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The non-dereliction in evolution: Trophic specialisation drives convergence in the radiation of red devil spiders (Araneae: Dysderidae) in the Canary Islands

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Abstract

Natural selection plays a key role in deterministic evolution, as clearly illustrated by adaptive radiations. Unlike most spiders, *Dysdera* species display a high variability of cheliceral morphologies, which has been suggested to reflect different levels of specialisation to feed on isopods. In this study, we integrate geometric morphometrics and experimental trials with a fully resolved phylogeny of the highly diverse endemic species from the Canary Islands to (1) characterize cheliceral morphologies, (2) unravel their dietary function, (3) examine if they evolved multiple times independently (4) verify whether convergent evolution of morphotypes has occurred and (5) test if specialization could lead to evolutionary irreversibility. We show the existence of nine cheliceral morphotypes and uncovered their significance for trophic ecology. Further, we demonstrate that similar ecomorphs evolved multiple times in the archipelago, providing a novel study system to explain how convergent evolution and irreversibility due to specialization may be combined to shape phenotypic diversification in adaptive radiations.

Keywords: Convergent evolution, irreversibility, geometric morphometrics, phylogenetic comparative methods, mitogenomics, adaptive radiation, ecological specialization

Understanding the origins and identifying the processes that shape biodiversity are among the major goals of biology ever since Darwin published his book “On the origin of species” (1859). Similarly, the ability to predict organismic response to natural selection lays at the very core of evolutionary biology (Blankers et al. 2017). One well-known process that has linked evolution and ecology is that of adaptive radiation (Givnish and Sytsma 1997), defined as the evolution of eco-phenotypically distinct species from a single common ancestor that has occurred in a short period of time (Schluter 2000). Typically, adaptive radiations are characterized by a fast synchronous diversification of lineages and phenotypes after the colonization of a new environment, which slows down as ecological niches are filled (Glor 2010). The study of adaptive radiations has greatly contributed to shed light on the mechanisms that drive speciation and promote eco-phenotypic diversification, along with many other related ecological and evolutionary processes (Gillespie et al. 2020).

A common pattern observed in adaptive radiations is the evolution of strikingly similar phenotypic outcomes (Losos et al. 1998) that arise as a result of the adaptation to similar ecological conditions (Rundle et al. 2000; Schluter 2000; Brakefield 2006; Losos 2011). The independent evolution of similarities as a result of common selective regimes, as opposed to those resemblances inherited from common ancestry, is known as convergent evolution (Arbuckle and Speed 2016). Patterns of convergent evolution have been extensively explored in famous cases of adaptive radiation across different taxa, e.g. cichlid fishes (Rüber et al. 1999; Stiassny and Meyer 1999; Hulsey et al. 2019), anole lizards (Kolbe et al. 2011; Mahler et al. 2013; Ord et al. 2013), crickets (Pascoal et al. 2014; Blankers et al. 2017) and spiders (Blackledge and Gillespie 2004). Furthermore, such convergent morphotypes are typically associated with similarities in ecological performance (Irschick and Higham 2015; De Meyer et

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al. 2019). For instance, the evolution of similar morphotypes in distantly related clades has been shown to be linked to the convergent evolution of specific dietary adaptations in several taxa such as marine tetrapods (Kelley and Motani 2015), rodents (Martinez et al. 2018), cichlid fishes (Muschick et al. 2012) and beetles (Baulechner et al. 2020).

The evolution of trophic adaptations has been frequently investigated as a dichotomy between stenophagus and euryphagous specializations (i.e. specialist vs. generalist), depending on the food resource availability and the fitness benefits gained from each strategy (Levins and MacArthur 1969). However, specialization could represent a wide spectrum of different definition of what types of adaptations species have (Ferry-Graham et al. 2002). Previous studies have defined stenophagy as species occupying a section of the euryphagous' niche breath (e.g. herbivore and carnivore as specialist and omnivorous as generalists), in such cases, generalist diets have been seen as macroevolutionary sinks (Burin et al. 2016; Bender and Luiz 2019; Borstein et al. 2019). However, some species presents narrow trophic requirements adapted to prey on a resource not (or less) exploited by wider trophic consumers. In such cases, the evolutionary consequences could be completely different, like an evolutionary irreversibility derived of the long term effect of this specialization (Day et al. 2016), compromising the ability of lineages for future evolutionary change (e.g. Cope 1896; Haldane 1951; Kelley and Farrell 1998; Nosil and Mooers 2005; Vamosi *et al.* 2014, Day *et al.* 2016). Stenophagous adaptations often translate into morphological traits that increase the efficiency of feeding on certain specific preys (Pekar et al. 2011) thus, the irreversibility to a more generalist diet by specialist species could translate into evolutionary irreversibility of their phenotypic characters.

But nevertheless, understanding all these evolutionary processes (adaptive radiation, convergent evolution and irreversibility) is sometimes hampered by the species niche

conservatism among closely related groups (but see Wiens & Graham 2005 for a review in this topic). Furthermore, patterns of species diversification on continental regions are often blurred by the large temporal and spatial scales involved (Gillespie 2016). Because of their clear boundaries, small size, well dated chronology and multiple replicates, oceanic islands, in contrast, provide natural experiments for studies of species proliferation and ecological adaptation (Losos et al. 1998; Losos and Schluter 2000; Beheregaray et al. 2004; Gillespie 2004; Emerson and Kolm 2005). The Atlantic archipelago of the Canary Islands is formed by 7 major islands and several islets of volcanic origin, located 100 kilometres from the north-western coast of Africa (Fig. 1a). The islands are geochronological arranged, with the oldest islands, Lanzarote and Fuerteventura (15 My and 23 My respectively), lying at the easternmost side, and becoming progressively younger towards the western side (from east to west): Gran Canaria (subaerial age 15 My), Tenerife (12 My), La Gomera (11 My), La Palma (1.7 My) and El Hierro (1.1 My) (Van Den Bogaard 2013). Local communities are highly structured by elevation as a result of the influence of the humid north-east trade winds, which explains the deep differences between the lush and habitat-rich western islands and the arid and highly eroded older islands of Fuerteventura and Lanzarote (Macías-Hernández et al. 2016). Like other isolated oceanic land masses with no former connection to the mainland, the Canary Islands exhibit high levels of endemism, which reaches 66% in the case of spiders (Banco de Datos de Biodiversidad de Canarias, 2021; Cardoso *et al.* 2010).

The red devil spider genus *Dysdera* (Dysderidae) (Fig. 2a), with approximately 300 described species (World Spider Catalog, 2022), have been repeatedly reported to feed on woodlice (Bristowe 1958; Sunderland and Sutton 1980; Hopkin and Martin 1985; Raupach 2005), a prey usually rejected by generalist predators (Pekár et al. 2016). Concomitantly,

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Dysdera spiders show an unusual variability in their mouthpart morphology, which has been suggested to be related to different levels of dietary specialization for feeding on isopods (i.e. oniscophagy) and prey capture strategies (Řezáč and Pekár 2007; Toft and Macías-Hernández 2017), as well as to metabolic adaptations (Toft and Macías-Hernández 2021).

With a western Palearctic distribution (Deeleman-Reinhold and Deeleman 1988), *Dysdera* species have colonized all Macaronesian archipelagos (Crespo et al. 2021), which represents the westernmost limit of its range (Arnedo and Ribera 1999; Arnedo et al. 2001). In the Canary Islands, and in a lesser degree in Madeira (Crespo et al. 2020), this genus has undergone major diversification processes. To date, 47 valid species have been described in the Canarian archipelago (Macías-Hernández et al. 2016), most of which are single-island endemics. The Canarian species assemblage offers an ideal experimental model to study patterns in species radiation as most of their species have diversified from a single common ancestor in the archipelago (Adrián-Serrano et al. 2020), and show a high variability of phenotypic modifications related to ecological performances (Toft and Macías-Hernández 2017, 2021; Řezáč et al. 2021). A former study based on traditional morphometric analysis, suggested the existence of five cheliceral morphotypes in the islands, and linked them to different prey-capture strategies (Řezáč et al. 2021). Additionally, oniscophagy was found to be ancestral for *Dysdera* in the Canary Islands, and it was suggested that a more generalist diet would have evolved secondarily from specialist ancestors.

In this study, we combined Geometric Morphometric (GM) tools, with experimental data and phylogenetic comparative methods, incorporating a new robust phylogeny of the group, to investigate the recurrent evolution of cheliceral ecotypes in *Dysdera* spiders from the Canary Islands. Specifically, we aim to 1) quantitatively delimit the different cheliceral morphotypes

present in the archipelago, 2) test their association to trophic specialisation, as reported for continental species 3) reconstruct the evolution of these ecomorphs throughout the diversification of the group, 4) test the hypothesis of convergent evolution of the different morphotypes and, 5) examine whether specialisation constitutes a case of evolutionary irreversibility in this group. By addressing these questions, we investigate the interaction between convergence and irreversibility in shaping phenotypic diversification, evolutionary processes that have not been previously explored in combination. To this end, we integrate experimental evidence on the role of oniscophagy in driving mouthpart evolution with multidimensional shape data in a phylogenetic framework to disentangle the evolution of functional diversity in this megadiverse spider genus.

Material & methods

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 -20°C 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

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Variation in the shape of different cheliceral views was quantified using landmark-based geometric morphometrics (Bookstein 1991; James Rohlf and Marcus 1993; Adams et al. 2004; Zelditch et al. 2004; Mitteroecker and 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 (Fig. 2c); and a hinged fang, which folds into the cheliceral groove and shows the opening of the venom conduct at its distal end (Fig. 2d). 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 all the species with a minimum of at least three specimens. 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 and Arnedo 1994; Arnedo and 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 (see Morphometric_sampling of the Supplementary material available on Dryad). 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 four fixed landmarks and 20 sliding semilandmarks on the lateral view; 14 landmarks on the dorsal view of the basal chelicera segment; and three fixed landmarks and 10 semilandmarks on the lateral view of the fang (Fig 2b-d).

Geometric Morphometric Analysis

To remove non-shape variation (i.e. related to location, scale and rotation) from individual landmark coordinates, each dataset was subjected to a Generalized Procrustes Analysis (GPA: Gower 1975; Rohlf & Slice 1990) as implemented in the `gpgen` function of the R package “geomorph” (Adams et al. 2021; Baken et al. 2021). During GPA superimposition the position of semilandmarks was optimized by minimizing bending energy. We used the function `combine.subsets` to combine all three views in a single landmark configuration that captured the total shape variation of the chelicera in our sample. With this combined configuration, we performed a second GPA to scale all views to unit centroid size and take their relative proportions into account (Collyer et al. 2020). With the resulting combined superimposed and scaled coordinates, we calculated the mean shape of each species. These mean shapes were subsequently used to perform a principal component analysis (PCA) using the function `gm.prcomp`. The first two principal components (PC) were used to explore and visualize shape variation across species.

Species clustering

To quantitatively delimit morphotypes present in the *Dysdera* species from the Canary Islands, we applied a hierarchical clustering analysis based on Procrustes distances among species means of cheliceral shape, using the function `hclust` of the *stats* R-package (R core team,

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200 2021). To select the best algorithm for clustering, we calculated the correlation between the
201 original distance matrix and the cophenetic distances on the phenogram derived by each method
202 (ward.D, ward.D2, single, complete, average, mcquitty, median and centroid) and chose the one
203 that maximized this correlation. To delimit clusters that could be interpreted as distinctive
204 morphotypes, we combined the resulting phenogram with pairwise p-values between species
205 obtained using permutation procedures as implemented in the function *pairwise* of RRPP R
206 package (Collyer and Adams 2018, 2021), over an ANOVA comparison of species means
207 performed using the function *procD.lm* of geomorph R package (Adams et al. 2021; Baken et al.
208 2021). Pairwise p-values were plotted as significant or non-significant considering significance
209 at $\alpha = 0.05$. To delimit morphotypes, we selected those clusters that included the highest number
210 of species with non-significant shape differences among them.

211 *Phylogenetic analyses*

212 We inferred relationships of the focal taxa based on mitogenomic information.
213 Mitogenomes were recovered from different sources and forms. We downloaded genomic and
214 transcriptomic information from public repositories (NCBI). In addition, we gathered new Low-
215 Coverage Whole Genome Sequencing data for selected species (see Molecular_Sampling of the
216 Supplementary material available on Dryad). We recovered mitochondrial genes using the
217 pipeline detailed in Adrián-Serrano *et al.* (2020). The concatenated data matrix was further
218 completed by adding species represented by at least one of the following genes obtained by
219 Sanger sequencing: cytochrome c oxidase 1 (COI), the NADH dehydrogenase subunit (nad1)
220 and the large (16S) and small (12S) ribosomal subunits, available either from public databases or
221 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 Molecular_Sampling of the Supplementary material available on Dryad). Additional *Dysdera* species from the mainland and Madeira were also included to polarize the tree. Finally, representatives of all the families within the Synesperiata 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 Synesperiata 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 and 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 Appendix 1, Table S1. 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

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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. Unless specified otherwise, we used the consensus tree for downstream analysis.

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 and Schliep 2019).

Phylogenetic signal

To evaluate to what extent the cheliceral morphological resemblance among *Dysdera* species is associated to their shared phylogenetic history, we measured phylogenetic signal using Blomberg’s *K* statistic (Blomberg et al. 2003), as generalized for high-dimensional and multivariate data (Kmult: Adams 2014). In addition, to test if Kmult values were significantly different from one, we developed a simulation procedure (see R scripts of the Supplementary material available on Dryad) that simulated multivariate phenotypes of the same dimensions as

our data, under the empirical evolutionary rate matrix observed in our dataset and using a Brownian Motion model of evolution. This produced a null distribution of expected values of $K_{mult} = 1$, against which we compared the value observed in our dataset. We calculated the corresponding p-value as the number of times the observed value was more extreme than that expected from the simulated phenotypes. In both analyses, we accounted for phylogenetic uncertainty by estimating K_{mult} and evaluating its significance over 1,000 trees subsampled from the posterior distribution of BEAST, using the function *physignal* of the “geomorph” R package.

Ancestral State Reconstruction

To reconstruct the evolutionary history of chelicera morphotypes, we used stochastic character mapping (Bollback 2006) with the function *make.simmap* of the R package *phytools* (Revell 2012). To take phylogenetic uncertainty into account, we used the same subsample of 1,000 trees as before from the posterior distribution of trees obtained from the BEAST analysis. We simulated 100 stochastic character maps under four different transition-rate scenarios, namely: a single-parameter model with equal rates (ER), a model with different rates for each transition (ARD), a model with equal forward and reverse rates (SYM), and a model with equal transition rates but with the constrain of irreversibility (constrained ER), where the generalist state can be lost but not regained. The last scenario allows to test the irreversibility of trophic specialization, as we allowed all transitions from generalist towards specialized ecomorph states (see Results) under an equal rate but set all rate parameters involving reversals from a generalist towards a specialist ecomorph equal to zero. We evaluated the fit of different models by comparing the AIC values of the alternative scenarios over the 1000 topologies by 100 *simmap* combinations.

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290 *Convergent evolution*

291 To test for evolutionary convergence of the different cheliceral groups, we quantified the
292 amount of independent evolution of each morphotype using the R package “convevol” (Stayton
293 2015a). For this, we used the first two PCs of cheliceral shape variation (accounting for 62.88%
294 of total variation, see results) and we excluded all cheliceral morphs with less than three species.
295 This was necessary because calculating and evaluating convergence statistics requires less
296 variables than number of species in each group. We investigated the degree of evolutionary
297 convergence using the distance-based statistics C1 and C3 (Stayton 2015). These two metrics
298 provide a complementary view on how exceptional the similarity between two taxa is, by
299 considering the ratio between the phenotypic distance between the two potentially convergent
300 taxa and the maximum distance observed between any pair of taxa (either ancestral or extant)
301 across the evolutionary history of these lineages. The significance of departure from the values
302 of C1 and C3 expected under Brownian Motion was assessed using 100 simulations performed
303 through the function *convratsig*, and again we accounted for phylogenetic uncertainty by using a
304 subset of 100 trees from the posterior distribution of BEAST analyses.

305 *Prey Preference analysis*

306 For prey preference analyses, we selected 14 phylogenetically spread-out *Dysdera* species,
307 of different morphotypes, for which we managed to collect enough specimens for the experiments
308 (Appendix 1, Table S2). Each species was represented by twenty specimens, except for *D.*
309 *alegranzaensis* (N = 16); *D. tilosensis* (N = 7); *D. insulana* (N = 3) and *D. levipes* (N = 6).

310 The spiders were maintained at room temperature and starved during the three weeks before
311 experiments. Individuals were placed singly into Petri dishes (30 mm diameter) with a moistened

312 piece of filter paper. The offered prey size was half to equal body length of the spider. We offered
313 five types of prey: spiders (*Pardosa prativaga*), flies (*Musca domestica*), carabids beetles
314 (*Bembidion lampros*), and two species of woodlice (*Porcellio scaber* and *Armadillidium vulgare*).
315 For the smallest species, *D. levipes* (range of size between 6.5-9 mm.), we tried six types of prey:
316 juvenile spiders (*Pardosa prativaga*), flies (*Drosophila* sp.), staphilinids beetles (Aleocharinae),
317 juveniles of woodlice (*Porcellio scaber* and *Armadillidium vulgare*) and collembola (*Sinella*
318 *curviseta*). The preys were offered randomly, and if they were not captured within 30 min. we
319 offered another one. We prevented the spider from consuming the prey to keep them hungry. The
320 percentage of times that each spider preyed on each type of prey were recorded and species average
321 values were calculated.

322 We first tested for a statistical association between cheliceral morphotype and the prey
323 preference, using a two-block partial least-squares (PLS) analysis implemented in the function
324 *phylo.integration* from the “geomorph” R package (Adams et al. 2021; Baken et al. 2021), with
325 the consensus phylogeny trimmed to match the species analysed. This test evaluates the degree
326 of covariation between two sets of variables, while considering the relationship between species.
327 Then, we used prey preference data to examine whether we could accurately identify species as
328 generalists or specialists. For this purpose, we calculated the proportion of isopod versus non-
329 isopod prey consumed by each individual. With the mean value per species, we then performed a
330 clustering analysis using the function *kmeans* with $k = 2$ to assign species into one of the
331 aforementioned trophic specialization categories. To evaluate whether these two groups differed
332 in their degree of specialization towards isopod consumption, we conducted an ANOVA test to
333 assess if species from the presumed generalist cluster exhibited a significantly lower preference
334 for accepting isopods as prey than those included in the specialist cluster.

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335 Results

336 *Characterization of cheliceral morphotypes*

337 The first two components of the PCA of the mean shapes of all species analysed
338 explained 49.85% and 13.02% of chelicera shape variation, respectively. PC1 captured shape
339 variation related to the elongation of the basal segment in dorsal view and the relative distance
340 from the distal cheliceral teeth to the distal part of the basal segment, as well as with the
341 bulginess of the basal segment in lateral view and relative fang length (Appendix 2, Fig. S1).
342 PC2 was mostly associated with variation in the relative length of the cheliceral groove, the
343 stoutness of the basal segment in lateral view, and the relative width of the fang (Appendix 2,
344 Fig. S2).

345 Of all the clustering algorithms used on the pairwise Procrustes distances among species
346 mean shapes, the unweighted pair group method with arithmetic mean (i.e. UPGMA) showed the
347 highest correlation between the node distance and the original distance matrix ($r=0.746$). The
348 combined examination of the resulting phenogram with the pairwise p-values between all pairs
349 of species, revealed several clusters that maximized non-significant within-group differences in
350 the shape of the chelicera (Fig. 1b). As the best arrangement of the cheliceral variability
351 displayed by *Dysdera* spiders in the Canary Islands, we identified nine morphotypes, hereafter
352 referred to by the letters A to I. Cheliceral morphotypes G, H, and I exhibit an elongation of their
353 views as described above with the extreme shapes in PC1. Morphotypes C and E have the
354 concavity of the chelicera slightly marked. Morphotype D is characterized by a protruding
355 cheliceral bulge, and morphotype F represents the stouter chelicera with a short and wide fang
356 (Fig. 3). The cheliceral morphotypes A, B, C, F, G and I are circumscribed to the western

islands, namely Gran Canaria, Tenerife, La Gomera, La Palma and El Hierro, while morphotypes D, E and H are exclusive to the eastern islands of Lanzarote and Fuerteventura (Fig. 1a). The time-stamped phylogeny inferred from mitogenome data (Appendix 2 Fig. S3), was pruned to include Canarian species with GM information for downstream analyses.

Relationships between morphotypes and dietary preferences

Prey preference behaviour varied extensively across the *Dysdera* species analysed, where the percentage of isopod acceptance ranged between 20% in some species and up to almost 92% in others (Fig. 4b). The phylogenetic PLS analysis showed a significant covariation between prey preference and cheliceral shape ($r\text{-PLS} = 0.863$, $P\text{-value} = 0.002$) (Fig 4a). The composition of the first PLS eigenvector for the prey block indicates that this association is mainly explained by the level of predatory preference for *Armadillidium* isopods (0.788), *Musca* flies (0.463) and *Porcellio* isopods (0.380) (Appendix 1, Table S3). In terms of shape variation, the preference for *Armadillidium* predation (characteristic of e.g. *D. insulana* and *D. tilosensis*: Appendix 1, Table S2) is associated to an elongation of all views analysed, and particularly of the fang and the anterior part of the chelicera (Fig. 4a). By contrast, higher levels of predation on *Porcellio* are associated to more compact chelicera with a relatively flat and short fang, as exemplified in *D. ramblae* (Fig. 4a, Appendix 1, Table S2). *Dysdera* species with intermediate shapes, occupying the central portion of the PLS space of association between cheliceral shape and dietary preference, exhibit higher levels of acceptance of *Musca* flies. The K-means cluster analysis based on the percentage of acceptance of isopods grouped together species belonging to morphotypes A and D, hereafter referred as generalists. By contrast, all other species were grouped together in the second cluster, the specialists, except for two species belonging to morphotypes B and G (Fig. 4b). To avoid over-splitting, we assigned these species to the cluster

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that included the remaining species of their morphotype. The ANOVA comparison indicated significant differences between the two clusters (p -value = 0.006), where generalist species consumed lower percentages of isopods than specialist ones (Fig. 4b).

Evolution of morphotypes

The best supported model of state transition to describe the evolutionary dynamics of different cheliceral morphotypes was the constrained ER encompassing irreversibility (Akaike Information Criterion, $AIC = 42.395 \pm 1.001$). The second and third most supported models were the ER and SYM models ($AIC = 154.468 \pm 18.780$ and 187.468 ± 6.035 respectively), and the least supported model was ARD ($AIC = 266.068 \pm 8.210$). All models, except for ARD, identified morphotype A as the most likely ancestral state of all *Dysdera* species from the Canary Islands (Fig. 5, Appendix 2, Fig. S4-6, Appendix 1, Table S4-7). The character reconstruction of all nine different cheliceral morphotypes, revealed that at least three (B, F and G) out of the six morphotypes represented by more than one species evolved more than once (Fig. 5). Cheliceral shape as represented by the combined coordinates of all three examined views exhibited significant phylogenetic signal ($K_{mult} = 0.657 \pm 0.015$, p -value = 0.0014 ± 0.0005), which suggested a phylogenetic component in the observed phenotypic variation. Furthermore, the observed K value was significantly smaller than one (p -value = 0.0144 ± 0.0023), as also supported by a lack of phylogenetic structure in phylomorphospace (Fig. 3).

For the three cheliceral morphotypes with more than three species analysed (A, B, G), distance-based measures of convergence revealed significantly higher observed C1 and C3 values than those expected by chance (p -value < 0.001). Specifically, the distance between any two species in morphotype A was closed between 69.28 to 75.54% by subsequent evolution

(C1), while convergence was responsible for 42.89 to 48.08% of the evolution between those lineages (C3). Morphotypes B and G also showed significant, and similar C values ($C1 = 50.53-63.67$ and $41.42-63.93\%$, $C3 = 24.86-37.67$ and $24.31-38.03\%$, respectively).

Discussion

With the present study, we have been able to untangle the effects of ecological pressures that have shaped the phenotypic diversity of a group with a shared evolutionary history. The repeated evolution of characters adapted to different levels of trophic specialization, has allowed to understand the deterministic evolution caused by the adaptation to the same ecological needs and the irreversibility effects triggered from those adaptations. Furthermore, our study has shown that, to capture complex characters involved in the evolution of the species, fine scale methods able to capture subtle changes are needed to understand the evolutionary pressures that has led the phenotypic disparity in the group.

Previous studies had uncovered the existence of particular cheliceral morphotypes among Canarian *Dysdera* spiders (Řezáč et al. 2021) and shown that these species differ in their metabolic ability to feed on isopods (Toft and Macías-Hernández 2017, 2021). Here, we statistically characterise for the first time cheliceral morphotypes using geometric morphometrics and experimentally demonstrate their association to different levels of diet specialisation (oniscophagy). The existence of different cheliceral morphotypes was first identified in continental *Dysdera* species, using data on the length of the basal segment and the fang relative to carapace length, along with qualitative observations (Řezáč et al. 2008). The same morphotypes were subsequently reported in Canarian *Dysdera*, in that case by combining several linear measurements of the chelicera (Řezáč et al. 2021). The finer GM data and more

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sophisticated statistical analyses used here allowed us to delimit nine different cheliceral morphotypes in the Canary Islands, some of which greatly overlapped, but not fully matched, the previously defined morphotypes. Morphotypes A, D and E identified here mostly corresponded with the “unmodified” morphotype of previous studies, morphotype B and H to “elongated”, morphotype F to “flattened” and morphotype G to “concave”. By contrast the morphotypes C and I were identified here for the first time. Interestingly, the different cheliceral morphotypes had been experimentally linked to specific strategies of prey capture (Řezáč et al. 2008, 2021). One of these strategies, “dagger” was reported only in one species, *D. levipes*, which our analysis assigned to its own distinct cheliceral morphotype, C. On the other hand, the cheliceral morphotype I, which had not been reported before in the Canaries, closely resembles the “very elongated” morphotype described in continental species (Řezáč et al. 2008).

Cheliceral morphotypes reflect different levels of diet specialisation

Our results demonstrate a close link between morphotype differentiation in *Dysdera* species from the Canary Islands, as reflected in cheliceral modifications, and diet specialisation, specifically in the increase in the capture and consumption of terrestrial isopods, as already suggested in former studies (Řezáč et al. 2008, 2021; Toft and Macías-Hernández 2017, 2021). This implies an ecological functionality for the different cheliceral morphologies present in the archipelago, which assigns them a strong evolutionary potential. Although dietary preference remain to be tested in natural conditions, our data are suggestive of different levels of trophic specialization in preying on woodlice among Canarian species. Indeed, species of *Dysdera* belonging to the identified morphotypes also group together in the morphology-diet PLS space, where different morphotypes occupy distinct areas (Fig. 4a). Across this space, the more extreme shapes are associated with specialization in feeding on different types of isopods, while the

species with unmodified cheliceral types exhibit higher levels of acceptance of non-isopod prey. Thus, selection related to dietary preferences seems to have been a strong evolutionary agent for the diversification of cheliceral morphology in the *Dysdera* species from the Canary islands, possibly in the same way as in continental species where similar morphotypes occur (Řezáč et al. 2008). Interestingly, both generalist and diet specialists are represented by multiple cheliceral morphotypes. Indeed, when analysing dietary preferences in isolation, species with different cheliceral morphotypes could be classified into two main groups, namely, generalists (A and D) and specialists (B, C, F and G). In the case of dietary specialists, the different morphotypes have been related to alternative strategies of woodlice capture and manipulation (Řezáč et al. 2008, 2021). The elongation of the chelicera, together with the presence of a marked concavity, has been linked to the ability of *Dysdera* species to insert the chelicera under the Isopod to expose the soft parts to the fang, meanwhile the more stout chelicera with the flattened fang are used to attack the prey between the dorsal sclerites (Řezáč et al. 2008). Importantly, our analyses also suggest the existence of two distinct morphotypes with a more generalist diet (A and D). While these two morphotypes are clearly distinct in cheliceral shape (Fig. 3), lab experiments are not sufficient to fully capture the dietary differences between them. To what extent those morphological differences exhibited by generalist species respond to different hunting strategies or dietary preferences remains to be tested, preferably with the use of approaches that may allow to detect prey consumed in the wild (eg. DNA, stable isotopes).

Island group and specialisation

It is worth noting that the eastern Canaries showed the higher proportion of generalist species. In this regard, *D. mahan*, a species that could not be included in the GM analyses, bears chelicerae that seem to correspond to morphotype D, the more widespread across these islands,

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thus likely also a generalist. The Fuerteventura endemic *D. sanborondon*, on the other hand, bears a unique cheliceral morph (E) that is closer to the woodlice feeding morphotype B in the morphospace (Fig. 3), but the species was reported as a non-oniscophagus in Rezac et al. (2021). The eastern Canaries also harbour lower number of endemic *Dysdera* species than other islands of similar size yet of younger age (e.g. Gran Canaria, Tenerife) (Cardoso et al. 2010), which has been suggested to be the result of higher levels of extinction as a result of the mass-wasting processes, and severe climatic degradation endured by these islands (Macías-Hernández et al. 2008). Habitat deterioration and aridification could have posed specific filters on the local ecosystems, which may have hindered the presence of specialist species in the eastern islands. In this regard, it has been reported that specialist predators at high trophic levels are less likely to be present in species-poor island communities (Piechnik et al. 2008). Additionally, the different environmental filtering or ecological pressures between the eastern and western islands could help to explain the non-overlapping of cheliceral morphotypes between the two islands groups (differentiated across the second PC in Fig. 3). The observation that the only specialised *Dysdera* in the eastern Canaries have an independent origin may also hint the different ecological constraints acting on the highly transformed eastern islands. *Dysdera lancerotensis* is the only species present in the eastern Canaries that potentially specialises in feeding on woodlice, as suggested by both its spatial closeness to morphotypes associated to higher proportions of woodlice in their diets (Fig. 3) and experimental data (Řezáč et al. 2021), and is also the only endemic that have their closest relatives on the continent, and thus most likely constitutes an independent colonization of these islands (Bidegaray-Batista et al. 2007; Macías-Hernández et al. 2010; Adrián-Serrano et al. 2020).

A generalist coloniser and convergent evolution of morphotypes

Contrary to former suggestions (Řezáč et al. 2021), our results support that morphotype A was the ancestral state for Canarian *Dysdera* (Fig. 5), and hence the ancestral coloniser was most likely a generalist species. The results of the former study, however, were compromised by the fragmentary evidence, including the use of linear measurements to quantify chelicera shape, sparse data ($n=1$ for many species) and the use of an outdated phylogeny. The more updated phylogeny and the use of geometric morphometrics that allows to capture shape variation at a finer scale, has most likely contributed to these dissonant results. This generalist ancestor in the Canarian *Dysdera*, is well aligned with the observation that specialist diets commonly evolved from generalist ones (Toft 1995; Stephens and Wiens 2003; Reynolds et al. 2016; Naik et al. 2021). Moreover, generalists are usually considered better colonizers, as they are able to more easily secure sufficient resources (Ebenhard 1991).

The evolution of these trophic adaptations during the diversification of the *Dysdera* species is, to some extent, related to their shared phylogenetic history, as reflected by significant phylogenetic signal observed for cheliceral shape. This result should not be surprising given the recent shared evolutionary history of these species and the lack of independence that this entails (Felsenstein 1988; Harvey and Pagel 1991; Revell et al. 2008). However, both the low values of K observed (<1), and the lack of phylogenetic structure that is apparent in the phylomorphospace plot (Fig. 3), are suggestive of distantly related species occupying closer phenotypic spaces than expected by chance. Accordingly, the ancestral reconstruction of morphotypes showed that the specialised slightly elongate, concave chelicera and the flat fang—morphotypes B, G, and F respectively—evolved several times, independently. The multiple evolution of at least three distinct cheliceral morphotypes (B, G and F), points towards a role of natural selection in the diversification of the group. The repeated evolution of convergent ecological and/or

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morphological traits is a common pattern in adaptive radiations, and is generally interpreted as a response to similar evolutionary pressures (Schluter and Nagel 1995; Losos et al. 1998, 2003; Rundle et al. 2000; Schluter 2000; Nosil et al. 2002; Blackledge and Gillespie 2004; Gillespie 2004; Gavrillets and Losos 2009; Muschick et al. 2012). Integrating evidence on convergent evolution with the statistical association between cheliceral shape and levels of trophic specialization (Fig. 4a), provides evidence that adaptations to increase woodlice consumption played a key role in the evolution and diversification of *Dysdera* in the Canary Islands. On the other hand, the morphotype D was found to be synapomorphic for the clade of endemic species that inhabits the eastern Canary Islands, which reveals the phylogenetic conservation of this trait in this group. In this case, some kind of environmental filtering may be playing an evolutionary constraint preventing the evolution or surviving of species with a more specialist chelicera.

Did the evolution of diet specialisation lead to an evolutionary irreversibility?

Although several studies have shown that transitions from specialist to generalist forms are indeed possible (Lanyon 1992; Muller 1996; Armbruster and Baldwin 1998), the reverse seems to be the common trend (Nosil 2002). Our character reconstruction analysis confirms the later observation, since the model that best fit our data encompassed transitions from generalist to specialist morphotypes but not the opposite, supporting the hypothesis that evolution of specialised chelicerae from generalist ancestors is the common pattern across Canarian *Dysdera* species. However, the convergence test gave a significative result for the repeated evolution of cheliceral morphotype A, related with a generalist diet. In the PCA, we can see that the internal nodes of cheliceral type A are outside the morphospace occupied by these species (Fig. 3), giving a positive result in convergence when tested for this cheliceral group, although there is an irreversibility in the specialization in the *Dysdera* species from the Canary Islands. In previous

studies, irreversibility has been tested in species with clear trophic preferences or well defined specializations (Day et al. 2016). In such cases, a compatibility between convergence of the ancestral state with an irreversibility of the derived state seems incongruent. Nevertheless, specialization in *Dysdera* seems to be facultative and gradual, and the phenotypic adaptations to these trophic preferences are translated in a continuum in shape variation, blurring the boundaries between specialized and generalist species.

Conclusions

Canarian *Dysdera* evolved a particular set of chelicerae shapes associated to different levels of diet specialisation, specifically to capture and feed on woodlice. Our implementation of geometric morphometrics approaches discerned cheliceral morphologies at finer scales than traditional measurements, as illustrated by the higher number of morphotypes compared to the ones described until now. Furthermore, the use of a newly inferred phylogenetic hypothesis based on mitogenomic data for a complete data set of Canarian endemics, revealed a new scenario for the phenotypic and ecological diversification of *Dysdera* in the Canary Islands. The ancestral coloniser of the archipelago was most likely a generalist species, from which specialised forms evolved convergently multiple times. The eastern and western islands harbour different cheliceral morphotypes, which hints at the existence of different ecological and evolutionary filters on each island group, probably related to the dramatic climatic and landscape changes undergone by the older and highly eroded Eastern Canaries. Our analysis provide support for a generalist origin in the diversification of the red devil spiders in the archipelago and specialisation as a derived trait that could lead to evolutionary irreversibility. Finally, we demonstrate how the categorization of trophic or phenotypic adaptations that are part of a

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continuum that blurs the boundaries between different groups, convincingly reconciles
evolutionary convergence and irreversibility.

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Author contributions

AB, MA and AK designed the study; AB and AK performed data analyses; SAS and MA conducted the “*de novo*” mitogenomic sequences and reconstructed the phylogeny; NMH and ST performed the prey preference experiments; AB wrote the first version of the manuscript. All authors reviewed and approved the final version of the manuscript.

Supplementary material

Data available from the Dryad Digital Repository: [http://dx.doi.org/10.5061/dryad.\[NNNN\]](http://dx.doi.org/10.5061/dryad.[NNNN])

Conflict of interest

The authors declare no competing financial interests.

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Figures

Figure 1 a) Map of the Canary Islands with the proportion of species of each cheliceral morphotype present in each island, as inferred through a clustering of species mean shapes. Numbers indicate the number of species analysed and the total number of species present in each island. b) Combined view of the clustering phenogram performed on pairwise distances among species means of the combined landmark configuration of the three cheliceral views, and the significance of pairwise comparisons of these distances. Black squares indicate no significant differences between pairs of species (p -value > 0.05), white squares indicate significant differences (p -value < 0.05). The horizontal dashed line across the top phenogram delimits the cheliceral morphotypes considered, which are highlighted by different colours (corresponding to letters A-I).

Figure 2 Male individual of *Dysdera simbeque* from Lanzarote (Photo credit M.A.) (a) and position of the landmarks (red) and semilandmarks (blue) considered on the dorsal (b) and ventral (c) view of the chelicera and the fang (d).

Figure 3 Cheliceral phylomorphospace with convex polygons depicting species of the same ecomorph group. Black dots represent the *Dysdera* species analysed, and the coloured dots correspond to the mean shape of each group. Deformation grids represent the mean shape of each group as compared to the global mean of all species.

Figure 4 a) PLS analysis between prey preference and cheliceral shape variation. Cheliceral morphotypes are represented with the colored dots. Deformation grids correspond to the minimum and maximum shape of the PLS axis for each cheliceral view. Shape patterns are magnified x1.3 to enhance visualization. b) Prey acceptance preference between generalist and specialist species. “Kmeans clustering” represents the species cluster delimitation made by the *kmeans* function. The “G/S cluster analysed” has been the clustering analysed made avoiding splitting species that belong to the same cheliceral morphotype.

Figure 5 Ancestral state reconstruction obtained under the constrained (irreversible) ER model. Pie-charts at each node represent the state probability obtained through the 100 stochastic character map simulations over 1000 phylogenetic trees sampled from the posterior distribution of BEAST.

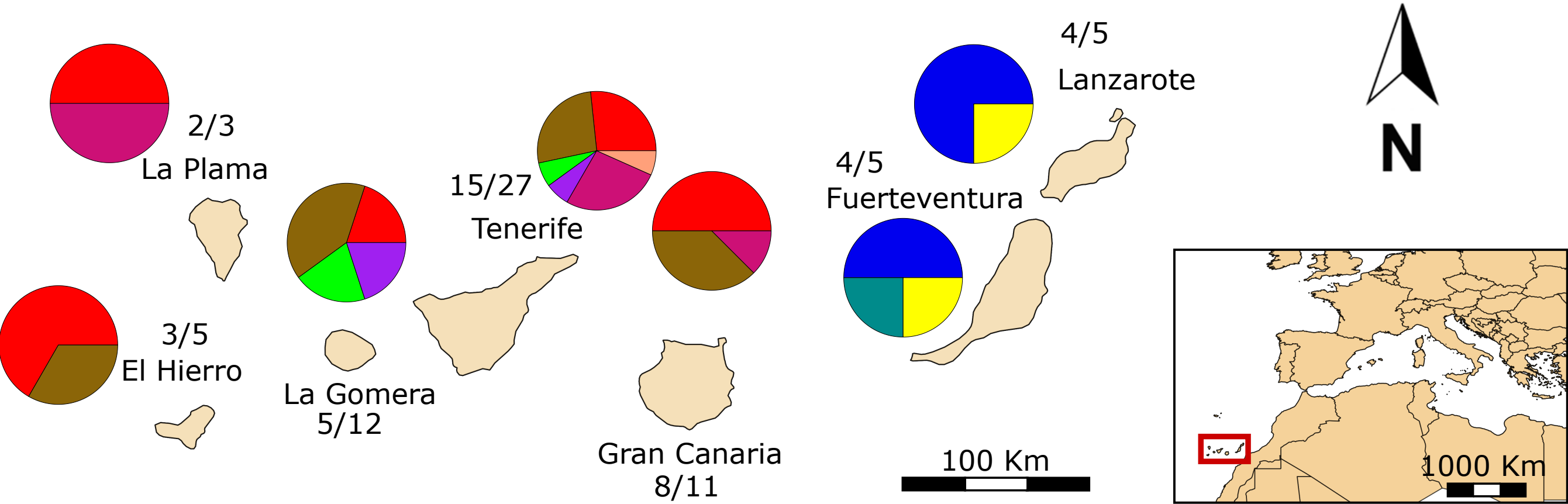
Appendices

Appendix 1. Supplementary Tables

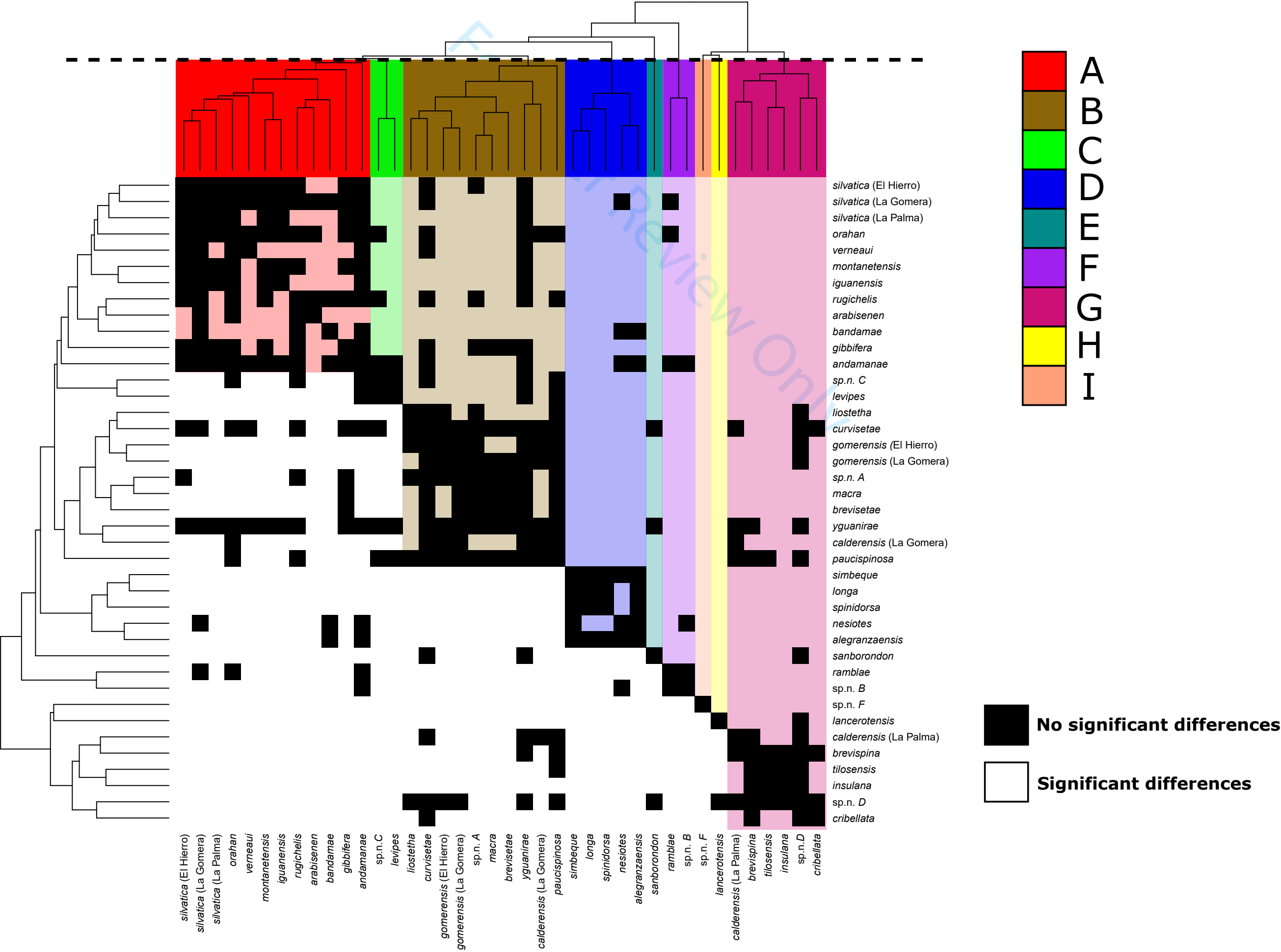
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- 924 **Table S1** Summary of the fossil and biogeographic calibrations used in the present study, with information on their
 925 implementation
- 926 **Table S2** Percentage of acceptance of each prey by each *Dysdera* species analysed
- 927 **Table S3** PLS eigenvector composition
- 928 **Table S4** Estimated total number of transitions from X to Y (upper matrix) and Y to X (lower matrix) for the constrained ER model
- 929 **Table S5** Estimated total number of transitions from X to Y (upper matrix) and Y to X (lower matrix) for the ER model
- 930 **Figure S4** Ancestral state reconstruction with ER model
- 931 **Table S6** Estimated total number of transitions from X to Y (upper matrix) and Y to X (lower matrix) for the SYM model
- 932 **Table S7** Estimated total number of transitions from X to Y (upper matrix) and Y to X (lower matrix) for the *ARD model*
- 933 **Appendix 2. Supplementary figures**
- 934 **Figure S1** Shape corresponding to the maximum and minimum extremes of PC1 for each cheliceral view, represented using
 935 deformation grids. Shape patterns are exaggerated by 1.3x to enhance visualization.
- 936 **Figure S2** Shape corresponding to the maximum and minimum extremes of PC2 for each cheliceral view, represented using
 937 deformation grids. Shape patterns are exaggerated by 1.3x to enhance visualization.
- 938 **Figure S3** *Time-stamped phylogeny including all described Dysdera species from the Canary Islands inferred from*
 939 *mitogenome data*
- 940 **Figure S4** Ancestral state reconstruction with ER model
- 941 **Figure S5** Ancestral state reconstruction with SYM model
- 942 **Figure S6** Ancestral state reconstruction with ARD model
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a)



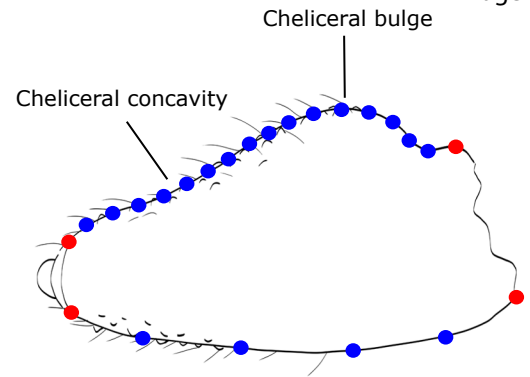
b)



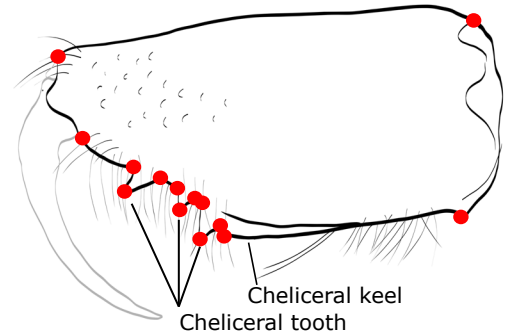
a)



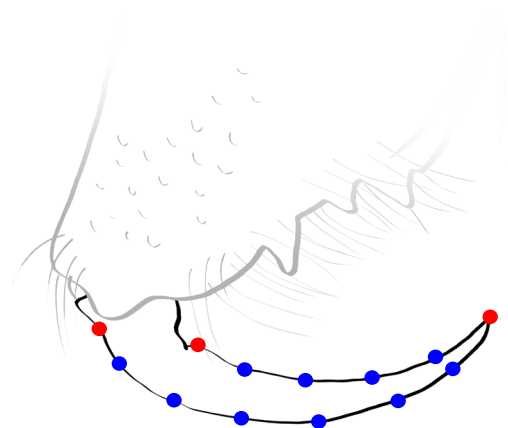
b)

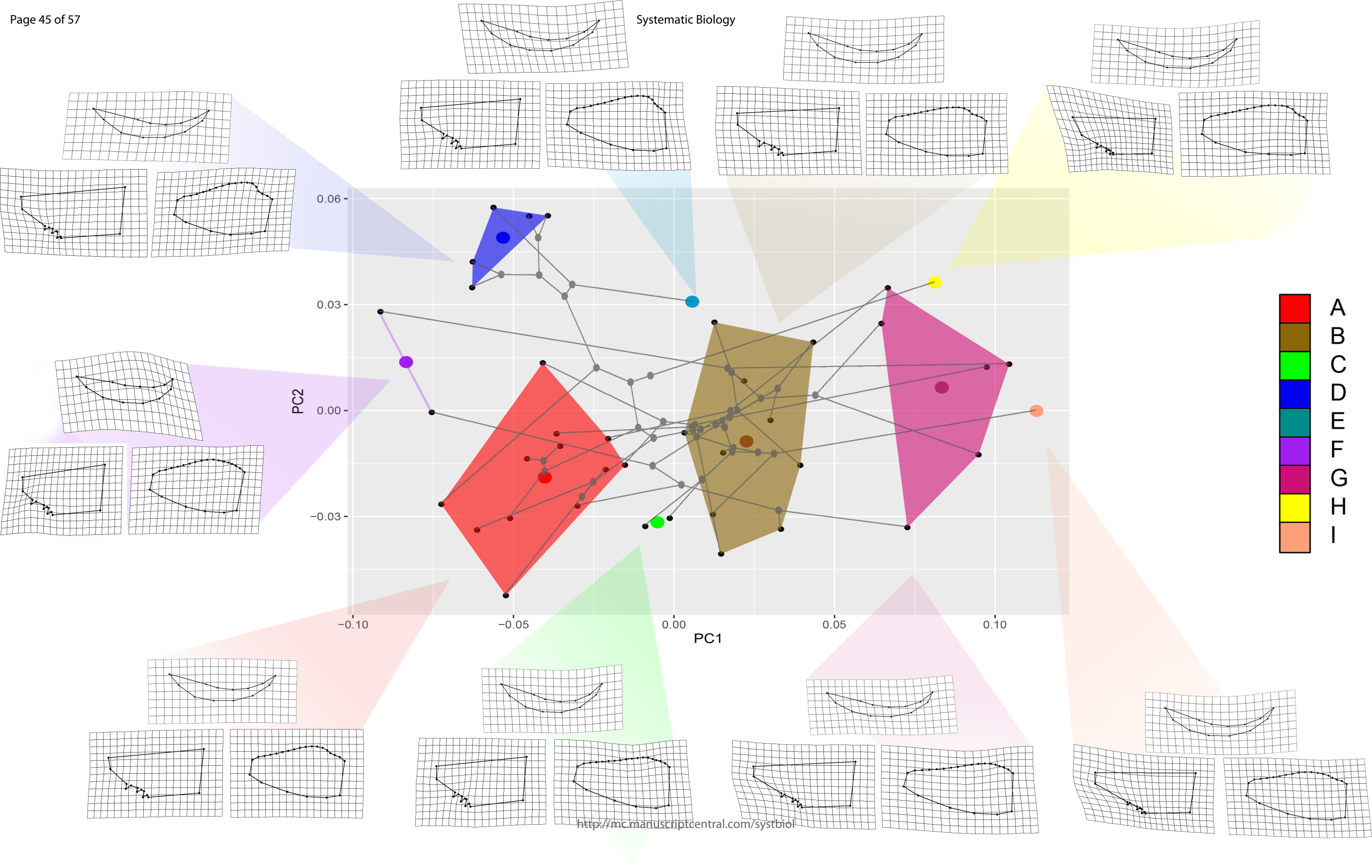


c)

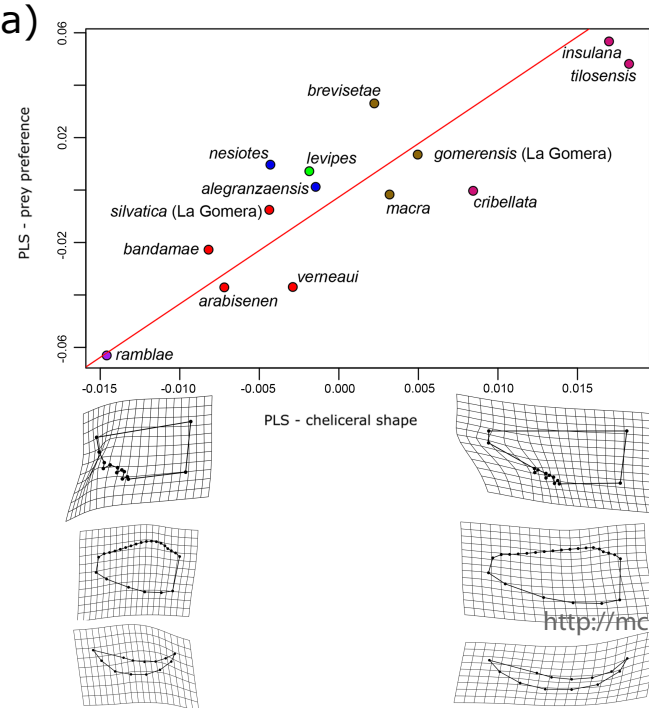


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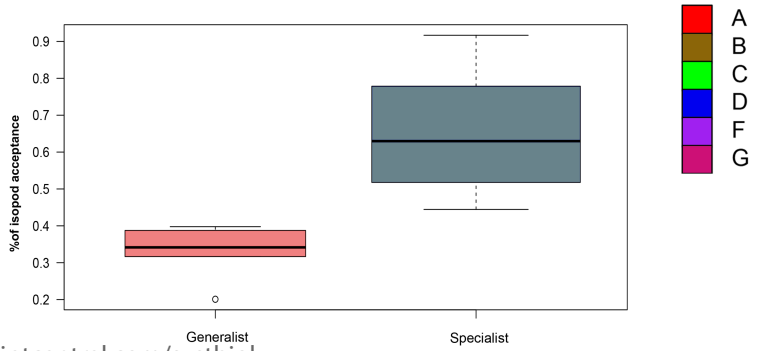




Systematic Biology



Species	<i>arabisenen</i>	<i>bandamae</i>	<i>silvatica</i>	<i>vermeaui</i>	<i>alegranzaensis</i>	<i>nesiotes</i>	<i>insulana</i>	<i>brevisetae</i>	<i>cribellata</i>	<i>macra</i>	<i>gomerensis</i>	<i>tilosensis</i>	<i>ramblae</i>	<i>levipes</i>
%of isopod acceptance	0.200	0.316	0.316	0.366	0.387	0.397	0.444	0.491	0.544	0.592	0.666	0.694	0.863	0.916
Cheliceral morphotype	A	A	A	A	B	B	C	D	E	E	E	F	F	G
Kmeans clustering	Cluster 1									Cluster 2				
G/S cluster analysed	Cluster 1									Cluster 2				



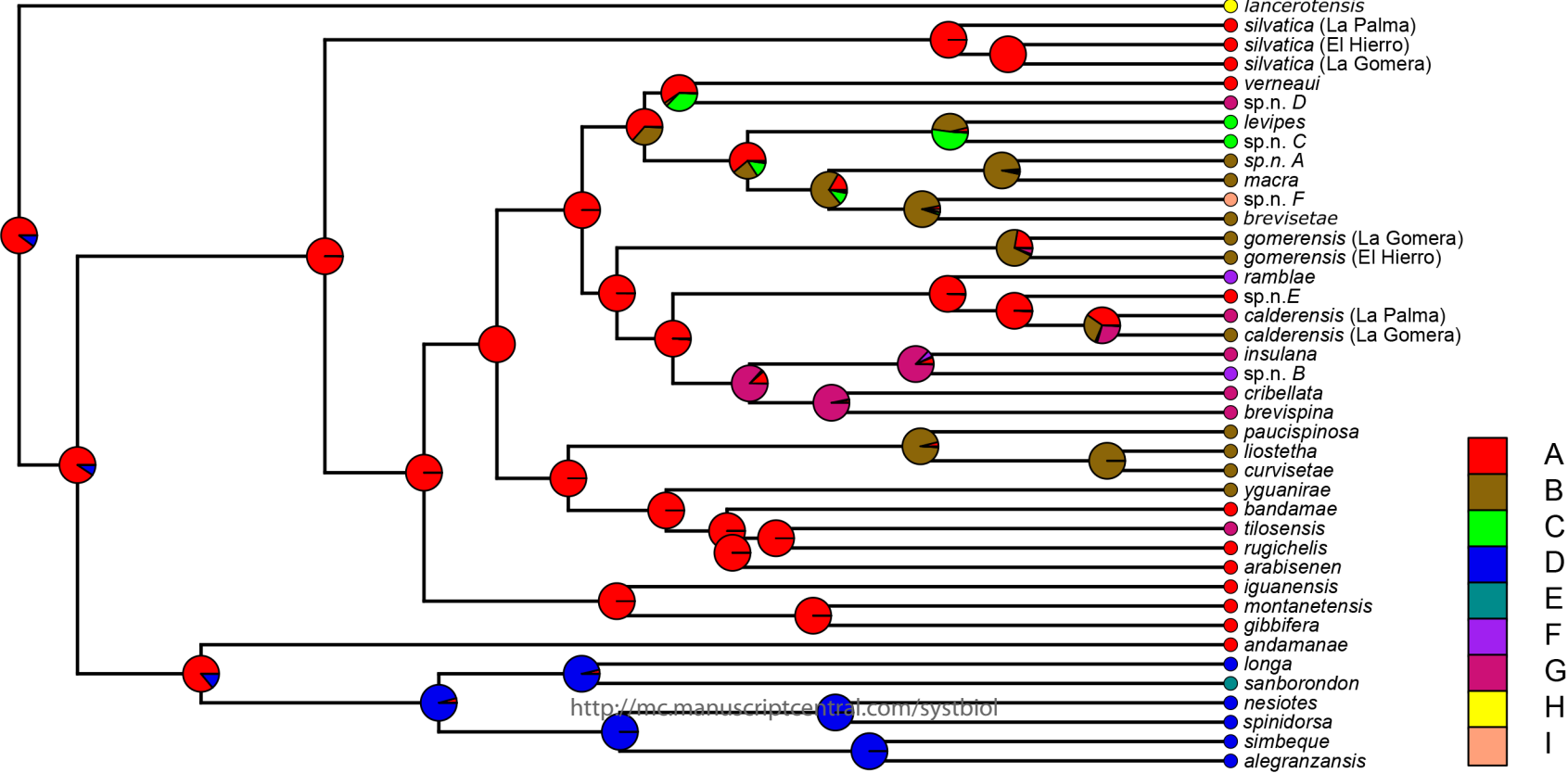


Table S1 Summary of the fossil and biogeographic calibrations used in the present study, with information on their implementation

CLADE	FOSSIL	MIN.AGE	MAX.AGE	ASSIGNABLE TO	PRIOR DISTRIBUTION	UPPER	LOWER	MEAN IN REAL SPACE	SD.	OFFSET	MEDIAN	97.5% HPD	REFERENCE
Mesothelae	<i>Palaeothela montceauensis</i> (Selden)	299	304	Mesothelae stem	Uniform	450	299*						Magalhaes et al. 2020
Mygalomorphae	<i>Rosamygale grauvogeli</i> Selden & Gall	242	247.2	Avicularioidea stem	lognormal			80	0.5	242.0	313	268-430	Magalhaes et al. 2020
Synspermiata	<i>Eoplectreurys gertschi</i> Selden & Huang	164	175.1	Synspermiata stem	lognormal			70	0.5	165.0	226	187-329	Magalhaes et al. 2020
Synspermiata	* <i>Montsecarachne amicum</i> Selden	125	129.4	Synspermiata crown	lognormal			70	0.5	125.0	187	148-290	Magalhaes et al. 2020
Synspermiata	* <i>Burmorchestia acuminata</i> Wunderlich	98.17	99.41	Oonopidae stem	lognormal			30	0.5	98.17	125	108-169	Magalhaes et al. 2020
Synspermiata	<i>Priscalecerera paucispinae</i> Wunderlich	98.17	99.41	Psilodercidae stem	lognormal			30	0.5	98.17	125	108-169	Magalhaes et al. 2020
Araneoidea	'Linyphiinae' indet.	125	135	Araneoidea stem	lognormal			70	0.5	125.0	187	148-290	Magalhaes et al. 2020
rta	* <i>Selenops</i> sp. indet.	53	56	Selenopidae stem	lognormal			20	0.75	53.0	68.1	56.5-119	Magalhaes et al. 2020
Synspermiata	<i>Dasumiana emicans</i> Wunderlich	43	47.8	Harpacteinae crown	lognormal			15	0.75	43.0	54.3	45.6-92.2	Magalhaes et al. 2020
Synspermiata	<i>Quamtana huberi</i> Penney	53	56	Pholcinae crown	lognormal			20	0.75	53.0	68.1	56.5-119	Magalhaes et al. 2020
Araneoidea	<i>Mesozygella dunlopi</i> Penney & Ortuño	115	121	Araneidae stem	lognormal			30	0.5	115.0	141	125-186	Penney & Ortuño 2006
Biogeographic even													
Parachtes	Hercinian split			Parachtes crown	normal			29	2.5		29	24.1-33.9	Bidegaray & Arnedo 2011
*EARLIEST EVIDENCE OF LAND ARTHROPODS (ARBITRARY UPPER CONSTRAINT ON ROOT)													

Table S2 Percentage of acceptance of each prey by each *Dysdera* species analysed

Species	n	Musca	Porcellio	Armadillidium	Pardosa	Carabid
<i>D. alegranzaensis</i>	18	93.75	87.5	43.75	68.75	31.25
<i>D. arabisenen</i>	19	100.00	35.29	29.41	35.29	11.76
<i>D. bandamae</i>	20	95.00	50.00	30.00	60.00	0.00
<i>D. brevisetae</i>	20	95.00	85.00	55.00	30.00	0.00
<i>D. cribellata</i>	20	78.95	94.74	36.84	57.89	0.00
<i>D. gomerensis</i> (La Gomera)	19	47.37	100.00	57.89	47.37	0.00
<i>D. insulana</i>	3	100.00	100.00	66.67	100.00	0.00
<i>D. levipes</i>	6	0.00	100.00	100.00	0.00	0.00
<i>D. macra</i>	19	73.68	94.74	42.11	21.05	10.53
<i>D. nesiotis</i>	20	100.00	90.91	40.91	72.73	9.09
<i>D. ramblae</i>	20	17.65	100.00	23.53	11.76	0.00
<i>D. silvatica</i> (La Gomera)	20	100.00	90.00	25.00	85.00	5.00
<i>D. tilosensis</i>	7	71.43	85.71	71.43	0.00	0.00
<i>D. verneuui</i>	20	100.00	75.00	15.00	60.00	10.00

Table S3 PLS eigenvector composition

Prey	Vector 1	Vector 2	Vector 3	Vector 4	Vector 5
Musca	0.46	-0.75	-0.45	0.14	0.00
Porcellio	0.38	0.14	0.35	0.63	0.56
Armadillidium	0.79	0.37	0.05	-0.48	-0.12
Pardosa	0.02	-0.52	0.80	-0.28	-0.07
carabid	-0.14	-0.09	-0.16	-0.53	0.82

Table S4 Estimated total number of transitions from X to Y (upper matrix) and Y to X (lower matrix) for the constrained ER model

X	A	B	C	D	E	F	G	H	I
A		4.05	0.83	1.14	0.32	1.22	3.41	0.77	0.31
B	0.00		0.46	0.08	0.08	0.08	0.46	0.11	0.82
C	0.00	0.35		0.05	0.05	0.05	0.10	0.08	0.06
D	0.00	0.16	0.07		0.75	0.09	0.14	0.11	0.07
E	0.00	0.12	0.03	0.09		0.05	0.10	0.07	0.03
F	0.00	0.12	0.03	0.05	0.05		0.16	0.07	0.03
G	0.00	0.44	0.06	0.07	0.07	0.82		0.10	0.06
H	0.00	0.12	0.03	0.05	0.05	0.05	0.10		0.03
I	0.00	0.16	0.04	0.04	0.05	0.05	0.10	0.07	

Table S5 Estimated total number of transitions from X to Y (upper matrix) and Y to X (lower matrix) for the ER model

X	A	B	C	D	E	F	G	H	I
A		10.01	8.53	8.93	8.37	8.52	9.57	8.56	8.36
B	9.48		8.75	8.47	8.32	8.52	9.27	8.33	8.77
C	8.78	8.76		8.43	8.27	8.30	8.49	8.27	8.24
D	8.94	8.69	8.34		8.65	8.33	8.52	8.33	8.27
E	8.75	8.67	8.29	8.46		8.28	8.49	8.27	8.24
F	8.80	8.69	8.29	8.41	8.25		8.53	8.27	8.23

G	9.13	9.18	8.36	8.46	8.27	9.05		8.30	8.27
H	8.80	8.67	8.28	8.43	8.24	8.30	8.47		8.24
I	8.76	8.66	8.28	8.42	8.25	8.28	8.49	8.26	

Table S6 Estimated total number of transitions from X to Y (upper matrix) and Y to X (lower matrix) for the SYM model

	X	A	B	C	D	E	F	G	H	I
Y										
A			1.55	0.16	0.77	0.07	0.46	3.79	0.78	0.16
B		1.36		0.44	0.19	0.01	1.20	24.05	0.05	1.12
C		0.14	0.03		4.34	20.49	0.70	0.01	1.77	4.23
D		0.13	0.18	4.09		11.15	0.41	0.00	6.95	1.68
E		0.01	0.00	20.51	10.63		0.23	0.00	2.14	1.10
F		0.50	0.91	1.03	0.44	0.24		0.66	0.23	1.13
G		4.31	26.06	0.10	0.01	0.01	2.76		0.01	0.12
H		0.16	0.02	1.79	7.03	2.15	0.18	0.00		1.29
I		0.17	0.23	4.88	1.75	1.14	0.63	0.01	1.34	

Table S7 Estimated total number of transitions from X to Y (upper matrix) and Y to X (lower matrix) for the ARD model

	X	A	B	C	D	E	F	G	H	I
Y										
A			0.06	0.64	4.61	0.99	0.33	0.60	2.33	0.01
B		0.61		2.33	0.79	0.45	2.51	37.54	0.77	2.56
C		2.72	3.02		1.44	1.45	4.37	3.77	2.08	2.77
D		4.16	1.74	1.46		2.32	1.34	1.32	1.20	0.53
E		1.65	1.30	1.37	1.65		1.15	1.24	1.43	0.76
F		1.83	3.42	4.96	1.22	1.63		4.60	1.97	2.92
G		1.12	30.69	6.21	2.20	1.36	8.55		1.91	3.61
H		2.25	1.88	1.93	1.35	1.47	1.45	1.80		0.95
I		1.38	2.76	2.49	0.66	0.93	2.39	2.48	1.19	

Additional references

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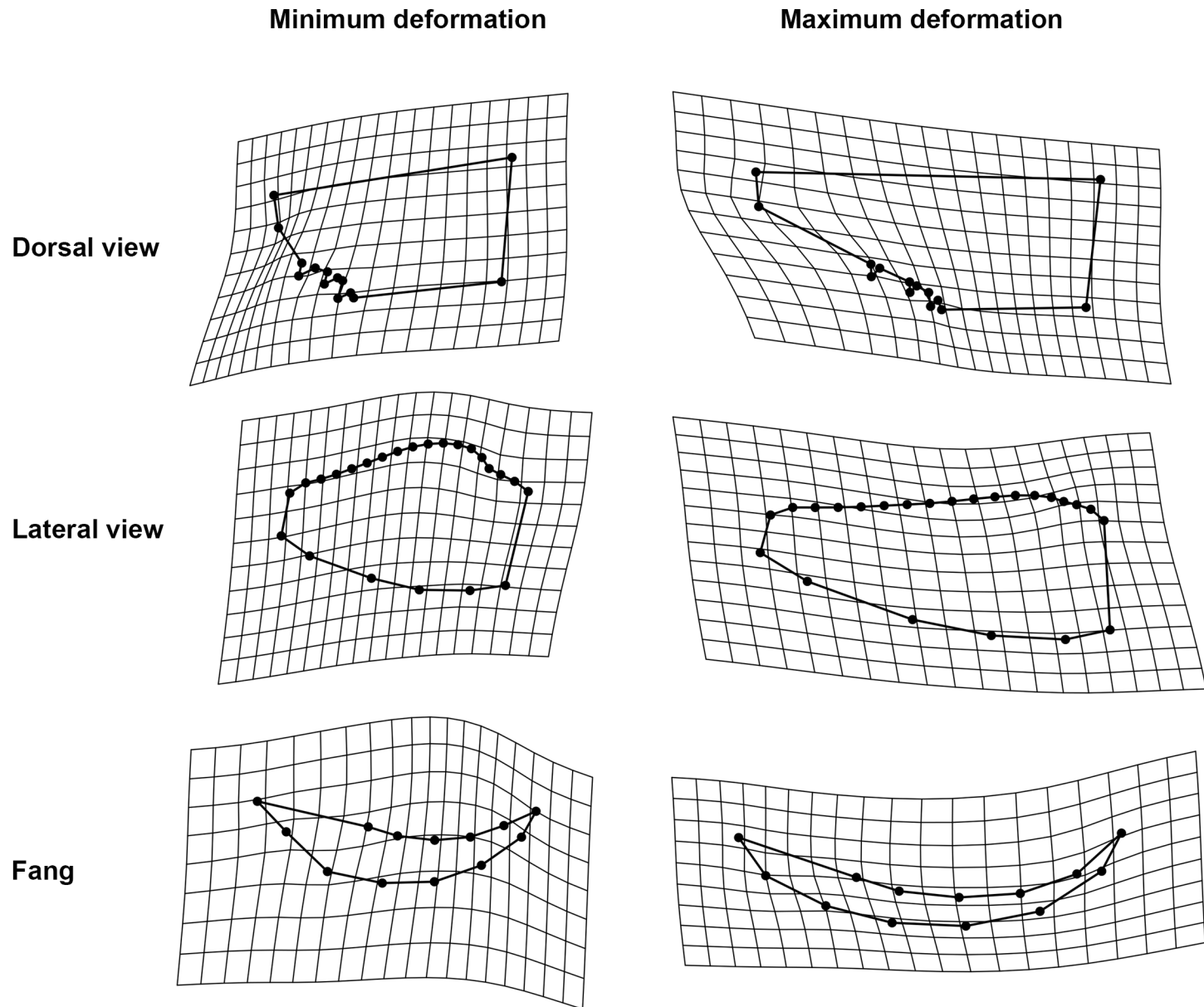
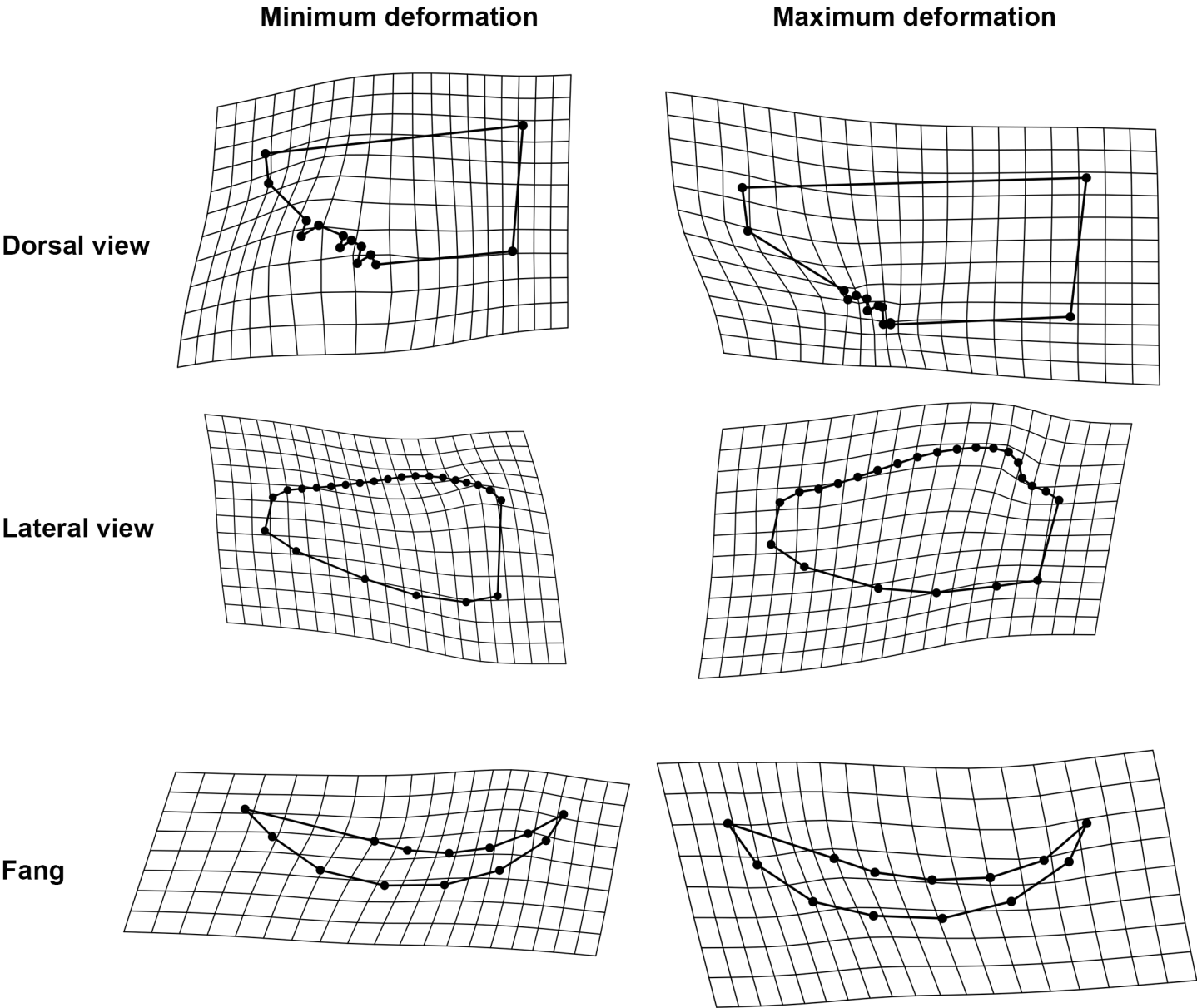
Fig. S1

Fig. S2



[illegible]

Eastern Canary Islands

Fig. S4

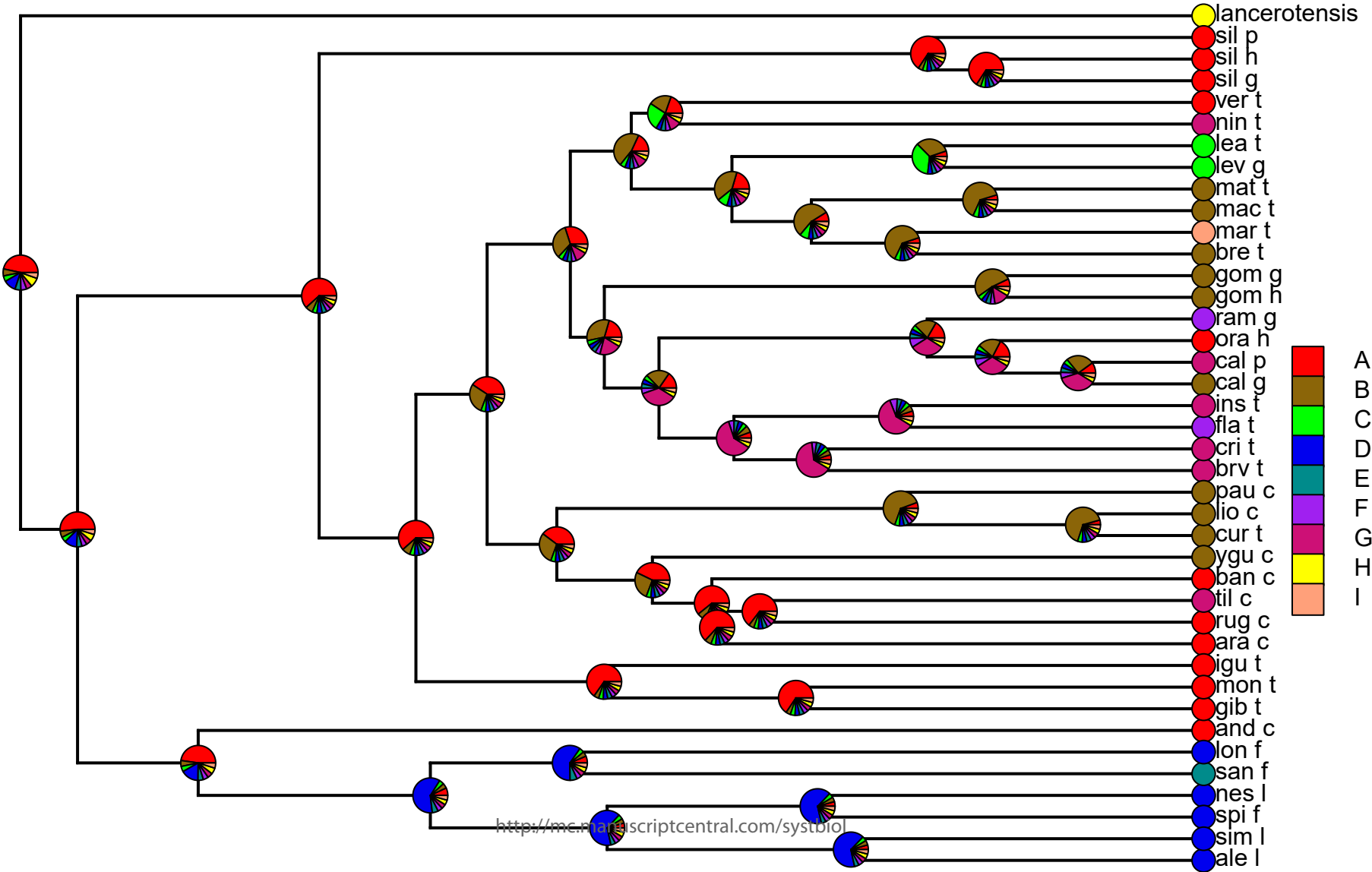


Fig. S5

