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ARTICLE

Unexpected discovery of a near cryptic *Dasypoda* species in southern Spain (Hymenoptera: Melittidae)

Guillaume GHISBAIN[1] • **Denis MICHEZ**[2] • **Paolo ROSA**[3] • **Sónia FERREIRA**[4] • **Thomas J. WOOD**[5]

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Abstract
A new species of pantaloon bee, *Dasypoda* (*Heterodasypoda*) *radchenkoi* GHISBAIN & WOOD sp. nov., is described from both males and females collected in the limestone mountains of south-western Spain. COI barcodes show an important differentiation from *D. morotei* QUILIS, 1928 of approximately 9.6% but the morphological divergence between the two species is extremely low. *Dasypoda radchenkoi* sp. nov. is the fifth species of *Heterodasypoda* found in Iberia, further confirming the peninsula as the center of diversity for the subgenus.

Keywords | Anthophila • Apoidea • taxonomy • Iberian Peninsula • endemic species • subgenus *Heterodasypoda*

Découverte inattendue d'une espèce cryptique de *Dasypoda* dans le sud de l'Espagne (Hymenoptera : Melittidae)

Résumé
Une nouvelle espèce de dasypode, *Dasypoda* (*Heterodasypoda*) *radchenkoi* GHISBAIN & WOOD sp. nov., est décrite à partir de mâles et femelles collectés dans les montagnes calcaires du sud-ouest de l'Espagne. Des codes-barres de la COI montrent une importante différenciation avec *D. morotei* QUILIS, 1928 d'environ 9.6 % mais la divergence morphologique des deux espèces est très faible. *Dasypoda radchenkoi* sp. nov. est la cinquième espèce d'*Heterodasypoda* trouvée sur la Péninsule ibérique, confirmant que ce lieu est le centre de diversité du sous-genre.

Mots-clefs | Anthophila • Apoidea • taxonomie • Péninsule Ibérique • espèce endémique • sous-genre *Heterodasypoda*

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Supplementary materials (on the website of the journal + archived in *Zenodo*) [https://doi.org/10.47446/OSMIA11.6]
Table S1. Sampling information on newly sequenced individuals [saved at https://doi.org/10.5281/zenodo.8172745]

INTRODUCTION

With a tenth of the global diversity of wild bees (Hymenoptera: Anthophila), the European continent is a playground for understanding the ecology, diversity, and conservation of this strikingly diverse insect group (GHISBAIN *et al.*, 2020; RASMONT *et al.*, 2021; WOOD *et al.*, 2022). Although the continent constitutes the most studied place for wild bee biology and taxonomy worldwide, we are still far from having fully examined and investigated all European regions (MICHEZ *et al.*, 2019).

The need for increased taxonomic and monitoring work across the continent is particularly well exemplified in southern and eastern European countries, for which national species totals are currently underestimated (NIETO *et al.*,

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[1] [GG] Spatial Epidemiology Lab (SpELL), Free University of Brussels, CP 264/3, avenue F. D. Roosevelt 50, B – 1050 Brussels, Belgium
+ Laboratory of Zoology, Research Institute for Biosciences, University of Mons, Place du Parc 20, B – 7000 Mons, Belgium • guillaume.ghisbain@umons.ac.be
https://orcid.org/0000-0003-2032-8081 • ZooBank https://zoobank.org/FD9CD87B-EAAB-4037-BFDE-58AE0E7349B7

[2] [DM] Laboratory of Zoology, Research Institute for Biosciences, University of Mons, Place du Parc 20, B – 7000 Mons, Belgium • denis.michez@umons.ac.be
https://orcid.org/0000-0001-8880-1838 • ZooBank https://zoobank.org/8B04585A-FE00-4D9A-AFD6-1BD2A1584CFA

[3] [PR] Laboratory of Zoology, Research Institute for Biosciences, University of Mons, Place du Parc 20, B – 7000 Mons, Belgium • paolo.rosa@umons.ac.be
https://orcid.org/0000-0003-2919-5297 • ZooBank https://zoobank.org/F1E8DF8A-6105-4AC8-96F9-31EDA674D320

[4] [SF] CIBIO, Centro de Investigação em Biodiversidade e Recursos Genéticos, InBIO Laboratório & BIOPOLIS Program in Genomics, Biodiversity and Land Planning Associado, Campus de Vairão, Universidade do Porto, P – 4485-661 Vairão, Portugal • hiporame@gmail.com
https://orcid.org/0000-0002-6884-3966 • ZooBank https://zoobank.org/931C5D9F-C380-4D37-9D3F-6C3C9A657877

[5] [TJW] Laboratory of Zoology, Research Institute for Biosciences, University of Mons, Place du Parc 20, B – 7000 Mons, Belgium • thomasjames.wood@umons.ac.be
https://orcid.org/0000-0001-5653-224X • ZooBank https://zoobank.org/37B8E7E4-5BA0-43AF-82AB-A86B4F3C8A3B

2014; RASMONT *et al.*, 2017). This is especially true for Mediterranean countries that display the climate type for which bee diversity is the highest globally (ORR *et al.*, 2021). With its combination of mountains, grasslands, coastal scrub, and forests, the Mediterranean basin constitutes a highly diversified mosaic of natural landscapes that host a large variety of bee communities, including a substantial proportion of endemic species (MICHENER, 2007; MICHEZ *et al.*, 2019). This can be explained by a unique conjunction of orography and climate, which led it to be considered as one of the 25 Global Biodiversity Hotspots (MYERS *et al.*, 2000). This high diversity, combined with relatively low historic and contemporary sampling effort across the region explains why discoveries and descriptions of new Mediterranean bee species are still underway (*e.g.*, RADCHENKO, 2017; WOOD & CROSS, 2017; KUHLMAN & SMIT, 2018; WOOD *et al.*, 2020, 2021).

Among the six bee families represented in the Mediterranean basin is the family Melittidae, a species-poor group including the pantaloon bees of the genus *Dasypoda* LATREILLE, 1802 (MICHEZ *et al.*, 2004a–b, 2008, 2009). This genus, remarkable by the presence of strongly developed pollen-carrying hair structures (scopae) on the tibiae and basitarsi of the hind legs of the females (MICHENER, 2007), has received renewed taxonomic attention in recent years. Two new cryptic species were described by RADCHENKO (2016, 2017) and additional surveys have further characterized their distribution, ecology, and conservation (RADCHENKO *et al.*, 2020; GHISBAIN *et al.*, 2021b). These steps are essential for the effective conservation of the group, as the combination of unambiguous taxonomic delineation and the accumulation of precise occurrence records constitute the starting point for accurate Red List assessments (GHISBAIN *et al.*, 2021 a–b; NIETO *et al.*, 2014).

In this work, we report the unexpected discovery of a new species of pantaloon bee from south-western Spain.

MATERIAL AND METHODS

The specimens used for the description of the new species were all collected as part of a field trip in south-western Spain conducted by GG and TJW during May and June 2021. Reference specimens of the other *Heterodasypoda* species are stored in the Laboratory of Zoology of the University of Mons (UMons). Some fresh specimens were used to sequence a barcode fragment of the cytochrome oxidase I gene (*COI*), with comparative sequences available in *GenBank* for all other described species of *Heterodasypoda* (GHISBAIN *et al.*, 2021b).

Genetic differentiation

We sequenced a fragment of the cytochrome c oxidase I (*COI*) gene from one male (paratype) and one female specimen (holotype) of our new species. DNA extraction and sequencing followed the general pipeline in use by the InBIO Barcoding Initiative. Genomic DNA was extracted from a middle leg of each individual using the EasySpin Genomic DNA Microplate Tissue Kit (Citomed, Odivelas, Portugal) according to the manufacturer's protocol. The cytochrome c oxidase I (*COI*) barcoding fragment was then amplified as two overlapping fragments (*LC* and *B2*), using two sets of primers: LCO1490 (FOLMER *et al.*, 1994) + Ill_C_R (SHOKRALLA *et al.*, 2015) (325 bp fragment) and BF2 + BR2 (422 bp fragment, ELBRECHT & LEESE, 2017). The PCR products were then sequenced in a MiSeq benchtop system. OBITools (https://git.metabarcoding.org/obitools/obitools) was used to process the initial sequences which were then assembled into a single 658 bp fragment using *Geneious 9.1.8.* (https://www.geneious.com). Additional sequences were generated by the Canadian Center for DNA barcoding (CCDB) in Guelph, Canada following their high throughput protocols (IVANOVA *et al.*, 2006) using the BeeCox1F1 primer (BLEIDORN & HENZE, 2021). After sequencing and alignment of the cleaned sequences, we ran a neighbor-joining phylogeny with 10,000 bootstraps including our new species *D. radchenkoi* sp. nov. (*n* = 2) along with representatives of each species of *Heterodasypoda*: *D. michezi* RADCHENKO, 2017 (*n* = 7), *D. morotei* QUILIS, 1928 (*n* = 6), *D. pyrotrichia* FÖRSTER, 1855 (*n* = 2), and *D. albimana* PÉREZ, 1905 (*n* = 3). We chose the more distant *Dasypoda* (*Dasypoda*) *morawitzi* RADCHENKO, 2016 as an outgroup. As the new species shows high phenotypic similarity with *D. morotei*, we included three near topotypic individuals of the latter from (i) the Madrid region (accession number MW401793) and (ii) the Sistema Central Mountain range, specifically from near to Aldeanueva de Atienza and Bustares (province of Guadalajara; accession numbers IBIHM1332-22 and IBIHM1331-22, respectively) which are 100 km to the north-east of the locus typicus of San Rafael in the Sierra de Guadarrama (province of Segovia) (QUILIS, 1928). We also include a *D. morotei* specimen from the Vale de Prazeres (Castelo Branco district, Portugal; accession number IBIHM882-21) which sits at the western end of the Sistema Central, thus covering the longitudinal extent of this mountain range (see Supplementary materials table S1: sampling information on newly sequenced individuals).

Taxonomic nomenclature

Based on the results of the COI barcoding, we further examined the morphology of the new species with all other *Heterodasypoda* species (*Dasypoda albimana*, *D. michezi*, *D. morotei* and *D. pyrotrichia*). High-resolution pictures of both sexes of the new species were taken using a Camera Olympus E-M1 Mark II with the Olympus Zuiko 60 mm objective and stacked with the software *Helicon Focus* (version 7.6); further image processing was completed with *Adobe Photoshop CS6.0* software program.

The following abbreviations (following the nomenclature of MICHENER, 2007) were used for morphological structures: A1, A2, … = first, second, … antennomere; S1, S2, … = first, second, … metasomal sternum; T1, T2, etc. = first, second, … metasomal tergum; L = length; W = width.

Conservation assessment

In the framework of an update of the last European Red List of Bees (see NIETO *et al.*, 2014), we assessed an IUCN status for *Dasypoda radchenkoi* sp. nov. following the Guidelines for using the IUCN Red List Categories and Criteria V. 15.1 (https://www.iucnredlist.org/resources/redlistguidelines, updated in July 2022).

RESULTS

Genetic results

The resulting phylogenetic tree based on *COI* is shown in figure 1. Branch support and length for *Dasypoda radchenkoi* sp. nov. unambiguously support its differentiation with all other species belonging to the subgenus *Heterodasypoda* and confirm the sex association between the holotype female and a male paratype. The close placement of the new species relative to *Dasypoda morotei* is in line with their strong phenotypic similarity. New sequences are available on *BOLD* with the accession numbers reported in figure 1.

Taxonomy and ecology

Dasypoda (*Heterodasypoda*) *radchenkoi*

GHISBAIN & WOOD, 2023 sp. nov.

ZooBank https://zoobank.org/FC629A42-7687-40C5-BD01-E7447BDA3B5E

Female

Head (figure 2d). Clypeus covered with whitish pilosity; supraclypeal and paraocular areas covered with long and erect whitish pilosity intermixed with black hairs, with narrow band of black hairs along internal margin of compound eye. Clypeus strongly punctured over its surface, with or without median impunctate carina, clypeal punctures denser basally and almost confluent, separated by less than one puncture diameter, becoming slightly sparser apically. Frons and vertex covered with sparse, erect, black pilosity, becoming pale posteriorly. Lateral parts of vertex, between lateral ocellus and compound eye, matt and densely shagreened (figure 2e). Gena weakly shiny with dense punctation, covered with black pilosity dorsally, becoming whitish distally. Malar space short, linear, clearly narrower than diameter of flagellum (figure 2f). Mandible strongly shagreened basally, becoming shiny medially, shagreened distally, its cuticle becoming weakly reddish medially and black elsewhere. Basal part of mandible with whitish hairs externally. Galea fully covered with small-rounded grains, surface dull. Scape ventrally covered with sparse whitish and black pilosity. Pedicel and flagellum cylindrical, with black cuticle and covered with very short whitish pilosity. A3 clearly longer than A4, even slightly exceeding A4+5 (figure 2c).

Mesosoma (figures 2a–b, h–I, 4b–d). Scutum shiny and strongly punctured medially, distance between points equal to one puncture diameter or smaller, and clearly more shagreened laterally (Figure 2h). Hairs on scutum black centrally and whitish-brown laterally, so that the darker pilosity appear to be a spot on scutum between wings (figures 2a–b). Scutellum shiny and densely punctured; propodeum with granular shagreen over majority of its area, shagreen transitioning into subtly but distinctly raised ridges covering basal half (figure 2i). Tegula dark brown and finely shagreened. Wing transparent, hyaline. External face of profemur and tibia covered with grey pilosity, except the apical part of internal face of femora with orangish pilosity. Mesofemur with light grey pilosity; external face of mesotibia basally with whitish hairs and apically with a tuft or adpressed orange pilosity, internal face with black pilosity. Mesobasitarsus with whitish pilosity on external face except dorsally and apically where pilosity is grey. Metafemur with whitish pilosity. Scopa on metatibia and metabasitarsus uniformly orange. Inner part of the pro- and mesobasitarsus with dense brush of bright orange pilosity. Metabasitarsus

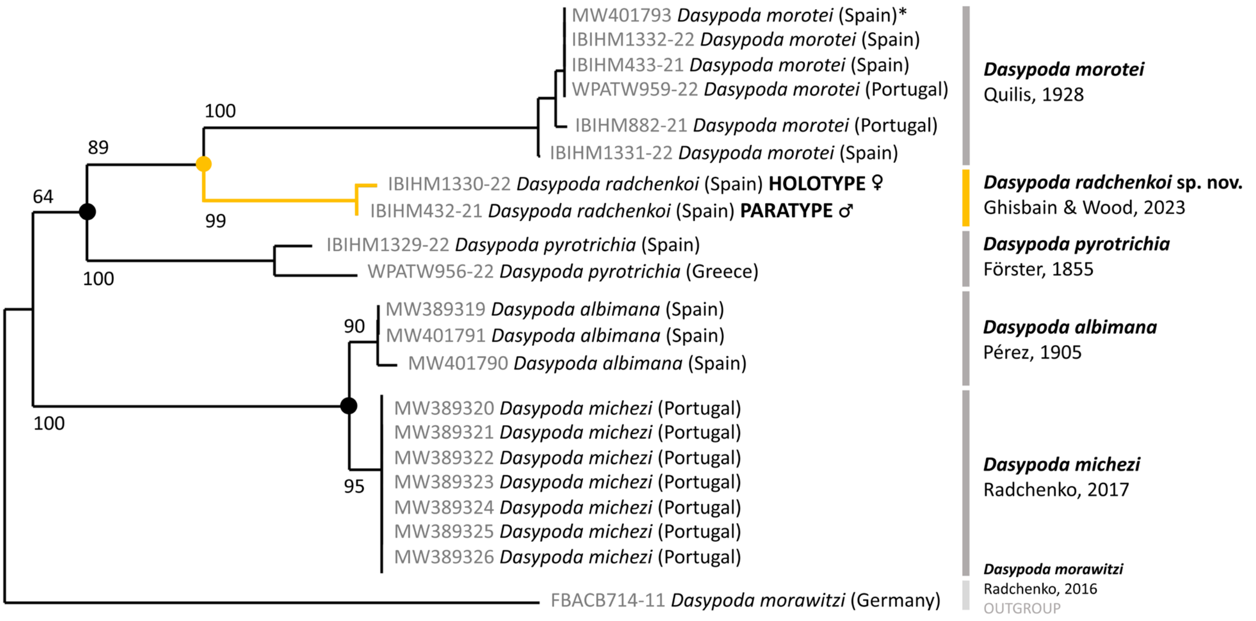

Figure 1. COI phylogeny of the five *Heterodasypoda* species, highlighting the separation of the newly discovered species *Dasypoda radchenkoi* sp. nov. from the closely related *Dasypoda morotei* QUILIS. The asterisk highlights the specimen of *D. morotei* that was collected close to the locus typicus. *Dasypoda* (*Dasypoda*) *morawitzi* RADCHENKO is used as an outgroup.

Figure 2. Female of *Dasypoda radchenkoi* sp. nov. **a.** Dorsal view. **b.** Lateral view. **c.** Antenna. **d.** Head in frontal view. **e.** Vertex and ocellar field. **f.** Left malar space. **g.** Galea and maxillary palps in lateral view. **h.** Sculpture of the scutum. **i.** Sculpture of the propodeum. **j.** Pygidial plate. **k.** Metasoma in ventral view. **l.** Metasoma in dorsal view.

other tarsomeres orange. Claws orange basally and dark brown apically, with strong internal tooth.

Metasoma (figures 2j–l). T1 basally covered with semi-erect white hairs, distance between points of cuticle equal to 1–2 puncture diameter, underlying surface finely shagreened. T2–3 discs covered with erect and semi-erect brown pilosity, marginal part with white pilosity, basally with brown pilosity centrally and white pilosity laterally, underlying surface clearly shagreened between points. T4 like T2–3 yet apical white band complete (although intermixed with brown hairs centrally). T5 disc covered with dense tuft of dark brown hairs basally, followed by dense tuft of white hairs medially, and an apical, very dense tuft of light brown hairs. T5 disc apically strongly punctured (figure 2l). Pygidial plate strongly depressed and relatively deeply excised apically, shagreened, dark red, hairless but surrounded by dark brown pilosity (figure 2j). Surface of all sterna strongly shagreened, discs covered with very short, whitish-orangish adpressed pilosity, apically with dense bands of brown-orange hairs with exception of S6 entirely covered with longer dark brown hairs (figure 2k).

Figure 3. Male of *Dasypoda radchenkoi* sp. nov. **a.** Dorsal view. **b.** Lateral view. **c.** Antenna. **d.** Head in oblique view. **e.** Vertex and ocellar field. **f.** Left malar space. **g.** Galea and maxillary palp in lateral view. **h.** Metasoma in ventral view. **i.** Metasoma in dorsal view. **j.** Sternite 7 in ventral view. **k.** Sternite 8 in ventral view. **l.** Genitalia in lateral view with a focus on the lobes of the gonostylus.

Male

Head (figures 3c–f). Clypeus, supraclypeal and paraocular areas covered by dense, long and erect orange pilosity (in some darker paratypes clypeus posteriorly and supraclypeal area interspersed with black hairs), with thin band of black hairs along internal margin of compound eye (figure 3d). Clypeus surface typically obscured by dense pilosity, densely punctate, punctures separated by < 1 puncture diameter. Vertex covered with sparse, erect, black pilosity (posteriorly intermixed with orange hairs). Vertex laterally, between lateral ocellus and the compound eye, matt and densely shagreened (figure 3e). Gena (behind compound eye) shiny and densely punctate, covered with black pilosity (turning into orange laterally). Malar space short (figure 3f). Mandible strongly shagreened basally and apically shiny, hairless and reddish. Base of mandible externally with dark hairs (interspersed with orange hairs in lighter paratypes). Galea fully covered with small grains (figure 3g). Ventral part of scape covered with dense, dark pilosity. Pedicel and flagellum black. Flagellomeres bent medially, making them concave ventrally and convex dorsally, densely covered with very short, brownish pilosity (figure 3c). A3–4 subequal.

Mesosoma. Scutum, scutellum, metanotum and propodeum densely shagreened between punctures, covered with orange erect pilosity, scutum medially with darker pilosity, extending to almost cover area between tegulae in darker paratypes. Wing slightly infuscate. Femur, tibia and basitarsus with cuticle black but all tarsi orangish. Leg color varies from dark to orange depending on paratype. Inner surface of metabasitarsus covered with a mix of orange and black hairs. Spur of the meso- and metatibia orange, even in darker paratypes.

Metasoma (figures 3h–l). Cuticle of T1–4 densely shagreened basally and mostly matt. T5–6 less shagreened and shinier. Distance between punctures equals 2–3 point diameters. Apical parts of terga dark orange, sometimes very slightly translucid. Color of tergal pilosity dependent on the overall color of paratype, but T1–2 always with long orangish hairs medially. In very light paratypes, T1–4 more or less covered with orange pilosity (mostly replaced by black pilosity in darker paratypes). T5 with dark pilosity, except at apex with relatively dense fringe of short, lighter pilosity (orange in light paratypes). S1–5 (figures 3h) densely shagreened and punctured, covered with dark pilosity (black centrally and basally, and brownish apically), even in lighter paratypes. S6 apically with dense tufts of black hairs that radiate from the centers of apical processes. S7 as in figure 3j. Apical part of S8 truncated, as in figure 3k. Genitalia with internal dorsal lobe of gonostylus rounded, external lobe narrow and curved (figure 3l). Penis valve curved.

Material examined

Holotype. ♀, SPAIN: Málaga, Gaucín, Gaucín to Puerto del Hacho, 36.5214 N, 5.3328 W, 1.VI.2022, *leg.* T. J. WOOD (online barcode accession number IBIHM1330-22, figure 1).

Paratypes. 10 ♂♂, 1 ♀, SPAIN: Andalucía, Málaga, Parque Nacional Sierra de las Nieves, mount peak South of Pinsapo Escalereta, 36.6621 N. 5.0362 W, 30.V.2021; 7 specimens (including the female) *leg.* G. GHISBAIN and 4 specimens *leg.* T. J. WOOD. One of these male specimens was barcoded, with the online barcode accession number IBIHM432-21 (figure 1). 1 ♂, Andalucía, Yunquera Mirador Puerto Saucillo to Puerto del Cuco, 36.7163 N, 4.9768 W, 31.V.2021, *leg.* G. GHISBAIN.

Deposit location. The holotype and three paratypes are deposited in the Oberösterreichisches Landesmuseum, Linz, Austria. Two paratypes are deposited at the Naturalis Biodiversity Center, Leiden, the Netherlands. The other paratypes are deposited at the Laboratory of Zoology of the University of Mons, Belgium.

Diagnosis. Females of *D. radchenkoi* sp. nov. have dark hairs medially the scutum (they are completely bright orange in *D. pyrotrichia*, without any dark hairs). The pubescence of the mesobasitarsus includes dark hairs (with only white hairs in *D. albimana*). The outer surface of the galea is covered in small rounded grains and is therefore dull (shiny in *D. albimana* and *D. pyrotrichia*, covered in wave-like sculpture in *D. michezi*). The pubescence of the tibial scopae is entirely orange both dorsally and ventrally (figure 4b) (tibial scopae with some darker hairs in *D. albimana*, *D. michezi*, *D. morotei* and *D. pyrotrichia*, particularly dorsally) (see figure 4a for the tibial scopa of the closely related *D. morotei*). The only consistent structural difference found between *D. radchenkoi* sp. nov. and *D. morotei* is the sculpture of the basal part of the propodeum, which has clearly visible ridges in the new species (figure 4d) and without such ridges in *D. morotei* (figure 4c), the entire surface covered with fine granular shagreen. We also observed that both our female specimens of *D. radchenkoi* sp. nov. have a completely orange tibial scopa, which is much less common in *D. morotei* (a single specimen of the latter species at our disposal presented a light orange scopa, which might however be a discoloration due to the old age of the specimen).

Males of *Dasypoda radchenkoi* sp. nov. can be easily differentiated from *D. albimana*, *D. michezi* and *D. pyrotrichia* by the sculpture of the galea, as it is covered with small, rounded grains (the galea is smooth apically in *D. albimana* and *D. pyrotrichia* and covered with wave-like structures in *D. michezi*). The clypeus of *D. radchenkoi* sp. nov. has an impunctate median band (this feature is almost invisible when pubescence is undamaged), likewise in *D. morotei* and *D. pyrotrichia*. As in *D. morotei*, *D. radchenkoi* sp. nov. has flagellomeres 3–8 slightly curved instead of cylindrical in the other species of the subgenus. It has also a relatively short glossa as in *D. morotei*. Overall, we have not been able to find any obvious morphological or color character separating the males of *D. radchenkoi* sp. nov. from those of *D. morotei*. Although the propodeum might be diagnostic (with a higher probability of finding narrow raised ridges in the new species than in *D. morotei*), this difference is much more subtle than in the female sex, and it is unclear whether this difference will be consistent across a larger series of specimens. Based on the material at our disposal however, we observed that the T3 seems to always bear some ginger hairs on the disc in *D. morotei* (the area can be all black in *D. radchenkoi* sp. nov.). The T4 also seems to always bear some ginger hairs on the disc in *D. morotei* (while this area most often bears a black pilosity in *D. radchenkoi* sp. nov.). The apical hair of the galea appear shorter on

Figure 4. a. Scopa of the metatibia of a *D. morotei* female, with its typical brown and orangecoloration. **b.** Scopa of the metatibia of the female paratype of *D. radchenkoi* sp. nov., with its typical fully orange coloration. **c.** Sculpture of the propodeum of a female of *D. morotei*. **d.** Sculpture of the propodeum of the female holotype of *D. radchenkoi* sp. nov, with its diagnostic basal ridges (upper part of the image).

average in *D. morotei* than in *D. radchenkoi* sp. nov. *D. morotei* can have small depressions at the base of S8 (illustrated in RADCHENKO, 2017) while this part of the S8 seems straighter on average in *D. radchenkoi* (figure 3k).

Because these characteristics are mostly based on small series of non-barcoded, relatively old museum specimens of *D. morotei*, as well as a small type series of *D. radchenkoi* sp. nov., it is possible that future investigation reveals other more consistent discriminating features. As for the females, the best way to separate *D. morotei* from *D. radchenkoi* sp. nov. remains both the sampling locality and genetic barcoding.

Etymology. The species is named after Prof. Vladimir RADCHENKO, Head of the Department of General Biology of the National Academy of Sciences of Ukraine, a worldwide authority on the ecology and taxonomy of bees.

Flight period. May-June (based on the few data available).

Ecology. The species was observed in high-altitude limestone steppes in the Sierra de las Nieves in south-western Spain (figure 5). The collecting site was at an altitude of 1600 m and slightly above the tree line. The vegetation was dominated by shrubby *Vella spinosa* (Brassicaceae) and *Lavandula* spp. (Lamiaceae), with scattered *Abies pinsapo* (Pinaceae). Within this vegetation community were small shrubs of *Cistus albidus* (Cistaceae), the observed host plant. The second collecting site at the Puerto del Hacho (Gaucín) was at a lower altitude of around 800 m and was below the tree line in a habitat comprising *Quercus* woodland mixed with Mediterranean scrub. The female specimen was also captured on *Cistus albidus*. Given that other members of the *Heterodasypoda* are either oligolegs of *Cistus* (including the sister species *D. morotei*; MICHEZ *et al.*, 2008) or polylectic with a preference for Cistaceae, it is not possible to definitively comment on the dietary niche of *D. radchenkoi* sp. nov., although clearly *Cistus* plays an important role.

Remark. QUILIS (1928), in his revision of Spanish *Dasypoda*, described *D. morotei* from female specimens only (locus typicus San Rafael in the Sierra de Guadarrama) and also *D. nigra* from a single male specimen from 'de Teja'. The location of 'de Teja' is unclear, but the specimens were collected by the Catalan collectors BOFILL and CODINA who collected predominantly around Barcelona, and so this may refer to the village of Teià or Teyá (L. CASTRO, *pers. comm.*). It is also an unavailable name, since *D. nigra* is preoccupied by *Dasypoda pyrotrichia* var. *nigra* FRIESE, 1923 which was described from south-eastern Europe. Moreover, this name was synonymized with *D. pyrotrichia* by Warncke (1973) (see the illustration of the genital capsule provided by Quilis, 1928: plate V, image 1).

Conservation status

Based on the guidelines of the IUCN (IUCN Standards and Petitions Committee 2022, section 10.3, p. 79), we assessed *Dasypoda radchenkoi* sp. nov. as Data Deficient (DD) based on two main arguments: (i) there is no direct or indirect information about the status or possible threats about the taxon and (ii) apart from the few verified records from our three sampling localities, it is unclear whether records originally assigned to *D. morotei* in southern Spain could belong to *D. radchenkoi* sp. nov. (*e.g.*, see the map presented in Ghisbain *et al.*, 2021b). We therefore consider that the available data are inadequate for inferring a conservation status for the newly described species.

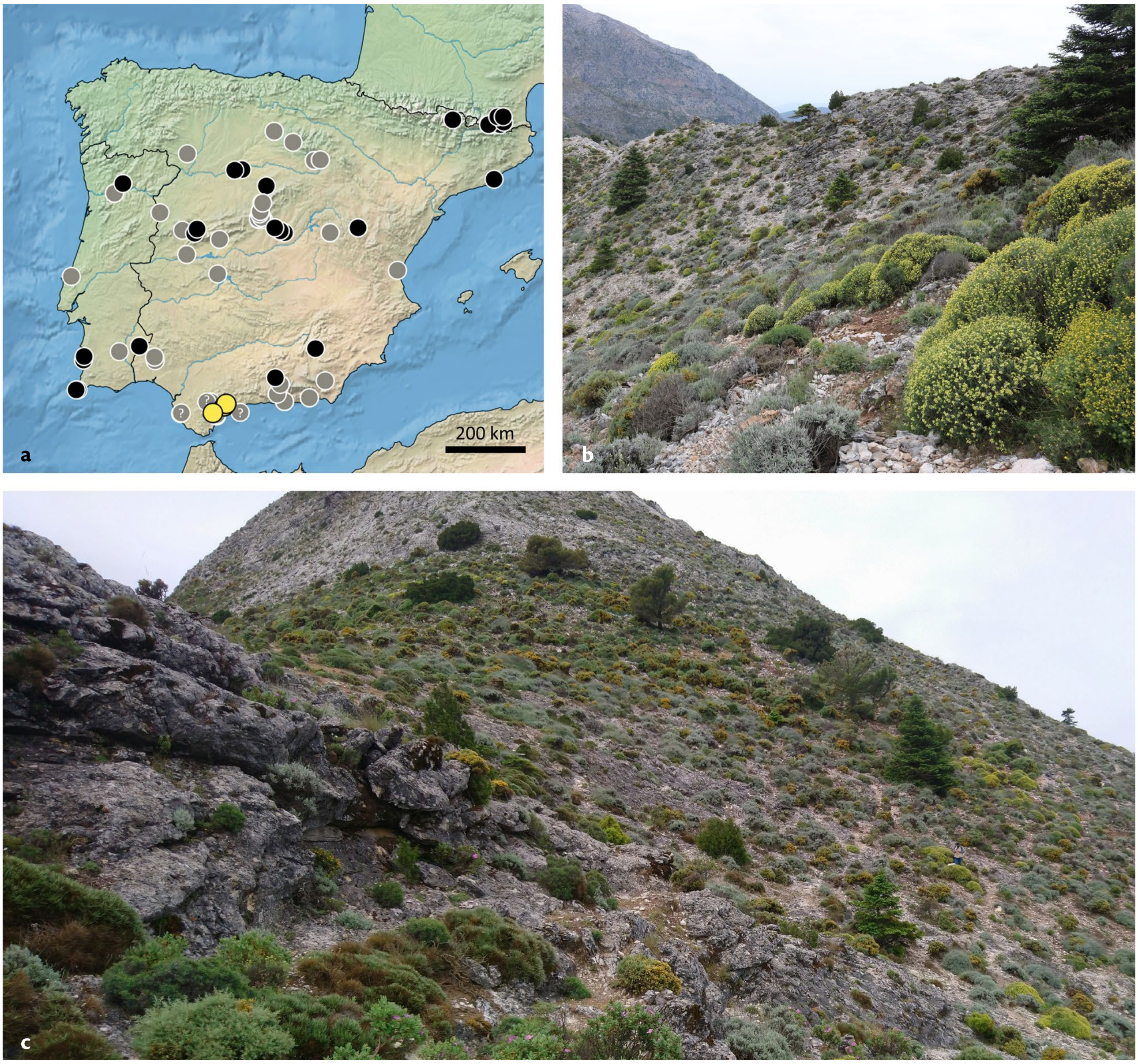

Figure 5. a. Map of the Iberian Peninsula, highlighting the known distribution of *Dasypoda radchenkoi* sp. nov. in yellow. Black dots represent localities where other *Heterodasypoda* have been recorded, and grey dots represent past records of *D. morotei* Quilis (based on Ghisbain *et al.*, 2021b). Revision of the southernmost records of the specimens attributed to the latter species are essential to assess their possible conspecificity with *D. radchenkoi* sp. nov. **b–c.** Sierra de las Nieves, mountain peak south of Pinsapo Escalereta (36.6610N, 5.0411W) where most specimens of *Dasypoda radchenkoi* sp. nov. from this study were collected (Spain, Andalusia, Málaga).

DISCUSSION

Taxonomy

We report the discovery of *Dasypoda* (*Heterodasypoda*) *radchenkoi* **sp. nov.**, a new pantaloon bee currently recorded from a few localities in south-western Spain. The species is part of the subgenus *Heterodasypoda* as described by Michez *et al.*, (2004b, with complements from Radchenko, 2017): males (i) have the apex of S6 with a dense, brown pubescence (figure 3h); (ii) the apex of the sternite 7 is slightly indented medially and with two lateral large structures (figure 3j); (iii) the base of sternite 8 does not present lateral hooks (figure 3k); (iv) the inner face of the apical lobe of the sternite 8 has a transverse carina (which is more or less carved); (v) they do not present a narrow tooth at the base of the gonostylus and (vi) have a scaly surface on the internal ventral lobe of the gonostylus.

Females also share most characters with other *Heterodasypoda*: (i) the maxillary palpae and galea of subequal length; (ii) the margin of the galea with setae along its length; (iii) the malar space shorter than the pedicel; (iv) the disc of T2 with a relatively straight marginal line; (v) the pygidial plate completely glabrous; and (vi) the nervulus prefurcal. However, females of *D. radchenkoi* sp. nov. contrast with most of the other *Heterodasypoda* by their completely orange scopa (although some of the orange hairs appear slightly darker on the metatibia).

Within *Heterodasypoda*, the phenotype of both males and females of *D. radchenkoi* sp. nov. is strongly reminiscent of the more widespread species *D. morotei*. The use of genetic tools, including the use of near topotypic samples of *D. morotei* from the Sistema Central in central Spain, was therefore pivotal for highlighting the species level differentiation of *D. radchenkoi* sp. nov. from *D. morotei*. In the case of *D. radchenkoi* sp. nov. and *D. morotei*, the challenge of separating species is exacerbated by the high color polymorphism that strongly overlaps among the males of both species. At the moment, the only sculptural character that seems consistent between the two species relates to the form of the female propodeum (see Diagnosis section and figure 4).

Ecology

The Iberian Peninsula has received a sustained interest in the last few years in terms of bee monitoring with a compiled data base of spatial records (Bartomeus *et al.*, 2022), and taxonomic advances with the description of many new wild bee species, including a new *Dasypoda* (Radchenko *et al.*, 2017) and even a new bee genus (Wood *et al.*, 2022). Based on these findings and new observations recently made by the authors as part of field trips in Spain and Portugal, there is no doubt that the peninsula still has surprises in store. The case of *D. radchenkoi* sp. nov. participates to demonstrate that continued efforts are strongly needed to comprehensively monitor the highly diversified wild bee fauna of the Iberian Peninsula (Ortiz-Sánchez, 2011, 2020) and also to systematically barcode collected specimens in order to uncover overlooked cryptic taxa.

In addition to the need to study in more detail the habitats in which our new species occurs, more effort is also required to characterize its phenology. Data from other *Heterodasypoda* bees show that their flight period occurs from late spring to late summer: May to July for *D. albimana*, April to May for *D. michezi* (based on very few occurrence records), and predominantly May to August for *D. morotei* and *D. pyrotrichia* (Ghisbain *et al.*, 2021b; Grace 2010; Michez *et al.*, 2003; Ornosa & Ortiz-Sánchez, 1998; Radchenko *et al.*, 2019). The few records presented here suggest that *D. radchenkoi* sp. nov. flies at least in May and June, although they are based on a single field trip. Moreover, the strong association of all our specimens with blooming *Cistus* plants is consistent with other *Heterodasypoda* bees (Michez *et al.*, 2008), suggesting a possible specialization on this plant genus. Overall, given the increasing evidence of substantial phenological shifts and plant-pollinator mismatches following ongoing eco-climatic changes (Duchenne *et al.*, 2020; Gérard *et al.*, 2020), delimiting in more detail the phenology and food specialization of bee species may be key to their effective monitoring and subsequent conservation.

Endemism and micro-endemism

It is important to note the ecological differences between the two loci typici of *D. radchenkoi* sp. nov. and *D. morotei*, namely the Serranía de Ronda/Sierra de las Nieves and the Sistema Central, to understand why two different Iberian endemic species have arisen. The collecting localities of DNA barcoded *D. morotei* in this study came from habitats on acidic soil, and specimens were collected from *Cistus laurifolius* in the Sistema Central and Sistema Ibérico. In contrast, *D. radchenkoi* sp. nov. was collected from basic soils with *Cistus albidus*. An important feature of major Iberian Mountain ranges (e.g. Pyrenees, Cantabrians, Sistema Central, and the Baetic system) is that they are largely isolated from one another, typically running more or less east-west rather than north-south. This may have resulted in species becoming isolated as they moved north and south during glacial-interglacial oscillations, potentially contributing to patterns of diversity as seen in bee groups such as *Andrena* (*Taeniandrena*) with related species displaying stratified distributions across the peninsula (Wood *et al.*, 2021). The 9.6% DNA barcode difference between *D. radchenkoi* sp. nov. and *D. morotei* implies that their divergence was not recent, even though very little morphological differences appear to have accumulated during this time. Indeed, if they possess the same dietary niche using *Cistus* pollen, then there would have been little selective pressure leading to changes in their morphologies. This observed microendemism may therefore be reflective of interrupted gene flow rather than the specific colonization of a different ecological niche. In this context, specimens of '*D. morotei*' reported from southern Spain (Ghisbain *et al.*, 2021b; Michez *et al.*, 2004a) must be reexamined morphologically and genetically in order to determine whether the two cryptic taxa occur in sympatry.

Perspectives for conservation

Future revisions unveiling the spatial distribution, ecology and evolutionary history of similar endemic species are encouraged to properly assess any potential threat that these pollinators could face in our changing world. More specimen data for *D. radchenkoi* sp. nov. are urgently needed to properly delineate its total distributional range and enable a conservation assessment under the IUCN criteria.

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