Data collection

All material examined was obtained through field campaigns conducted by the authors and other colleagues during the last 20 years and is deposited at the Centre de Recursos de Biodiversitat Animal of the Universitat de Barcelona (CRBA) and the Departamento de Zoología de la Universidad de La Laguna (DZUL), Tenerife, Canary Islands collection. The species were mostly captured by active sampling, searching under rocks, logs and tree barks. The captured specimens were either preserved in EtOH at 75% in collections at room temperature at both institutions or at 95% in -20ºC freezers at CRBA. All specimens were collected following institutional and governmental regulations and permits were granted by the different Cabildos of each island or by the governing body of each natural reserve.

Specimen imaging

Variation in the shape of different cheliceral views was quantified using landmark-based geometric morphometrics (Bookstein 1991; James Rohlf & Marcus 1993; Adams *et al.* 2004; Zelditch *et al.* 2004; Mitteroecker & Gunz 2009). For this purpose, we took high-resolution photographs of different views for all the specimens with a digital camera LEICA DFC 450 attached to a LEICA MZ16A stereoscopic microscope using the software Leica Application Software (LAS) v.4.4 (Leica Microsystems Ltd, Switzerland). The chelicera of spiders is formed by two segments. In *Dysdera*, the basal segment carries three teeth (distal, medial and basal) on a groove in its internal margin, and the basal tooth develops into a keel that extends proximally; and a hinged fang, which folds into the cheliceral groove and shows the opening of the venom conduct at its distal end (Fig. 2). Following a revision of the structure of variation in different character views in the *Dysdera* species from the Canary Islands (Bellvert et al. in prep.), we chose three different views to capture the variations in shape of the chelicera, namely the dorsal and lateral view of the basal segment, and the ventral view of the fang. Whenever possible, we digitized the left chelicera for five females and five males of each species. In case of damaged or missing left structures, we assumed symmetry in the vertical plane and the right side was imaged and inverted. Eight Canarian *Dysdera* species are exclusively known from caves and show somatic adaptations to the underground life (e.g. appendage elongation, eye reduction, loss of pigmentation) (Ribera *et al.* 1985; Ribera & Arnedo 1994; Arnedo & Ribera 1996, 1999; Arnedo *et al.* 2007). As selective pressures other than prey segregation may be acting differentially on these species compared to their epigean counterparts, we excluded them from the downstream analyses. A total of 400 specimens comprising 40 of the 57 (including nominal and undescribed) total species of the group were photographed.

Photographs for each view of the chelicera were compiled using the software TpsUtil (Rohlf 2015) and landmarks and semilandmarks were digitized using TpsDig2 (Rohlf 2017). Specifically, we considered 14 landmarks on the dorsal view of the basal chelicera segment; four fixed landmarks and 20 sliding semilandmarks on the lateral view; and three fixed landmarks and 10 semilandmarks on the lateral view of the fang (Fig 2).

Phylogenetic analyses

We inferred relationships of the focal taxa based on mitogenomic information. Mitogenomes were recovered from different sources and forms. We downloaded genomic and transcriptomic information from public repositories (NCBI). In addition, we gathered new Low-Coverage Whole Genome Sequencing data for selected species (see supplementary Table S2). We recovered mitochondrial genes using the pipeline detailed in Adrián‐Serrano *et al.* (2020). The concatenated data matrix was further completed by adding species represented by at least one of the following genes obtained by Sanger sequencing: cytochrome c oxidase 1 (COI), the NADH dehydrogenase subunit (nad1) and the large (16S) and small (12S) ribosomal subunits, available either from public databases or generated in house.

We included all *Dysdera* species from the Canary Islands, represented at least by two individuals and in the case of multi-island species, individuals from each island, with few exceptions (see supplementary Table S2). Additional *Dysdera* species from the mainland and Madeira were also included to polarize the tree. Finally, representatives of all the families within the Synespermiata clade, including members of the Superfamily Dysderoidea were considered to provide fossil information for calibration. All trees were rooted, assuming a sister group relationship of the families Hypochilidae and Filistatidae with the Synespermiata clade (Garrison *et al.* 2016; Wheeler *et al.* 2017; Fernández *et al.* 2018; Michalik *et al.* 2019; Kulkarni *et al.* 2020, 2021; Kallal *et al.* 2021; Ramírez *et al.* 2021).

Mitochondrial genes were manipulated and concatenated with Geneious Prime 2020.2.4 (www.geneious.com). Each gene was aligned independently using the software MAFFT (Katoh & Standley 2013) as implemented in Geneious, with the G-INS-I algorithm and default values (0.53 for gap penalty, 0.123 for offset value). We inferred a time-stamped phylogeny under a Bayesian uncorrelated relaxed molecular clock approach as implemented in BEAST v2.6.3 (Bouckaert *et al.* 2019). The concatenated data matrix was partitioned by gene, and the best evolutionary model for each gene partition selected with PartitionFinder 2 (Lanfear *et al.* 2017). Individual log-normal clocks were defined for each gene, and the tree prior was set to the Birth-Death model.

Calibration information used to constrain nodes for time estimation is summarized in supplementary table S3. In short, we combined 10 fossil calibrations with one biogeographic event. Fossil information was included as lognormal prior distribution on specific nodes, except for the root which was assigned a uniform prior. The biogeographic information (the Hercynian split of the Iberian plate into present-day major western Mediterranean islands) was defined as a normal prior distribution. We further enforced topological constraints on specific nodes (monophyly of Segestriidae, Oonopidae, Orsolobiidae and Dysderidae, Segestriidae sister to Oonopidae, Orsolobiidae and Dysderidae) following results of recent phylogenomic analyses (Kallal *et al.* 2021; Kulkarni *et al.* 2021). A starting tree including time and topological constraints was generated with the program PATHd8 (Britton *et al.* 2007).

We run three independent chains under selected priors for 100 million generations, sampling every 10,000 generations. Convergence among runs and correct mixing of the chains was monitored with TRACER v.1.7 (http://tree.bio.ed.ac.uk/software/tracer/). The burn-in was removed (10%) and the runs combined with the help of the BEAST accompanying programs LOGCOMBINER and TREEANNOTATOR.

Non-Canarian and cave-dwelling *Dysdera* taxa, as well as taxa for which phenotypic data was not available, were pruned from the final posterior distribution of time-stamped trees with the help of the R package ‘ape’ (Paradis & Schliep 2019).

Prey preference analysis

For prey preference analyses, we selected 14 phylogenetically spread-out *Dysdera* species, of different morphotypes, for which we managed to collect enough specimens for the experiments (Fig.4). Each species was represented by twenty specimens, except for *D. alegranzaensis* (N = 16); *D. tilosensis* (N = 7); *D. insulana* (N = 3) and *D. levipes* (N = 6).

The spiders were maintained at room temperature and starved during the three weeks before experiments. Individuals were placed singly into Petri dishes (30 mm diameter) with a moistened piece of filter paper. The offered prey size was half to equal body length of the spider. We offered five types of prey: spiders (*Pardosa prativaga*), flies (*Musca domestica*), carabids beetles (*Bembidion lampros*), and two species of woodlice (*Porcellio* *scaber* and *Armadillidium vulgare*). For the smallest species, *D. levipes* (range of size between 6.5-9 mm.), we tried six types of prey: juvenile spiders (*Pardosa prativaga*), flies (*Drosophila* sp.),staphilinids beetles (Aleocharinae), juveniles of woodlice (*Porcellio* *scaber* and *Armadillidium vulgare*) and collembola (*Sinella curviseta*). The preys were offered randomly, and if they were not captured within 30 min. we offered another one. We prevented the spider from consuming the prey to keep them hungry. The percentage of times that each spider preyed on each type of prey were recorded and species average values were calculated.

**References**

Adams, D.C., Rohlf, F.J. & Slice, D.E. (2004). Geometric morphometrics: Ten years of progress following the ‘revolution.’ *Ital. J. Zool.*, 71, 5–16.

Adrián‐Serrano, S., Lozano‐Fernandez, J., Pons, J., Rozas, J. & Arnedo, M.A. (2020). On the shoulder of giants: Mitogenome recovery from non‐targeted genome projects for phylogenetic inference and molecular evolution studies. *J. Zool. Syst. Evol. Res.*, 1–26.

Arnedo, M. & Ribera, C. (1996). Dysdera ratonensis Wunderlich, 1991 (Arachnida, Araneae) a troglomorphic species from La Palma, Canary Islands: Description of the male and redescription of the female. *Rev. Arachnol.*, 11, 109–122.

Arnedo, M.A., Oromí, P., Múrria, C., Macías-Hernández, N. & Ribera, C. (2007). The dark side of an island radiation: systematics and evolution of troglobitic spiders of the genus Dysdera Latreille (Araneae : Dysderidae) in the Canary Islands. *Invertebr. Syst.*, 21, 623.

Arnedo, M.A. & Ribera, C. (1999). Radiation in the genus Dysdera (Araneae, Dysderidae) in the Canary Islands: The island of Tenerife. *J. Arachnol.*, 27, 604–662.

Bookstein, F.L. (1991). *Morphometric tools for landmark data: Geometry and biology*. Cambridge University Press, New York.

Bouckaert, R., Vaughan, T.G., Barido-Sottani, J., Duchêne, S., Fourment, M., Gavryushkina, A., *et al.* (2019). BEAST 2.5: An advanced software platform for Bayesian evolutionary analysis. *PLoS Comput. Biol.*, 15, 1–28.

Britton, T., Anderson, C.L., Jacquet, D., Lundqvist, S. & Bremer, K. (2007). Estimating divergence times in large phylogenetic trees. *Syst. Biol.*, 56, 741–752.

Fernández, R., Kallal, R.J., Dimitrov, D., Ballesteros, J.A., Arnedo, M.A., Giribet, G., *et al.* (2018). Phylogenomics, Diversification Dynamics, and Comparative Transcriptomics across the Spider Tree of Life. *Curr. Biol.*, 28, 1489-1497.e5.

Garrison, N.L., Rodriguez, J., Agnarsson, I., Coddington, J.A., Griswold, C.E., Hamilton, C.A., *et al.* (2016). Spider phylogenomics: Untangling the Spider Tree of Life. *PeerJ*, 2016.

James Rohlf, F. & Marcus, L.F. (1993). A revolution morphometrics. *Trends Ecol. Evol.*, 8, 129–132.

Kallal, R.J., Kulkarni, S.S., Dimitrov, D., Benavides, L.R., Arnedo, M.A., Giribet, G., *et al.* (2021). Converging on the orb: denser taxon sampling elucidates spider phylogeny and new analytical methods support repeated evolution of the orb web. *Cladistics*, 37, 298–316.

Katoh, K. & Standley, D.M. (2013). MAFFT multiple sequence alignment software version 7: Improvements in performance and usability. *Mol. Biol. Evol.*, 30, 772–780.

Kulkarni, S., Kallal, R.J., Wood, H., Dimitrov, D., Giribet, G. & Hormiga, G. (2021). Interrogating Genomic-Scale Data to Resolve Recalcitrant Nodes in the Spider Tree of Life. *Mol. Biol. Evol.*, 38, 891–903.

Kulkarni, S., Wood, H., Lloyd, M. & Hormiga, G. (2020). Spider-specific probe set for ultraconserved elements offers new perspectives on the evolutionary history of spiders (Arachnida, Araneae). *Mol. Ecol. Resour.*, 20, 185–203.

Lanfear, R., Frandsen, P.B., Wright, A.M., Senfeld, T. & Calcott, B. (2017). Partitionfinder 2: New methods for selecting partitioned models of evolution for molecular and morphological phylogenetic analyses. *Mol. Biol. Evol.*, 34, 772–773.

Michalik, P., Hormiga, G., Kallal, R. & Giribet, G. (2019). Phylogenomics and genital morphology of cave raptor spiders ( Araneae , Trogloraptoridae ) reveal an independent origin of a flow ‐ through female genital system, 1–11.

Mitteroecker, P. & Gunz, P. (2009). Advances in Geometric morphometrics. *Evol. Biol.*, 36, 235–247.

Paradis, E. & Schliep, K. (2019). Ape 5.0: An environment for modern phylogenetics and evolutionary analyses in R. *Bioinformatics*, 35, 526–528.

Ramírez, M.J., Magalhaes, I.L.F., Derkarabetian, S., Ledford, J., Griswold, C.E., Wood, H.M., *et al.* (2021). Sequence capture phylogenomics of true spiders reveals convergent evolution of respiratory systems. *Syst. Biol.*, 70, 14–20.

Ribera, C. & Arnedo, M.A. (1994). Description of Dysdera gollumi (Araneae, Haplogynae), a new troglobitic species from Tenerife, Canaray Islands, with some comments on Canarian Dysdera. *Mémoires de Biospéologie*, 21, 115–119.

Ribera, C., Fernandez, M.A. & Blasco, A. (1985). Araneidos cavernicolas de Canarias II. *Mémoires de Biospéologie*, 51–68.

Rohlf, F.J. (2015). The tps series of software. *Hystrix*, 26, 1–4.

Rohlf, F.J. (2017). tpsDig2.

Wheeler, W.C., Coddington, J.A., Crowley, L.M., Dimitrov, D., Goloboff, P.A., Griswold, C.E., *et al.* (2017). The spider tree of life: phylogeny of Araneae based on target-gene analyses from an extensive taxon sampling. *Cladistics*, 33, 574–616.

Zelditch, M.L., Swiderski, D.L., Sheets, H.D. & Fink, W.L. (2004). *Geometric morphometrics for biologists: A primer*. Elsevier Academic Press, New York.