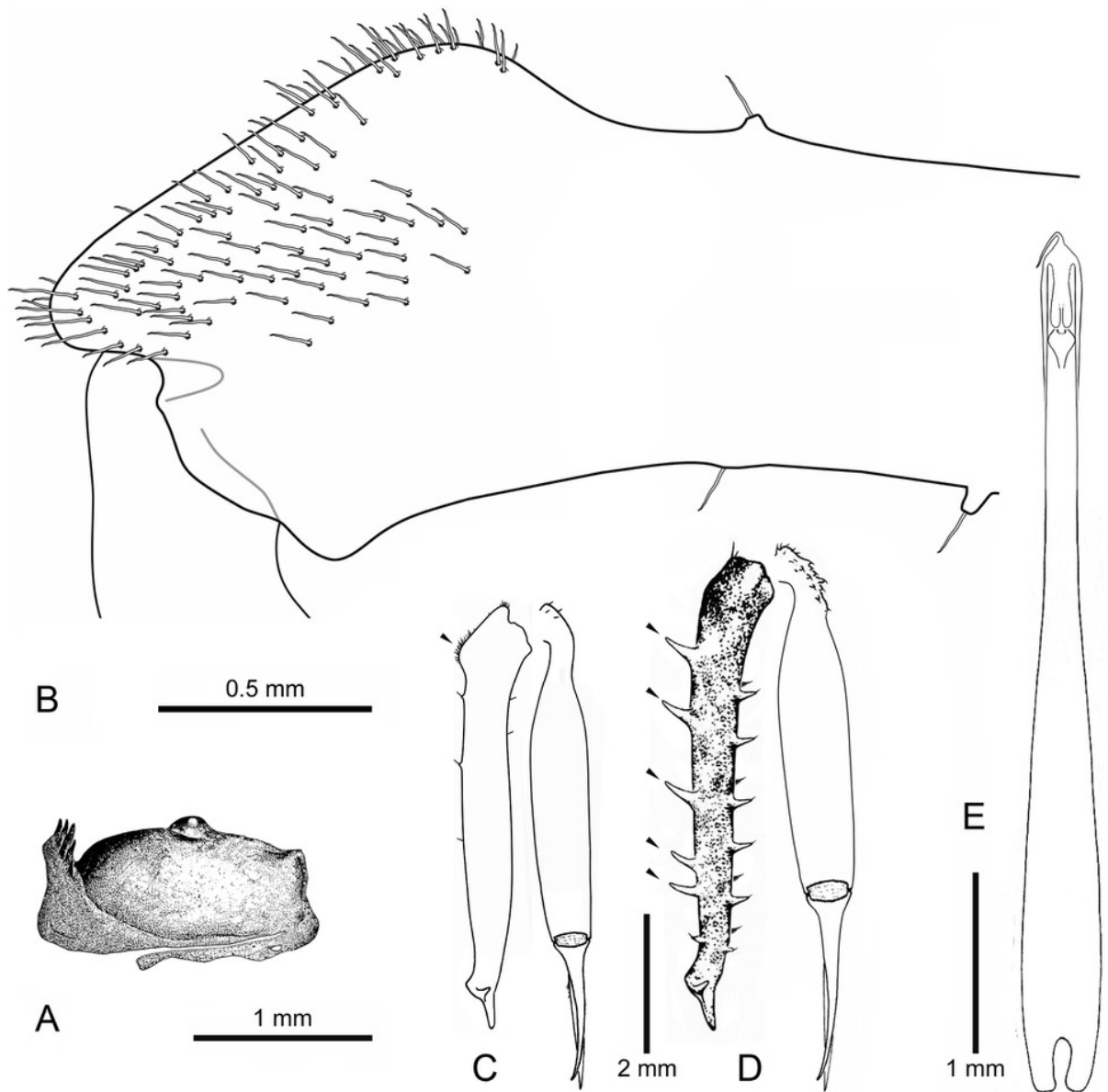


Fig. 8. A–B, E. *Ischyropsalis aguerana* Luque & Labrada sp. nov. A–B, E. Holotype, ♂ (MNCN 20.02/17333), from La Pasada Cave. C–D. Paratype from Hoyo de las Fuentes Cave (CRBA 97563), chelicera. A. Prosoma, lateral view. B. Distal part of male chelicerae (medial view), apophyses and cheliceral secretion-extruding-field indicated by bristle-field. C. Male. D. Female. E. Penis, ventral view. Arrows indicate diagnostic characters mentioned in the text.

OVIPOSITOR. Bilobulate, with the ventral apex covered with few setae. The seminal receptacle has four short finger-like ampullae. In this respect, it does not differ from other species of *Ischyropsalis* in the region (see Dresco 1968: fig. 19; Martens 1969b: figs 66, 68).

Variability

Although species within the *dispar*-group may exhibit some degree of intraspecific variation, such as more developed female spination in larger specimens (Martens 1969b; Luque & Labrada 2012), the examined material of *I. aguerana* sp. nov. shows limited morphological variability.

Distribution

Endemic to the mountainous area of the middle Agüera Basin. Only found in a particular karst area and cave system in the northern foothills of the Cantabrian range. The Agüera area corresponds to thick Urgonian reef limestones from the Cretaceous, within a radius of less than 4 km² (Figs 1–2, 15D). Legal

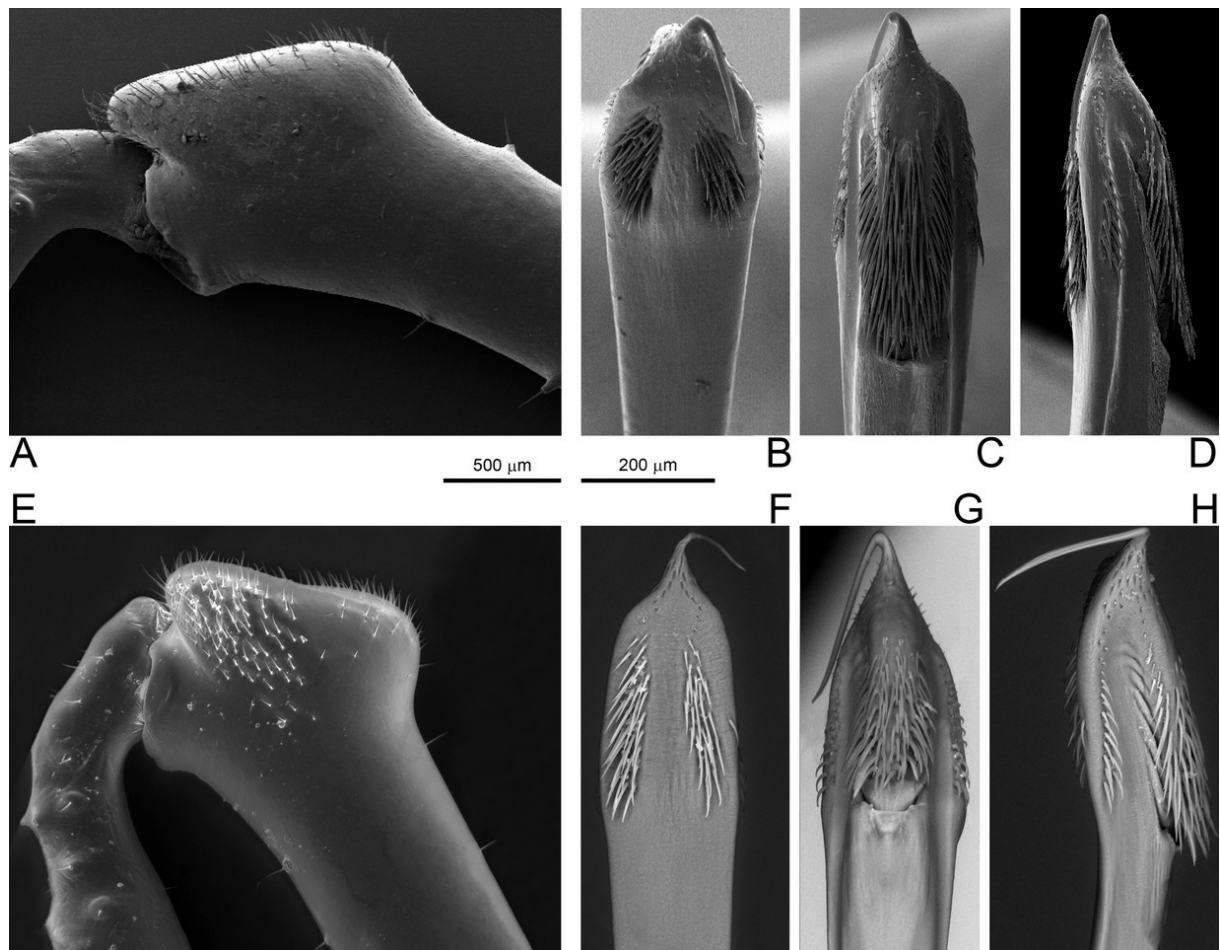


Fig. 9. *Ischyropsalis aguerana* Luque & Labrada sp. nov. **A–D.** Paratype, ♂ from La Pasada Cave, MNCN 20.02/12859 (type locality: Agüera, Guriezo, Cantabria); see also Luque & Labrada 2012: figs 54–57). **E–H.** *Ischyropsalis dispar* Simon, 1872 from Albia Cave, MNCN 20.02/17337 (type locality: 42.96869° N, 3.07397° W, 810 m a.s.l., Villalba de Losa, Burgos, 19 Jul. 2008, C.G. Luque & L. Labrada leg. **A, E.** Male cheliceral apophysis on distal end of basal article with basal part of second article of right chelicera (medial view) and cheliceral secretion-extruding-field indicated by bristle-field. **B–D, F–H.** Distal part of penis with glans and stylus. **B, F.** Dorsal view. **C, G.** Ventral view. **D, H.** Lateral view.

ownership of the Agüera area, in dispute with neighbouring Biscay since 1552, has been assigned to Cantabria since 2008 (Baró-Pazos 2010).

Ecology

The present records suggest that *I. aguerana* sp. nov. is a troglobiont species. It has only been recorded in a deep cave environment which is inhabited due to its cool, moist and dark microclimate. Hoyo de las Fuentes is found with a direct access to a subterranean river (the unsaturated or vadose zone of underground).

Ischyropsalis damiani sp. nov.

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Figs 10–13

Diagnosis

A species from the *nodifera*-group (see *I. nodifera* synonymized species in Martens 1969b) with short and slender chelicerae. The species of this group can be told apart by morphological characters and by allopatric range. Males are discernible by the distal flat pronounced bulge on the basal cheliceral article (Figs 11–12), the mace-shaped distal end with a narrow bristle area (Fig. 11A) and by their distinct penis morphology (Figs 11F, 13). Females are discernible by the presence of short dorsal spines on the distal end of the basal article and the presence of several large dorsal and ventral spines on the basal cheliceral article (Fig. 11E).

Etymology

The specific epithet ‘*damiani*’ is a patronym formed as a noun in the genitive case, honoring Damián García Tudelilla in recognition of his friendship with the first author.

Type material

Holotype

SPAIN – Asturias Province • ♂; Somines, Grado, Elvira Cave; 43.36943° N, 6.01764° W; 100 m a.s.l.; 2 Oct. 2023; R. López-Alonso, C. González-Toral and E. Pascual-Parra leg.; BOS-OPI-00007.

Allotype

SPAIN – Asturias Province • ♀; same collection data as for holotype; BOS-OPI-00008.

Other paratypes (only adult specimens)

SPAIN – Asturias Province • 10 ♂♂, 3 ♀♀, in 14 vials; same collection data as for holotype; BOS-OPI-00003 to BOS-OPI-00006, BOS-OPI-00009 to BOS-OPI-00017.

Genetic vouchers of *I. damiani* sp. nov. correspond to specimens E1 (BOS-OPI-00003), E2 (BOS-OPI-00004), E3 (BOS-OPI-00005), E11 (BOS-OPI-00014), E12 (BOS-OPI-00015), and E14 (BOS-OPI-00017).

Description

Measurements (mm) were taken for the holotype, female allotype and paratypes (11 ♂♂, 4 ♀♀). Body length: holotype 5.28, ♂♂ paratypes 5.33–6.03, female paratype 6.54, ♀♀ paratypes 6.55–6.99; length of basal chelicerae segment (paratypes in parentheses): holotype 3.84 (3.82–4.07), female paratype 4.36 (3.71–4.46). Females show a wider range in body length probably due to different stages of gravidity, but their chelicerae are less variable in size than in male. Total length of leg II: 29.16 (28.24–33.08), 28.79 (27.73–30.58). Length of the leg II segments: femur: 6.81 (5.81–7.04), 6.04 (5.67–6.74); patella: 1.55 (1.38–1.71), 1.66 (1.55–1.64); tibia: 5.59 (5.15–5.95), 5.66 (4.96–5.95); metatarsus: 8.04 (7.57–8.92),

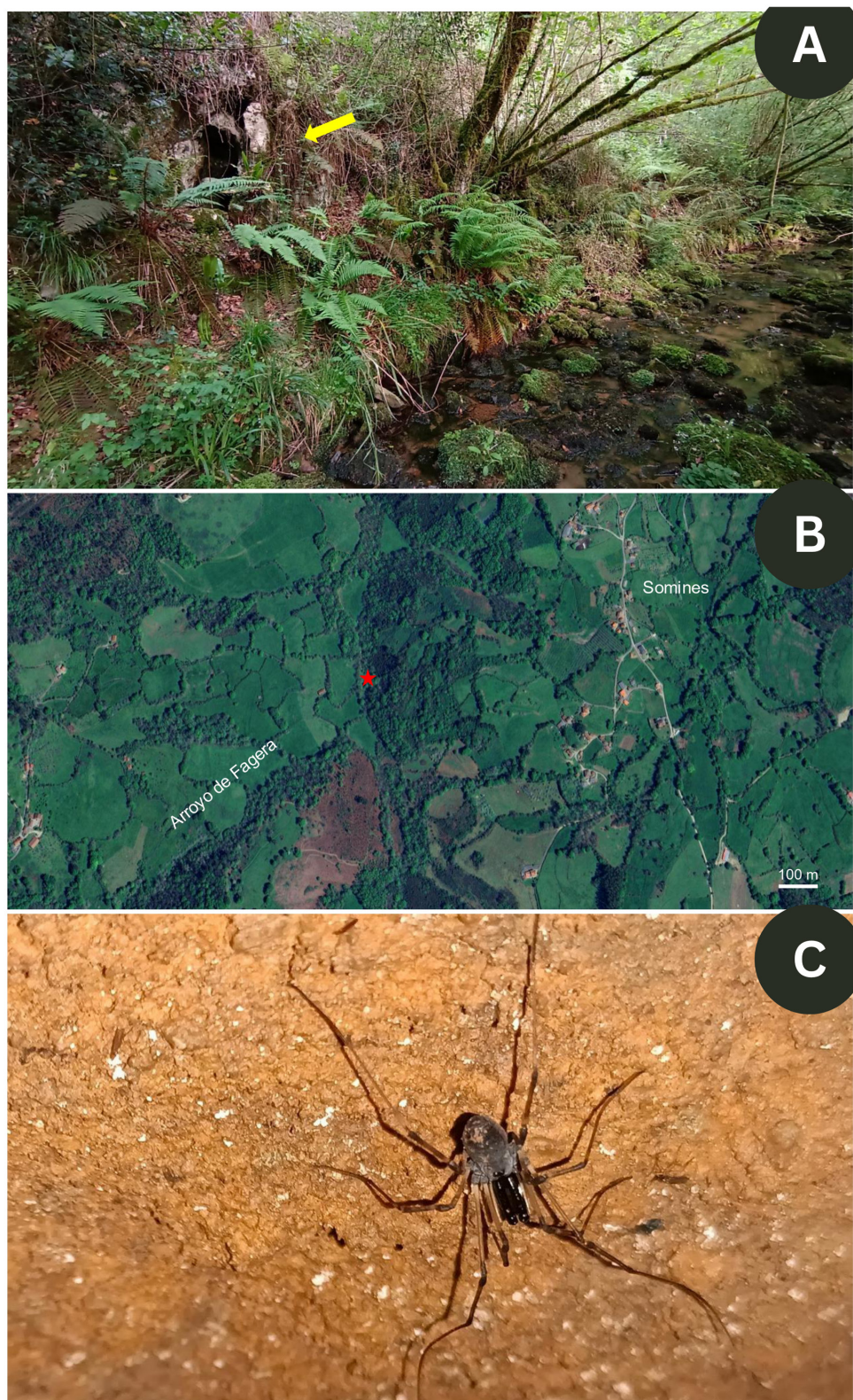


Fig. 10. Type locality and habitus of *Ischyropsalis damiani* sp. nov. **A.** Entrance of the Elvira Cave. **B.** Digital orthophoto with projection of the plan view of the studied cave (surveyed August 2022 by R. López-Alonso & C. González-Toral), the sampling site of the new species (star). **C.** Live male from Elvira Cave.

7.65 (7.53–8.49); tarsus: 7.55 (7.52–8.92), 7.44 (7.44–7.76). The measurements of the male (holotype) and female (in parentheses) femora from the first to the fourth pair of legs are 4.75 (4.32), 6.81 (6.32), 3.85 (3.52), 5.61 (5.11).

Male

PROSOMA. Low prosoma, height at the eye area about 0.8 of its length. Fine and densely roughened, grey to black, slightly ascent from border of second thoracic tergite. This tergite ornamented with a transversal row of bristles with ten or more denticles of different sizes. Eye mound weakly developed, eyes widely separated, with small lenses (Fig. 11B).

OPISTHOSOMA. Dark grey sclerotized areas, finely granulated, surrounded by transversal grooves between shallow tergites. Each tergite with transverse rows of inconspicuous small tubercles, each one carrying a thick and short hair. Males and females with scutum parvum (Fig. 10C).

CHELICERAE. Sexually dimorph. In general, short and slender, grey to black (can be up to ca twice body length). Gracile and smooth male basal article (Figs 11C, 12B) with only few short hairs, which widens

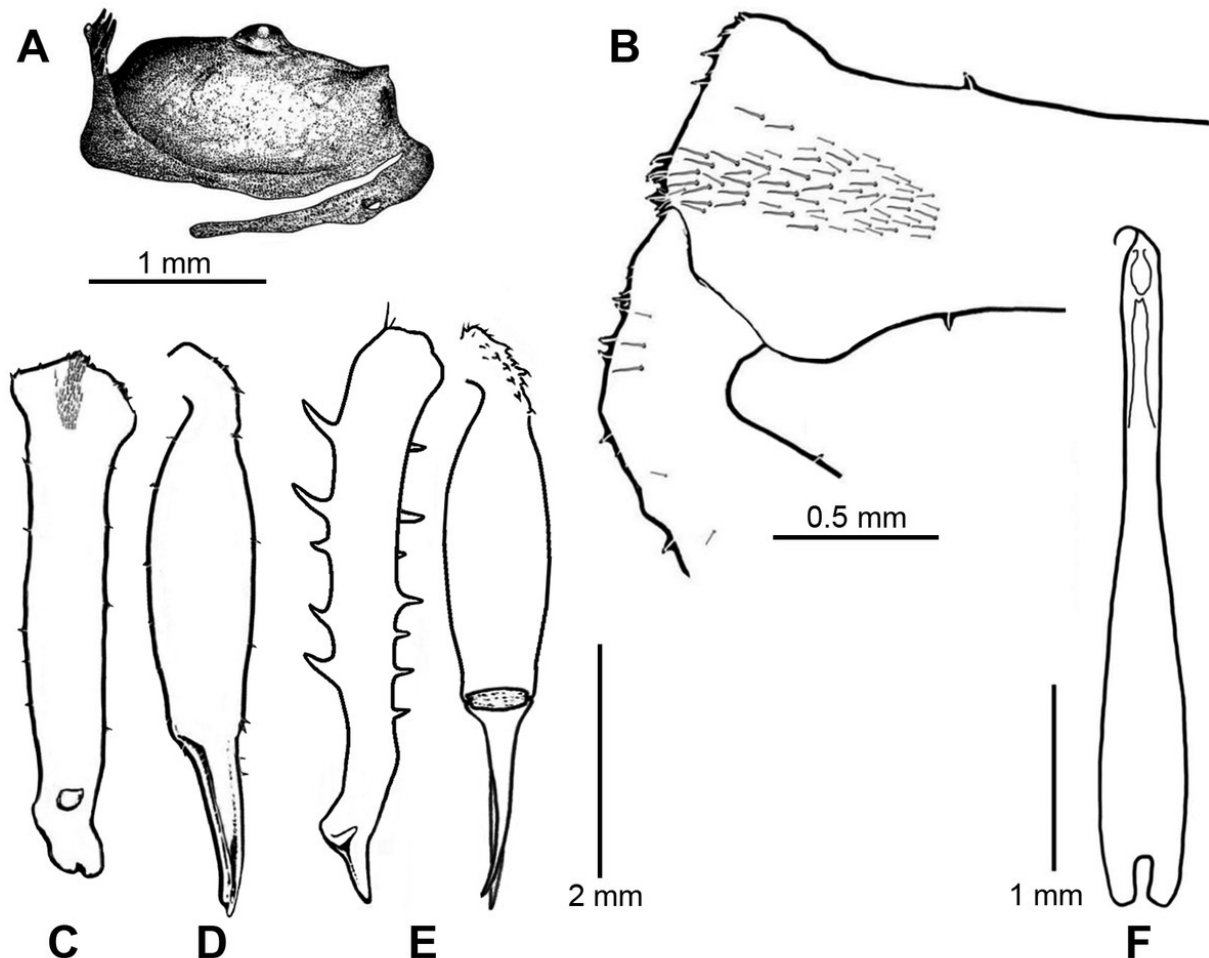


Fig. 11. *Ischyropsalis damiani* sp. nov. **A–D, F.** Holotype, ♂ (BOS-OPI-00007), from Elvira Cave. **E.** Allotype, ♀ (BOS-OPI-00008). **A.** Prosoma, lateral view. **B.** Distal part of chelicerae (medial view), apophyses and cheliceral secretion-extruding-field indicated by bristle-field. **C–E.** Chelicera. **F.** Total view of ventral penis.

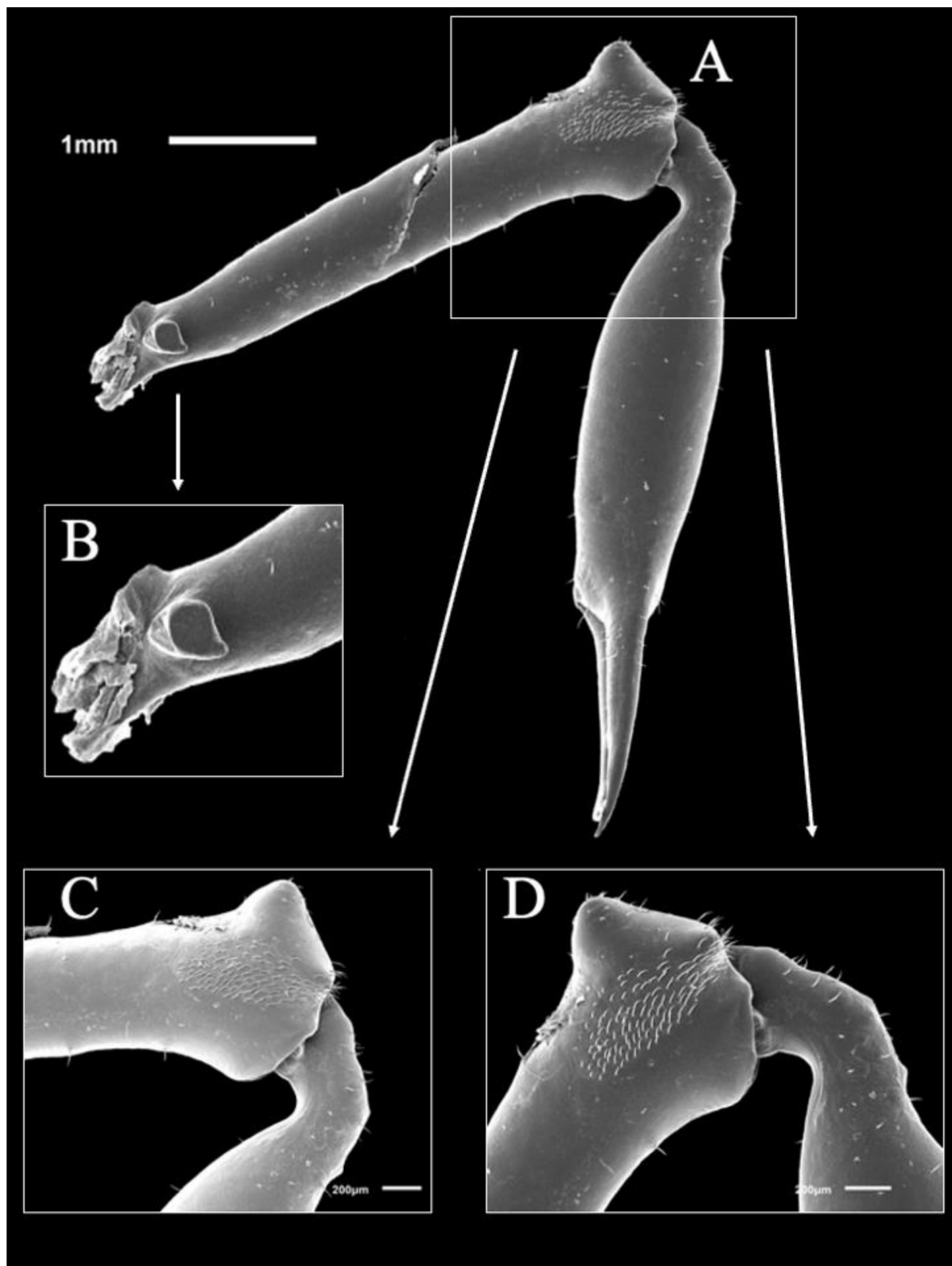


Fig. 12. *Ischyropsalis damiani* sp. nov., paratype, ♂ (BOS-OPI-00003), from Elvira Cave (type locality, Somines, Grado, Asturias). **A.** Male chelicera. **B.** Proximal part of male chelicera. **C–D.** Cheliceral secretion-extruding-field indicated by bristle-field.

distally to a pronounced bulge (Figs 11A, 12C–D). Male basal cheliceral article widens distally to a weakly pronounced bulge (Fig. 12A–D). The cheliceral apophysis is large, triangular and pointed with a rounded edge (lateral view), which harbours a medium-sized and wide glandular and hairy area (Figs 11A, 12A–D), bristles-like, short and blunt, set into a basal socket (see Luque & Labrada 2012). The distal article with numerous and large piliferous tubercles, and rounded shape at the dorsal knee (Figs 11A, 12C–D) and some short hairs at the base of the movable finger.

PENIS. Total length of the penis 4.8 mm. Penis truncus (Fig. 11F) from base to below glans gradually narrowing. Below glans slightly widening, not bulging dorsally (dorso-lateral and dorsal views, Fig. 13) but a rather straight prolongation of the remainder of the truncus. Glans with conical apex gradually narrowing into a long stylus. Glans separated into two sclerotised areas: dorsally with numerous setae (Fig. 13). The basal end divided into two lobes with long, abundant and dense bristles, with numerous setae towards the distal part of the glans (Fig. 13).

LEGS. Slender, of medium length, all segments rounded in cross section. Light grey metatarsus. Yellowish tarsus and base of femora. Smooth femora without sculpture elements (spines or tubercles), only with microtrichia and setae. Trochanter and coxa with several types of sculpture elements: granules, tubercles and cones, and long macrotrichia, which arise from a basal socket.

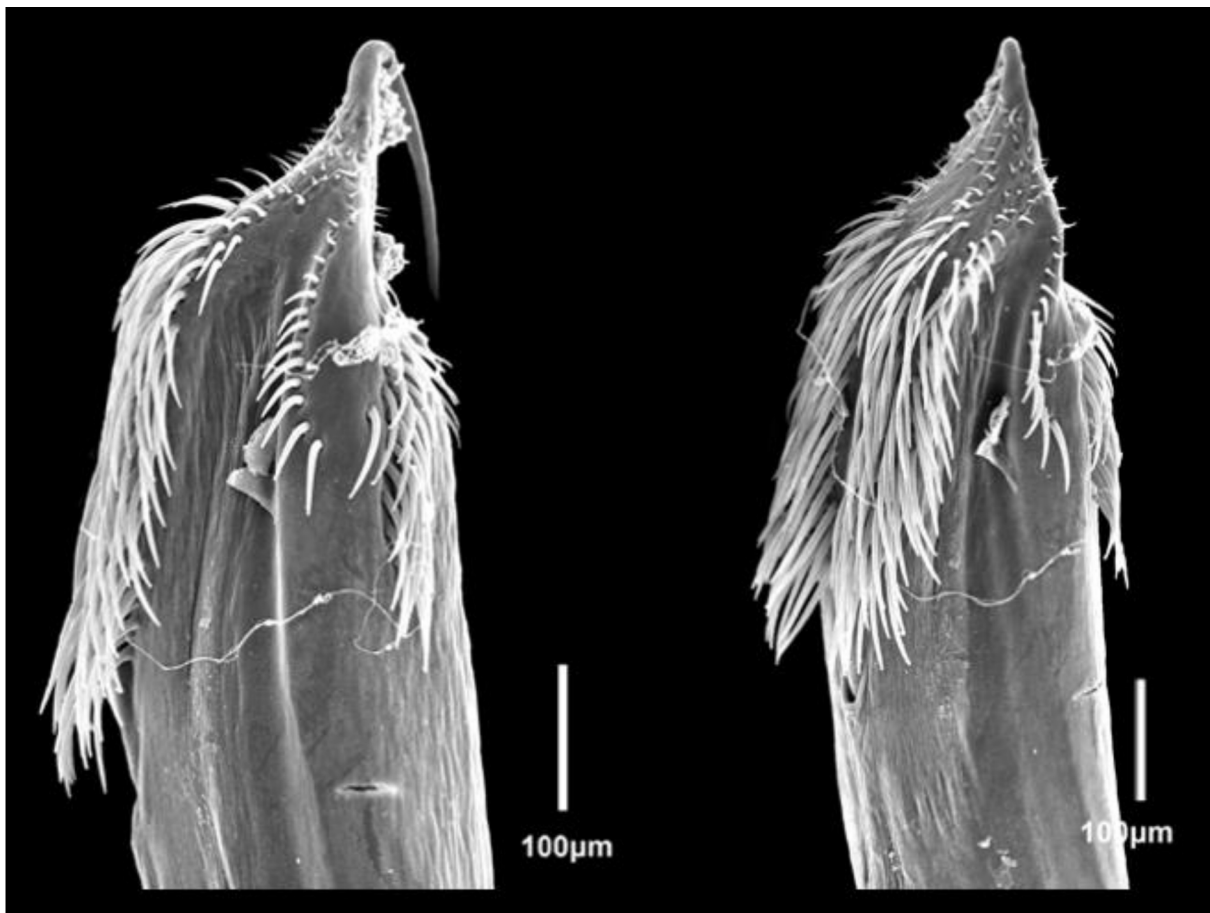


Fig. 13. *Ischyropsalis damiani* sp. nov., paratype, ♂ (BOS-OPI-00003), from Elvira Cave (type locality, Somines, Grado, Asturias). Distal part of penis with glans and stylus: frontal and lateral view.

Female

CHELICERAE. Female basal article more robust and armed with numerous and long spines of different sizes across its whole length (3–5 longer spines on dorsal surface, 4–6 ventro-medially, and 3–4 ventro-laterally).

OVIPOSITOR. Bilobulate, with the ventral apex covered with few setae. The seminal receptacle has four short finger-like ampullae. In this respect it does not differ from other species of *Ischyropsalis* in the region (see Dresco 1968: fig. 19; Martens 1969b: figs 66, 68).

Variability

Although species within the *nodifera*-group show notable interspecific differences (Luque & Labrada 2012), no extraordinary intraspecific variation was observed in the examined material. For example, the female spination, which may be more developed in larger specimens (Martens 1969b: 222–224), did not display unusual variability. The body is sometimes depigmented but the scutum parvum is uniformly light grey.

Comparative remarks

The differences with *Ischyropsalis asturica* Roewer, 1950, which is known from a single female specimen collected in Oviedo, Asturias, have also been taken into account. Although *I. asturica* is only represented by a single specimen, our material (n = 14) displays a denser cheliceral armature (with more spines) and a slenderer cheliceral shape. Martens (1969: fig. 49h, vial with code SMF/RII/5042) synonymized *I. asturica* with *I. nodifera*, recognizing several nominal species because of the high variability of cheliceral morphology within *I. nodifera*. Given the well-known morphological similarity among *Ischyropsalis* females and the unresolved species boundaries within the genus, we have carefully evaluated these differences. In addition to the consistent morphological distinctions, our species is also genetically distinct from *I. nodifera*, further supporting its recognition as a new and separate species.

Distribution

Endemic to the central region of Asturias, restricted to an isolated karst area and cave in the central part of Asturias. This area is formed by a Carboniferous limestone rock-type, called Grey or Mountain Limestone (Fig. 3).

Ecology

The present records suggest that *I. damiani* sp. nov. is a troglophile species. It has only been recorded in the cave environment which is cool, moist and dark. The cave, approximately five metres long, is notable because its passage runs parallel to the course of the Faguera stream (Fig. 10A).

Phylogenetical analyses

The COI α EF1- α dataset consisted of 21 different taxa and 35 different sequences, with a length of 963 pb, 206 parsimonious-informative sites (Table 1).

The COI α EF1- α analyses retrieved topologies (Fig. 14) in which *Ischyropsalis* was monophyletic (BS-ML:100; BI-PP:100). Within this topology, the Iberian-Alpine clade (BI-PP: 76), formed by COI α EF1- α -III, COI α EF1- α -IV, COI α EF1- α -V and COI α EF1- α -VI; the *manicata*-group (BS-ML: 81; BI-PP: 94), comprising COI α EF1- α -II and the basal *hellwigii*-group, formed by COI α EF1- α -I (BS-ML: 97; BI-PP: 100), could be found. Within the Iberian-Alpine (BI-PP: 76), the Iberian subclade (BI-PP: 84) formed by COI α EF1- α -VI; and the Alpine subclade (BS-ML: 67; BI-PP: 100), comprising COI α EF1- α -III, could also be distinguished. Within the retrieved Iberian subclade, the supported Iberian Pyrenean subclade (BI-PP: 99), formed by COI α EF1- α -IV and the supported Iberian Cantabrian subclade (BS-ML: 100; BI-PP: 100), comprising COI α EF1- α -V is obtained. Within the latter subclade, individuals from *I. damiani*

Table 1. Main features of the mitochondrial *Cytochrome Oxidase Subunit I* (COI), *Elongation Factor 1 Alpha* (EF1- α) and Combined (COI $\times$ EF1- α) alignments used for the *Ischyropsalis* C.L. Koch, 1839 phylogenetic analyses. The alignment positions that contain at least two different nucleotides were considered as variables, while when each of the different nucleotides occurring in apposition had a minimum frequency of two, those positions were considered parsimonious-informative.

Combined dataset (COI $\times$ EF1- α)	
Number of <i>Ischyropsalis</i> taxa	35
Number of sequences	21
Sequences length range (pb)	833–942
Alignment length (pb)	963
(C+G) %	40.3
Conserved sites	638
Variable sites	316
Parsimony-informative sites	206

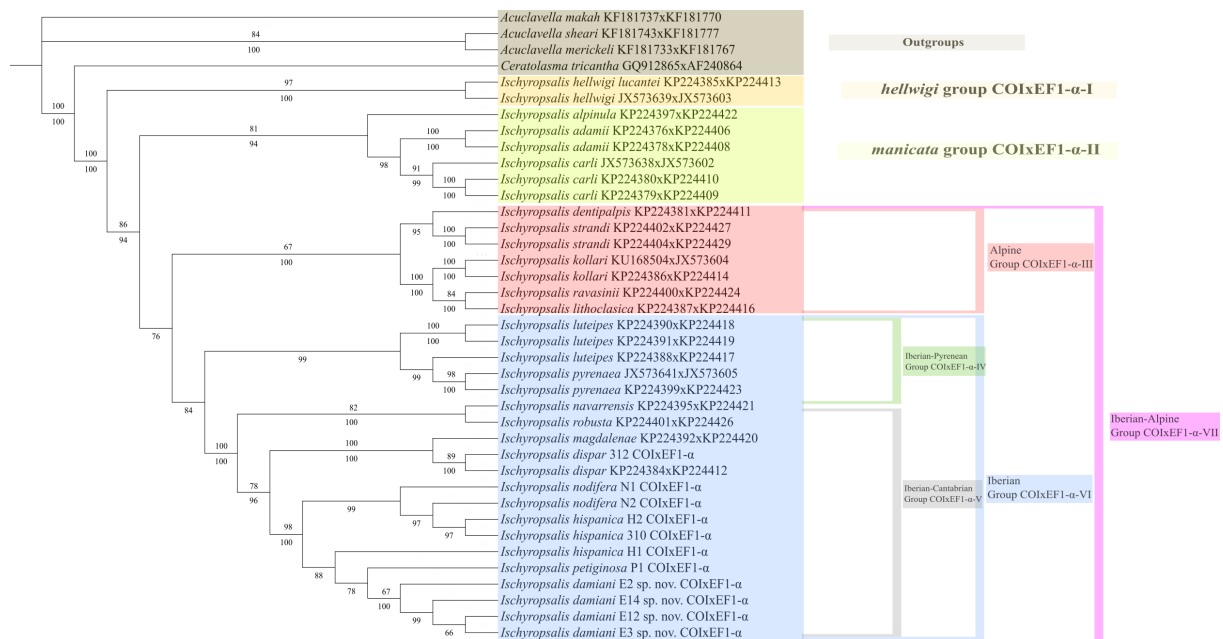


Fig. 14. Consensus topology obtained from the Maximum Likelihood (ML) and Bayesian Inference (BI) analyses of the COI $\times$ EF1- α dataset. Values beneath the branches indicate the Posterior Probability (PP) support from the BI analysis, while values above the branches represent the ultrafast bootstrap (BT) support from the ML analysis.

sp. nov. (BS-ML: 67; BI-PP: 100) and *I. dispar* (BS-ML: 89; BI-PP: 100) formed species-specific clades. The *I. petiginosa* individual has a moderate to sister relationship with the *I. damiani* exclusive clade (BI-PP: 78). Similarly, the *I. magdalenae* individual was closely related to the *I. dispar* exclusive clade with high support (BS-ML:100; BI-PP:100). On the other hand, the *I. nodifera* sequenced individuals formed a species-exclusive clade with *I. hispanica* (BI-PP:99). The other sequences of *I. hispanica* formed a species exclusive clade (BI-PP: 88).

In the present study, the discovery of *Ischyropsalis damiani* sp. nov. and its clear molecular and morphological differentiation from closely related taxa supports the proposal of a new *nodifera*-group, including *I. hispanica*, *I. petiginosa* and *I. nodifera*.

Discussion

The use of phylogenetic methods based on nuclear and mitochondrial markers (Prieto *et al.* 2013; Schönhofer *et al.* 2015), together with male genital and cheliceral morphology (e.g., Martens 1978; Schmidt *et al.* 2024), may clarify the taxonomy of *Ischyropsalis*. In this study, morphological data and molecular methods based on *COI* and *EFL-α* have provided new insights into the taxonomy of the Cantabrian taxa of *Ischyropsalis*. Our results support the existence of 16 species of *Ischyropsalis* occurring exclusively in the Iberian Peninsula, including three new species (*I. damiani* sp. nov., *I. impressa* sp. nov. and *I. aguerana* sp. nov.), which were placed within the Iberian clade sensu Schönhofer *et al.* (2015). Furthermore, the COIxEFL-α topologies supported the existence of the Iberian Pyrenean subclade, which includes species occurring in the Pyrenean (including *I. luteipes*, which also occurs in the French Massif Central (Prieto *et al.* 2013)), and the Iberian Cantabrian subclade, with species belonging to the *dispar*-group, *nodifera*-group and our newly sequenced species (*I. hispanica*, *I. petiginosa* and *I. damiani*). Compellingly, the *I. luteipes* western-pyrenees individuals (KP224419, KP224418) formed a subclade separate from the mid-pyrenees individual (KP224417). Given the complex nature of the phylogenetic relationships within this genus, this deserves further study.

The morphological analysis of the Iberian taxa revealed that their cheliceral apophysis is generally large and often triangular and pointed with a rounded edge (in lateral view), while the female chelicerae is provided with numerous spines of variable sizes. Furthermore, a well-marked character in *I. impressa* sp. nov. and *I. aguerana* sp. nov. is a divided two-lobed sclerite of the glans. According to this morphological character, the new species from Cantabria (*I. aguerana* and *I. impressa*) would belong to the *dispar*-group sensu Schönhofer *et al.* (2015), which comprises the Iberian-Cantabrian taxa (*I. dispar*, *I. magdalenae*, *I. cantabrica*, *I. noltei* and *I. gigantea*), which is also supported by the phylogenetic position of *I. aguerana* and *I. impressa* in the COI-based topology. There are also morphological differences between *I. aguerana* and *I. impressa* with the *dispar*-group sensu Schönhofer *et al.* (2015) and in particular with the northern Spanish taxa studied by Luque & Labrada (2012), which allows these taxa to be distinguished based on morphology, including the shape of the glans penis. For example, on the dorsal side, the glans has a mass of bristles at its end, arranged into two separate vertical rows that have a particular number and position for each species (Figs 2, 6, 9): *I. navarrensis* bears only 17 setae at each side, *I. impressa* 21 setae, *I. cantabrica* 25 setae, *I. magdalenae* 29 setae, *I. dispar* 31 setae and *I. aguerana* 35 setae.

Within the Iberian-Cantabrian clade, two main morphological groups can be recognized: the *dispar*-group and the *nodifera*-group. The *dispar*-group includes *I. dispar*, *I. magdalenae*, *I. noltei*, *I. cantabrica*, *I. aguerana* sp. nov. and *I. impressa* sp. nov. while the *nodifera*-group includes *I. nodifera*, *I. hispanica*, *I. petiginosa* and *I. damiani* sp. nov. *Ischyropsalis impressa* and *I. aguerana* share diagnostic characters, such as the morphology of the chelicerae and the glans, with the *dispar*-group as defined by Schönhofer *et al.* (2015). In these species, the chelicerae apophysis is large, triangular and pointed, with a rounded

edge in lateral view, and the male penis glans bears a dorsal mass of bristles arranged into two distinct vertical rows, with species-specific numbers of setae.

By contrast, *I. damiani* sp. nov., is highly similar to other *nodifera*-group taxa. The cheliceral apophysis and penis glans morphology are highly similar across species in this group, as previously noted by Schönhofer *et al.* (2015) making species delimitation based solely on morphology difficult without supporting molecular data. This morphological overlap led Martens (1969b) to synonymize *I. hispanica* and *I. petiginosa* with *I. nodifera*, while Prieto (1990) later reinstated *I. hispanica* as a valid species, treating it as distinct from *I. nodifera*, while Luque (1991) later reinstated *I. petiginosa* as a valid species, treating it as distinct from *I. nodifera*.

In addition to this, *I. damiani* sp. nov. also presents morphological similarities with this group of species, especially regarding the glans morphology. All these taxa belong to the Iberian-Cantabrian subclade, whose internal clades are rarely species-specific when only a single marker is used to build the topology, a pattern also reported by Schönhofer *et al.* (2015) in their first phylogeny of *Ischyropsalis*. The species-specific clades recovered in the combined COI α EF1- α dataset correspond to *I. petiginosa* and *I. damiani*. Interestingly, all of them have a close phylogenetic relationship, especially in the case of *I. petiginosa* and *I. damiani*. However, the fact that these two taxa present two different life-styles, as *I. petiginosa* has an epigean behaviour (Luque 1991; Luque & Labrada 2012), further supports our view that both are valid species.

A distinct clade is also recovered for *I. hispanica* together with another one for *I. nodifera*, although one *I. hispanica* specimen clusters alone in the COI α EF1- α dataset. The systematic status of *I. nodifera* and *I. hispanica* remains contentious due to overlapping morphological characters and conflicting interpretations. SEM analyses (Schönhofer *et al.* 2015) revealed striking similarities in chelicerae microstructure and bristle field, supporting Martens' synonymy hypothesis (Martens 1969b) and underscoring the morphological cohesion of the *nodifera*-group. In this context, a detailed integrative reassessment of other nominal taxa, such as *I. asturica* (junior subjective synonym of *I. nodifera*), including topotypic sampling and molecular data, would be highly desirable to clarify their taxonomic status and to resolve long-standing uncertainties within the group.

The existence of two Iberian subclades (the Iberian Pyrenean and the Iberian Cantabrian ones) suggests two different biogeographic lineages: one occurring in the mountainous systems of the Eastern Iberian Peninsula (Schönhofer *et al.* 2015) and another whose members are present in the mountainous systems of the Western Iberian Peninsula (Luque & Labrada 2012). These two sister lineages have a close phylogenetic relationship with the Alpine clade, thus suggesting that the glacial-interglacial cycles played an important role in the separation of these three lineages.

Climatic oscillations during the Pleistocene, particularly the glacial-interglacial cycles, are known to have played a major role in shaping patterns of diversification in arachnids due to their restricted mobility (Pinto-Da-Rocha *et al.* 2007). Glacial advances and retreats repeatedly fragmented and isolated populations, promoting allopatric speciation across a wide range of habitats. Although these processes affected many epigean species, their impact was especially pronounced in hypogean taxa, where caves acted as stable refugia amid climatic instability (Hedin 1997; Culver & Pipan 2009; Ribera *et al.* 2010; Schönhofer & Martens 2010). Similarly to the case of the southern alpine taxa, the speciation process in the northern Cantabrian Range is generally considered to be due to glacial cycles during the Quaternary period, which caused repeated isolations in non-glaciated 'karst refuge areas' (Schönhofer & Martens 2010; Luque & Labrada 2012). The preference of species of *Ischyropsalis* for permanently cool and humid environments, would have favoured the colonisation of caves (Mammola *et al.* 2019) as their fairly stable climate conditions would have meant that these were the only refuges during the cold periods of the Quaternary. The karst caves inhabited by *I. gigantea* are a clear example of the species' temperature

preference, average temperatures were recorded within the range of 6–9°C in the Recuistro Cave up in the Miera mountains at 1300 m a.s.l. (C.G. Luque unpubl. obs.). *Ischyropsalis cantabrica*, *I. gigantea* and our three new species are the westernmost cave-dwelling representatives of Ischyropsalidinae, likely

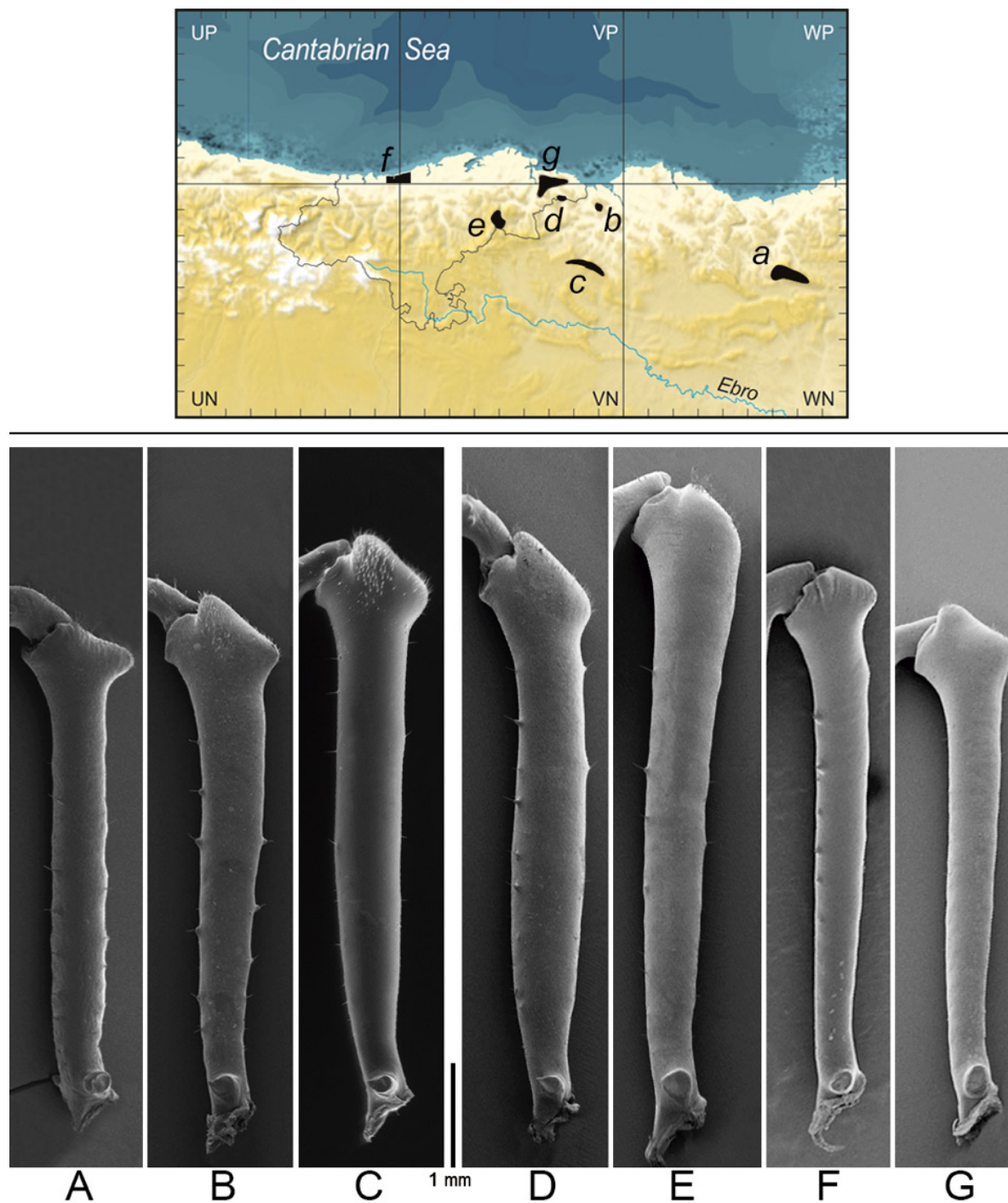


Fig. 15. Distribution of the seven Cantabrian troglobiont species of *Ischyropsalis* in the Cantabrian Mountains, northern Spain, and the basal cheliceral article of male (medial view). **A.** *I. navarrensis* Roewer, 1950. **B.** *I. magdalenae* Simon, 1881. **C.** *I. dispar* Simon, 1872. **D.** *I. aguerana* Luque & Labrada sp. nov. **E.** *I. gigantea* Dresco, 1968. **F.** *I. cantabrica* Luque & Labrada, 2012. **G.** *I. impressa* Luque & Labrada sp. nov.; for details, see Luque & Labrada (2012).

isolated during the last glaciation period (Fig. 1), as the closest karst areas to the localities of these three new species lie on the Sierra Salvada Massif (Burgos–Álava), inhabited by *I. dispar* and the Triano-Galdames Mountains (Biscay), inhabited by *I. magdalenae* (Fig. 15). In this context, the extensive glaciation in the Cantabrian Mountains Range was controlled not only by the preexisting landforms and the altitude, but also by the available humidity, related to their distance from the sea (Santos *et al.* 2013; Serrano *et al.* 2013, 2017). This led to species isolation and small-scale endemism, and might have been an important factor promoting divergence in karst areas in northern Spain (Sendra 2023). A comparable example is provided by the cave crickets of the genus *Dolichopoda* Bolívar, 1880, in the Italian Apennines, where geographic isolation among karst systems promoted strong genetic structuring, lineage divergence and local endemism (Garzia *et al.* 2025).

The species delineation presented here provides additional support for this hypothesis and suggests the need for further research on populations across the broad and fragmented distribution range of the genus in northern Spain (Luque & Labrada 2012).

In Cantabria and Asturias, the most common conservation strategy for the subterranean ecosystems is focused on the establishment of protected areas (Adrados-González 2011; Ballesteros *et al.* 2013; Luque & Labrada 2015, 2022). Regarding troglobiont species of *Ischyropsalis*, only two such species are found in protected areas designated under EU Natura 2000 network (declared December 2004): *I. cantabrica* that inhabits the Rogeria Cave SAC, and *I. gigantea* present in the Eastern Mountain-Montaña Oriental SAC, a group of mountains in the Upper Asón and Miera Rivers Basins. Here are located the largest and most important caverns in Spain and they harbour a wide diversity of poorly understood specialized organisms that are of interest from both a conservational and evolutionary perspective (Labrada *et al.* 2010; Angus *et al.* 2012; Luque & Labrada 2012, 2017; Salgado *et al.* 2012; Faille *et al.* 2021; Sendra 2023; Fresneda *et al.* 2024).

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Data availability

The data used to support the findings of this study are included within the manuscript. The DNA sequences generated for this research have been uploaded to GenBank (<https://www.ncbi.nlm.nih.gov/genbank>) and BOLD (<https://www.boldsystems.org/>).

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Competing interests

The authors have declared that no competing interests exist.

Author contributions

Ricardo López-Alonso (conceptualization, methodology, formal analysis, investigation, writing—original draft preparation, writing—review and editing), Esteban Pascual-Parra (conceptualization, methodology, formal analysis, investigation, writing—original draft preparation, writing—review and editing), Lucia Labrada (conceptualization, methodology, investigation, writing—original draft preparation, writing—review and editing), Carlos G. Luque (conceptualization, methodology, investigation, writing—original draft preparation, writing—review and editing), Andrés Arias (conceptualization, writing—review and editing, funding acquisition), Eduardo Cires (conceptualization, writing—review and editing, supervision, funding acquisition), and Claudia González-Toral (conceptualization, methodology, formal analysis, investigation, writing—original draft preparation, writing—review and editing).

References

- Adrados-González L. 2011. *Costa oriental de Asturias. Un Paisaje singular*. Adrados. Oviedo.
- Akaike H. 1974. A new look at the statistical model identification. *IEEE Transactions on Automatic Control* 19 (6): 716–723. Available from <https://ieeexplore.ieee.org/document/1100705> [accessed 26 Aug. 2026].
- Altekar G., Dwarkadas S., Huelsenbeck J.P. & Ronquist F. 2004. Parallel metropolis-coupled Markov chain Monte Carlo for Bayesian phylogenetic inference. *Bioinformatics* 20: 407–415. <https://doi.org/10.1093/bioinformatics/btg427>
- Angus R.B., Edwards D.B., Luque C.G. & Labrada L. 2012. A chromosomal investigation of some European Leiodidae (Coleoptera), with particular focus on Spanish subterranean Leptodirini. *Comparative Cytogenetics* 6 (2): 127–139. <https://doi.org/10.3897/CompCytogen.v6i2.2575>
- Ballesteros D., Jiménez-Sánchez M. & García-Sansegundo J. 2013. Patrimonio geológico subterráneo en espacios naturales protegidos: caracterización geomorfológica preliminar de sistemas kársticos profundos en el Parque Nacional de los Picos de Europa (España). In: Vega J., Salazar A., Díaz-Martínez E. & Marchán C. (eds) *Patrimonio geológico, un Recurso para el Desarrollo*: 361–370. Instituto Geológico y Minero de España. Madrid.

- Baró-Pazos J. 2010. *Los Hitos de un histórico Conflicto territorial entre Cantabria y el País Vasco: el Caso Agüera (Guriezo) y Trucíos-Turtzioz. Desde sus Orígenes (Siglo XVI) hasta la Sentencia del Tribunal Supremo de 9 de abril de 2008*. Regional Government of Cantabria (Consejería de Presidencia y Justicia), Santander.
- Chernomor O., Von Haeseler A. & Quang Minh B. 2016. Terrace aware data structure for phylogenomic inference from supermatrices downloaded from supermatrices. *Systematic Biology* 65 (6): 997–1008. <https://doi.org/10.1093/sysbio/syw037>
- Culver D.C. & Pipan T. 2009. *The Biology of Caves and Other Subterranean Habitats*. Oxford University Press. <https://doi.org/10.1093/oso/9780198820765.001.0001>
- Culver D.C., Deharveng L., Bedos A., Lewis J.J., Madden M., Reddell J.R., Sket B., Trontelj P. & White D. 2006. The mid-latitude biodiversity ridge in terrestrial cave fauna. *Ecography* 29: 120–128. <https://doi.org/10.1111/j.2005.0906-7590.04435.x>
- Darriba D., Taboada G.L., Doallo R. & Posada D. 2012. JModelTest 2: more models, new heuristics and parallel computing. *Nature Methods* 9 (8): 772. <https://doi.org/10.1038/nmeth.2109>
- Dresco E. 1968. Recherches sur les opilions du genre *Ischyropsalis* (Fam. Ischyropsalidae). VII. *Ischyropsalis gigantea* sp. nov. *Bulletin du Museum national d'Histoire naturelle, 2^e série* 40 (2): 308–314.
- Dresco E. 1970. Recherches sur la variabilité et la phylogénie chez les opilions du genre *Ischyropsalis* C.L. Koch (Fam. Ischyropsalidae), avec création de la famille nouvelle des Sabaconidae. *Bulletin du Muséum national d'Histoire naturelle* 41: 1200–1213.
- Edgar R.C. 2004. MUSCLE: multiple sequence alignment with high accuracy and high throughput. *Nucleic Acids Research* 32 (5): 1792–1797. <https://doi.org/10.1093/nar/gkh340>
- Faille A., Balart-García P., Fresneda J., Bourdeau C. & Ribera I. 2021. A remarkable new genus of Iberian troglobitic Trechodina (Coleoptera: Carabidae: Trechinae: Trechini), with a revisited molecular phylogeny of the subtribe. *Annales de la Société entomologique de France (N.S.)* 57: 85–106. <https://doi.org/10.1080/00379271.2021.1880339>
- Federación Cántabra de Espeología (FCE). Available from <https://www.espeleocantabria.net> [accessed 26 Aug. 2026].
- Federación Española de Espeología (FEE). Available from <https://feespeleo.com> [accessed 26 Aug. 2026].
- Federación d'Espeoloxía del Principáu d'Asturies (FESPA). Available from <https://fespasturies.com> [accessed 26 Aug. 2026].
- 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.
- Fresneda F., Giachino P.M., Salgado J.M., Faille A., Bourdeau C., Cieslak A. & Ribera I. 2024. A phylogenetic classification of Leptodirini (Coleoptera, Leioididae, Cholevinae). *Memorie della Società Entomologica Italiana* 101: 3–936. <https://doi.org/10.4081/memoriesei.2024.3>
- García de la Noceda C., De Toledo F.O. & Del Pozo Gómez M. 1999. *Programa de Actualización del Inventario Hidrogeológico (PAIH). Análisis del Conocimiento actual Evaluación y Programación de Estudios en las Cuencas intercomunitarias*. Instituto Geológico y Minero de España, Madrid.

- Garzia M., Berrilli E., Lunghi E., Coppari L., Delcour N. & Salvi D. 2025. Genetic diversity and distribution of Italian cave crickets (*Dolichopoda*): toward a better understanding of lineage structure. *Animals* 15 (16): 2429. <https://doi.org/10.3390/ani15162429>
- Hastings W.K. 1970. Monte Carlo sampling methods using Markov chains and their applications. *Biometrika* 57 (1): 97–103. <https://doi.org/10.2307/2334940>
- Hedin M. 1997. Molecular phylogenetics at the population/species interface in cave spiders of the southern Appalachians (Araneae: Nesticidae: *Nesticus*). *Molecular Biology and Evolution* 14 (3): 309–324. <https://doi.org/10.1093/oxfordjournals.molbev.a025766>
- Hedin M., Derkarabetian S., McCormack M., Richart C. & Shultz J.W. 2010. The phylogenetic utility of the nuclear protein-coding gene EF1- α for resolving recent divergences in Opiliones, emphasizing intron evolution. *The Journal of Arachnology* 38: 9–20. <https://doi.org/10.1636/ha09-49.1>
- Helversen O. Von. & Martens J. 1972. Unrichtige Fundort-Angaben in der Arachniden-Sammlung Roewer. *Senckenbergiana Biologica* 53: 109–123.
- Hoang D.T., Chernomor O., Von Haeseler A., Quang Minh B. & Sy Vinh L. 2017. UFBoot2: omproving the ultrafast bootstrap approximation. *Molecular Biology and Evolution* 30 (5): 1188–1195. <https://doi.org/10.1093/molbev/msx281>
- Kearse M., Moir R., Wilson A., Stones-Havas S., Cheung M., Sturrock S., Buxton S., Cooper A., Markowitz S., Duran C., Thierer T., Ashton B., Meintjes P. & Drummond A. 2012. Geneious Basic: an integrated and extendable desktop software platform for the organization and analysis of sequence data. *Bioinformatics* 28 (12): 1647–1649. <https://doi.org/10.1093/bioinformatics/bts199>
- Kury A.B., Mendes A.C., Cardoso L., Kury M.S., Granado A. de A., Giribet G., Cruz-López J.A., Longhorn S.J., Medrano M., Oliveira A.B.R. de, Kury I.S. & Souza-Kury M.A. 2024. *World Catalogue of Opiliones*. WCO-Lite version 2.7. Available from <https://wcolite.com/> [accessed 26 Aug. 2026].
- Labrada L., Salgado J.M. & Luque C.G. 2010. Fauna invertebrada de las cavidades de Cantabria. *Locustella, Anuario de la Naturaleza de Cantabria* 7: 29–43. Available from http://www.impress.es/docs/publicaciones/Locustella_2010.pdf [accessed 26 Aug. 2026].
- Labrada L., Tosal L., Gutiérrez M., Ranero S.L., Tosal E. & Luque C.G. in press. Life in the dark: Preliminary list of the strictly adapted cave fauna in Cantabria region (Northern Spain). *Bulletin of the Cantabrian Cave Research & Caving Federation* 18, ver. 2026.1.1 (last updated on 10 Oct. 2026). Santander.
- Leray M., Yang J.Y., Meyer C.P., Mills S.C., Agudelo N., Ranwez V., Boehm J.T. & Machida R.J. 2013. A new versatile primer set targeting a short fragment of the mitochondrial COI region for metabarcoding metazoan diversity: application for characterizing coral reef fish gut contents. *Zoology* 10 (34): 1–14. <https://doi.org/10.1186/1742-9994-10-34>
- Luque C.G. 1991. Los Ischyropsalidoidea de la Cornisa Cantábrica: Cantabria (Arachnida: Opiliones: Palpatores: Ischyropsalididae y Sabaconidae). *Arquenas* 1: 1–96.
- Luque C.G. 1992. Contribución al conocimiento de los Opiliones epigeos e hipogeos de Cantabria (España). Primeras aportaciones al catálogo (Arachnida: Opiliones: Palpatores). *Proceedings of the 5th Spanish Meeting of Speleology, Camargo-Santander, Spain 1–4 November 1990*: 143–157 + errata sheet. Cantabrian Caving Federation, Santander.
- Luque C.G. & Labrada L. 2012. A new cave-dwelling endemic *Ischyropsalis* C.L. Koch, 1839 (Opiliones: Dyspnoi: Ischyropsalididae) from the karstic region of Cantabria (Spain). *Zootaxa* 3506: 26–42. <https://doi.org/10.11646/zootaxa.3506.1.2>

- Luque C.G. & Labrada L. 2015. Organizing biogeomorphic information aimed at the planning and management of protected areas in Cantabria (Spain). The case of El Pendo and Peñajorao Caves – an example of Special Interest Area and UNESCO World Heritage Site. *Boletín de la real Sociedad española de Historia natural (Sección Geológica)* 109: 5–25.
Available from <http://www.rsehn.es/publicaciones-geologica/art278> [accessed 26 Aug. 2026].
- Luque C.G. & Labrada L. 2017. Nuevas localizaciones de la especie *Cantabrodesmus lorioli* Mauriès, 1971 en Cantabria, España (Polydesmida, Diplopoda, Chelodesmidae). *Boletín de la real Sociedad española de Historia natural (Sección Biológica)* 111: 51–60.
Available from <http://www.rsehn.es/publicaciones-biologica/art370> [accessed 26 Aug. 2026].
- Luque C.G. & Labrada L. 2022. Paisaje, territorio y patrimonio natural. In: Fernández Vega P.A. (coord.) *Historia y Patrimonio de Cantabria*: 1–40. Regional Government of Cantabria (Consejería de Educación). Santander.
- Mammola S., Schönhofer A.L. & Isaia M. 2019. Tracking the ice: subterranean harvestmen distribution matches ancient glacier margins. *Journal of Zoological Systematics and Evolutionary Research* 57: 548–554. <https://doi.org/10.1111/jzs.12264>
- Martens J. 1969a. Die Sekretarbeit während des Paarungsverhaltens von *Ischyropsalis* C.L. Koch (Opiliones). *Zeitschrift für Tierpsychologie* 26: 513–523.
- Martens J. 1969b. Die Abgrenzung von Biospezies auf biologisch-ethologischer und morphologischer Grundlage am Beispiel der Gattung *Ischyropsalis* C.L. Koch 1839 (Opiliones, Ischyropsalididae). *Zoologische Jahrbücher Abteilung für Systematik* 96: 133–264.
Available from <https://www.researchgate.net/publication/292124925> [accessed 26 Aug. 2026].
- Martens J. 1978. Spinnentiere, Arachnida – Weberknechte, Opiliones. *Die Tierwelt Deutschlands* 64: 1–464.
- Metropolis N., Rosenbluth A.W., Rosenbluth M.N., Teller A.H. & Teller E. 1953. Equation of state calculations by fast computing machines. *Journal of Chemical Physics* 21: 1087–1092.
<https://doi.org/10.1063/1.1699114>
- Minh B.Q., Nguyen M.A.T. & Von Haeseler A. 2013. Ultrafast approximation for phylogenetic bootstrap. *Molecular Biology and Evolution* 30 (5): 1188–1195. <https://doi.org/10.1093/molbev/mst024>
- Moore G.W., Goodman M. & Barnabas J. 1973. An iterative approach from the standpoint of the additive hypothesis to the dendrogram problem posed by molecular data sets. *Journal of Theoretical Biology* 38 (3): 423–457. [https://doi.org/10.1016/0022-5193\(73\)90251-8](https://doi.org/10.1016/0022-5193(73)90251-8)
- Nguyen L.T., Schmidt H.A., Von Haeseler A. & Minh B.Q. 2015. IQ-TREE: A fast and effective stochastic algorithm for estimating maximum-likelihood phylogenies. *Molecular Biology and Evolution* 32 (1): 268–274. <https://doi.org/10.1093/molbev/msu300>
- Pinto-da-Rocha R., Machado G. & Giribet G. (eds). 2007. *Harvestmen: The Biology of Opiliones*. Harvard University Press, Cambridge, UK.
- Prieto C.E. 1990. The genus *Ischyropsalis* C.L. Koch (Opiliones, Ischyropsalididae) on the Iberian Peninsula. I. Non-troglobitic species. *Acta Zoologica Fennica* 190: 315–320.
- Prieto C., Malumbres-Olarte J., Luque C.G., Labrada L. & Schönhofer A. 2013. The *Ischyropsalis* radiation from the Basque-Cantabrian region: Multiple colonization of caves or recolonization of epigean habitats? *III Encuentro Ibérico de Biología Subterránea, Barcelona* (Poster).
Available from <https://www.researchgate.net/publication/321252883> [accessed 26 Aug. 2026].
- Rambaut A., Drummond A.J., Xie D., Baelz G. & Suchard M.A. 2018. Posterior summarization in Bayesian phylogenetics using Tracer 1.7. *Systematic Biology* 67 (5): 901–904.
<https://doi.org/10.1093/sysbio/syy032>

- Ribera I., Fresneda J., Bucur R., Izquierdo A., Vogler A.P., Salgado J.M. & Cieslak A. 2010. Ancient origin of a Western Mediterranean radiation of subterranean beetles. *BMC Evolutionary Biology* 10 (29): 1–14. <https://doi.org/10.1186/1471-2148-10-29>
- Robinson D.F. 1971. Comparison of labeled trees with valency three. *Journal of Combinatorial Theory, Series B* 11 (2): 105–119. [https://doi.org/10.1016/0095-8956\(71\)90020-7](https://doi.org/10.1016/0095-8956(71)90020-7)
- Roewer C.F. 1950. Über Ischyropsalididae and Trogulidae: Weitere Weberknechte XV. *Senckenbergiana* 28: 7–57.
- Ronquist F., Teslenko M., Van Der Mark P., Ayres D.L., Darling A., Höhna S., Larget B., Liu L., Suchard M.A. & Huelsenbeck J.P. 2012. MrBayes 3.2: Efficient bayesian phylogenetic inference and model choice across a large model space. *Systematic Biology* 61 (3): 539–542. <https://doi.org/10.1093/sysbio/sys029>
- Rozwałka R., Mazur A. & Staręga W.K. 2012. Distribution of harvestmen of the genus *Ischyropsalis* C.L. Koch (Arachnida: Opiliones) in Poland. *Fragmenta Faunistica* 55 (1): 11–18. <https://doi.org/10.3161/00159301FF2012.55.1.011>
- Ruiz-Cobo J., Muñoz-Fernández E. & Luque C.G. 2008. Revisión del complejo arqueológico del karst de la Baja (Laredo, Cantabria). *Sautuola, Journal of Institute of Prehistory and Archaeology of Santander* 14: 41–58. Available from <https://institutosautuola.es/descargas-sautuola/> [accessed 26 Aug. 2026].
- Salgado J.M., Luque C.G., Labrada L., Fresneda J. & Ribera I. 2012. Revisión del género *Cantabrogeus* Salgado, 2000, con la descripción de tres nuevas especies hipogeas endémicas de la Cordillera Cantábrica (Coleoptera, Leiodidae, Cholevinae, Leptodirini). *Animal Biodiversity and Conservation* 35 (1): 27–50. <https://doi.org/10.32800/abc.2012.35.0027>
- Santos J., Redondo J.M., González-Gutiérrez R.B. & Gómez-Villar A. 2013. Applying the AABR method to reconstruct equilibrium-line altitudes from the last glacial maximum in the Cantabrian Mountains (SW Europe). *Palaeogeography, Palaeoclimatology & Palaeoecology* 387: 185–199. <https://doi.org/10.1016/j.palaeo.2013.07.025>
- Schmidt M., Achterhold K., Pfeiffer F. & Melzer R.R. 2024. Kinematics of elongate harvestmen chelicerae: comparative range of motion analysis in extant *Ischyropsalis* (Dyspnoi, Opiliones). *Zoology* 167: 1–11. <https://doi.org/10.1016/j.zool.2024.126219>
- Schönhofer A.L. 2013. A taxonomic catalogue of the Dyspnoi Hansen & Sørensen, 1904 (Arachnida: Opiliones). *Zootaxa* 3679: 1–68. <https://doi.org/10.11646/zootaxa.3679.1.1>
- Schönhofer A.L. & Martens J. 2010. On the identity of *Ischyropsalis dentipalpis* Canestrini, 1872 and description of *Ischyropsalis lithoclasica* sp. n. (Opiliones: Ischyropsalididae). *Zootaxa* 2613 (1): 1–14. <https://doi.org/10.11646/zootaxa.2613.1.1>
- Schönhofer A.L., Vernesi C., Martens J. & Hedin M. 2015. Molecular phylogeny, biogeographic history, and evolution of cave-dwelling taxa in the European harvestman genus *Ischyropsalis* (Opiliones: Dyspnoi). *The Journal of Arachnology* 43: 40–53. <https://doi.org/10.1636/H14-39.1>
- Sendra A. (ed.) 2023. *Habitantes de la Oscuridad. Fauna Ibero-balear de las Cuevas*. Sociedad Entomológica Aragonesa, Zaragoza.
- Serrano E., González-Trueba J.J., Pellitero R. & Gómez-Lende M. 2013. Quaternary glacial evolution in the Central Cantabrian Mountains (Northern Spain). *Geomorphology* 196: 65–82. <https://doi.org/10.1016/j.geomorph.2012.05.001>
- Serrano E., González-Trueba J.J., Pellitero R. & Gómez-Lende M. 2017. Quaternary glacial history of the Cantabrian Mountains of northern Spain: a new synthesis. *Geological Society London* 433: 55–85. <https://doi.org/10.1144/SP433.8>

Tavaré S. 1986. Some probabilistic and statistical problems in the analysis of DNA sequences. *In*: Miura E.R.M. (ed.) Lectures on Mathematics in the Life Sciences. *American Mathematical Society* 17 (2): 57–86..

Thorell T. 1876. *Sopra alcuni Opilioni (Phalangidea) d'Europa e dell'Asia occidentale, con un quadro dei generi europei di quest'Ordine*. *Annali del Museo Civico di Storia Naturale di Genova* 1 (8): 452–508. <https://doi.org/10.5962/bhl.part.10418>

Trifinopoulos J., Nguyen L.T., von Haeseler A. & Minh B.Q. 2016. W-IQ-TREE: a fast online phylogenetic tool for maximum likelihood analysis. *Nucleic Acids Research* 44 (1): 232–235. <https://doi.org/10.1093/NAR/GKW256>

Vink C.J., Thomas S.M., Paquin P., Hayashi C.Y. & Hedin M. 2005. The effects of preservatives and temperatures on arachnids DNA. *Invertebrate Systematics* 19: 99–104. <https://doi.org/10.1071/IS04039>

Waterhouse A.M., Procter J.B., Martin D.M.A., Clamp M. & Barton G.J. 2009. Jalview Version 2-A multiple sequence alignment editor and analysis workbench. *Bioinformatics* 25 (9): 1189–1191. <https://doi.org/10.1093/bioinformatics/btp03>

Yang Z. 1994. Maximum likelihood phylogenetic estimation from DNA sequences with variable rates over sites: approximate methods. *Journal of Molecular Evolution* 39: 306–314. <https://doi.org/10.1007/BF00160154>

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Supplementary files

Supp. file 1. List of *EF1- α* and *COI* sequences from GenBank used for the molecular analyses in this study. Species name, countries, GenBank accession numbers and references are given. <https://doi.org/10.5852/ejt.2026.1104.3336.14633>

Supp. file 2. Additional figures. <https://doi.org/10.5852/ejt.2026.1104.3336.14634>