



Endophallus structure: a promising tool for cryptic species identification in *Timarcha* Samouelle, 1819 (Coleoptera: Chrysomelidae: Chrysomelinae)

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Abstract

Contrary to the subtle differences of habitus found between many species of *Timarcha* their internal sacs of male genitalia have shown a remarkable variation. Thirty-two Palaeartic taxa, mostly from the Iberian Peninsula, have been analyzed for this trait, which can be used for species diagnosis and also for establishing species groups of close relatedness in agreement mainly with genetic analyses. According with this trait, new synonymies and taxonomical changes are proposed: *T. intermedia carmelenae* Petitpierre, 2013 **stat. nov.**, *T. intermedia kiesenwetteri* Kraatz, 1879 **stat. nov.**, *T. intermedia lugens* Rosenhauer, 1856 **stat. nov.**; *T. sinuatocollis monserratisensis* Bechyné, 1962 **comb. nov.**; *T. piochardi* Fairmaire, 1874 **stat. nov.**; *T. tortosensis* Bechyné, 1948 **stat. nov.**; *T. perezii* Fairmaire, 1884 **syn. nov.** and *T. asturiensis* Kraatz, 1879 **syn. nov.** = *T. geniculata* Germar, 1824. Furthermore, the endophalli of *T. hummeli*, *T. carmelenae*, *T. kiesenwetteri*, *T. lugens*, *T. tenebricosa*, *T. parvicollis*, *T. insparsa*, *T. marginicollis*, *T. balearica*, *T. strangulata* spp., *T. calceata*, *T. scabripennis*, *T. espanoli*, *T. monticola*, *T. cyanescens*, *T. interstitialis*, *T. aurichalcea*, *T. oblongula*, *T. hispanica* and *T. granadensis* are illustrated. One new species, *T. aitanae* **sp. nov.** is described.

Key words: *Timarcha*, endophallus, male genitalia, taxonomy, species groupings

Introduction

The genus *Timarcha* Samouelle, 1819 is considered to be amongst the most difficult of the 40 Palaeartic genera of the subfamily Chrysomelinae (Kippenberg 2010). With a total of 317 described taxa (Gómez-Zurita & Kippenberg 2010; Petitpierre & Daccordi 2013) and including at least two Western North America species (Jolivet *et al.* 2014), the number of well-defined species is subject to a high degree of uncertainty. As a good example of this, Gómez-Zurita (2008) stated that only 80 species and 48 subspecies are of stable taxa, whereas other 196 are unstable due to synonymies, status changes, or alternative placements. Warchałowski (2003) asserts that roughly half of the described species should probably fall in synonymy. Later on, Warchałowski (2010) wrote that at least 80% of about 240 described species and subspecies should be downgraded to infrasubspecific level, and very recently, Jolivet *et al.* (2014) reported a tentative number of roughly 100 species and 50 subspecies for the genus. On the other hand, most authors agree in considering only three subgenera, *Americanotimarcha* Jolivet, 1948 restricted to North America, *Metallotimarcha* Motschulsky, 1860 mostly from mountains in Western and Central Europe and eastern ranges from Anatolia, and *Timarcha s. str.* Samouelle, 1819 from Western Palaeartic (Daccordi 1994; Gómez-Zurita 2004; Winkelman & Debreuil 2008; Kippenberg 2010; Tiberghien 2014). More than 95% of the described taxa belong to the last mentioned subgenus. Nearly one third of the species are found in the Iberian Peninsula and a great majority of them are endemics, making their study particularly relevant. Contrary to any other Palaeartic genus of the subfamily Chrysomelinae, the species of *Timarcha* cannot be identified by the shape of the median lobes of their male genitalia, being very similar and useless out of two rare exceptions: southern Iberian *T. marginicollis* Rosenhauer, 1856 and the Balearic endemism *T. balearica* Gory, 1844 (Petitpierre 1970a). The apodemes (flagellum + manubrium) of endophalli of *Timarcha* have been studied in few species which have shown an unsuspected variation (Stockmann 1967; Petitpierre 1970a, 1973). However, the complete endophallus, that includes both soft parts and apodemes, had only been illustrated by Stockmann (1967) in three species: *T. goettingensis* (Linnaeus, 1758), *T. maritima* Perris, 1855 and *T. tenebricosa* (Fabricius, 1775).

The “lock and key” hypothesis assumes that in insects the male and female genitalia should be exactly fitting during copula, so genitalia are very important traits for ensuring sexual reproduction and also for preventing the reproductive isolation in case of possible matings between different species (Shapiro & Porter 1989). Nevertheless, this hypothesis should not be literally taken as it has been shown for ground beetles (Takami 2002, 2007).

A recently published technique for studying endophalli in leaf beetles has been successfully applied to separate species of the chrysomeline genus *Oreina* Chevrolat, 1836 (Bontems 2013). Moreover, some years before, Sassi (2001a, 2001b) used the internal sac sclerites to differentiate among three allied species of the *Cryptocephalus aureolus* group (Cryptocephalinae) occurring in Northern Italy and among five of the *C. marginellus* group in Southern Europa, respectively. Herein, we have used this technique to investigate its potential as a diagnostic character to discriminate species of *Timarcha* mostly from the Iberian Peninsula.

Our results on the endophalli of *Timarcha* will be discussed taking into account the proposed 28 morphological groups of species by Bechyné (1948a) and by Warchałowski (2010), their host-plant affiliations (Jolivet & Petitpierre 1973), karyotypes (Petitpierre 1970b, 1976, 2016; Gómez-Zurita *et al.* 2004), mitochondrial DNA phylogeny (Gómez-Zurita *et al.* 2000a, 2000b), or an ITS-2 nuclear DNA phylogeny (Gómez-Zurita *et al.* 2000c), and mainly on a molecular systematics using a combined DNA phylogeny based on two mitochondrial (16S rDNA and CO2) and two nuclear (ITS-2 and 18S rDNA) gene markers (Gómez-Zurita & Vogler 2003; Gómez-Zurita 2004). This is only a first study on the endophalli of *Timarcha*, further and more extensive analyses will be reported later.

Materials and methods

The studied taxa, number of specimens and their geographic origins, dates and collectors, are given in the results with the checked species. They are deposited in the collections of Daugavpils University Beetle Collection (DUBC) in Latvia, Alexandr Anichtchenko Collection (AAC) and Eduard Petitpierre Collection (EPC), both in Spain (Madrid and Palma de Mallorca, respectively). Types are deposited at the Museo Nacional de Ciencias Naturales (MNCN) in Madrid, and others as indicated.

The method for preparing and using the aedeagal endophallus was clearly detailed by Janovska *et al.* (2013). The internal sacs were photographed in their dorsal (D), and/or ventral (V), and lateral (La) views. In addition, the protuberances are named according with their locations: apical (A) or basal (B), left (L) or right (R). Thus, namely DB = dorso-basal, DAL = dorso-apical or DAR = dorso-apical left or right, respectively, VA = ventro-apical, VB = ventro-basal, VAL or VAR = ventro-apical left or right, respectively, and VBL or VBR = ventro-basal left or right, respectively. Most of these different parts of an endophallus of *Timarcha* are depicted in a schematic diagram (see fig. 1).

Results

The endophalli of the aedeagus of *Timarcha* display remarkable diversity contrasting with the high uniformity shown by the median lobes of most species. Although our sampled taxa and populations are still meager, we can distinguish groups of species based on their types of internal sacs. It is noticeable that the species of the so-called *T. goettingensis* complex, rather well-characterized from their morphological and chromosomal features (Petitpierre 1970b, 1976; Gómez-Zurita *et al.* 2004), and later confirmed by mitochondrial and nuclear DNA analyses (Gómez-Zurita *et al.* 2000a, 2000b, 2000c; Gómez-Zurita & Vogler 2003; Gómez-Zurita 2004), do not show homogeneous endophalli.

Subgenus *Metallochimarcha* Motschulsky, 1860

Type species *T. metallica* (Laicharting, 1781)

Description: Sexual dimorphism feebly pronounced in body size and tarsal thickness. Upper side with bronzy or cupreous sheen. Pronotum without marginal ridges. Mesoventrite broad and flat. Aedeagus with a long apical part of tegmen. Four European species, one of them reaching Caucasus Mts. and Armenia. One single species studied:

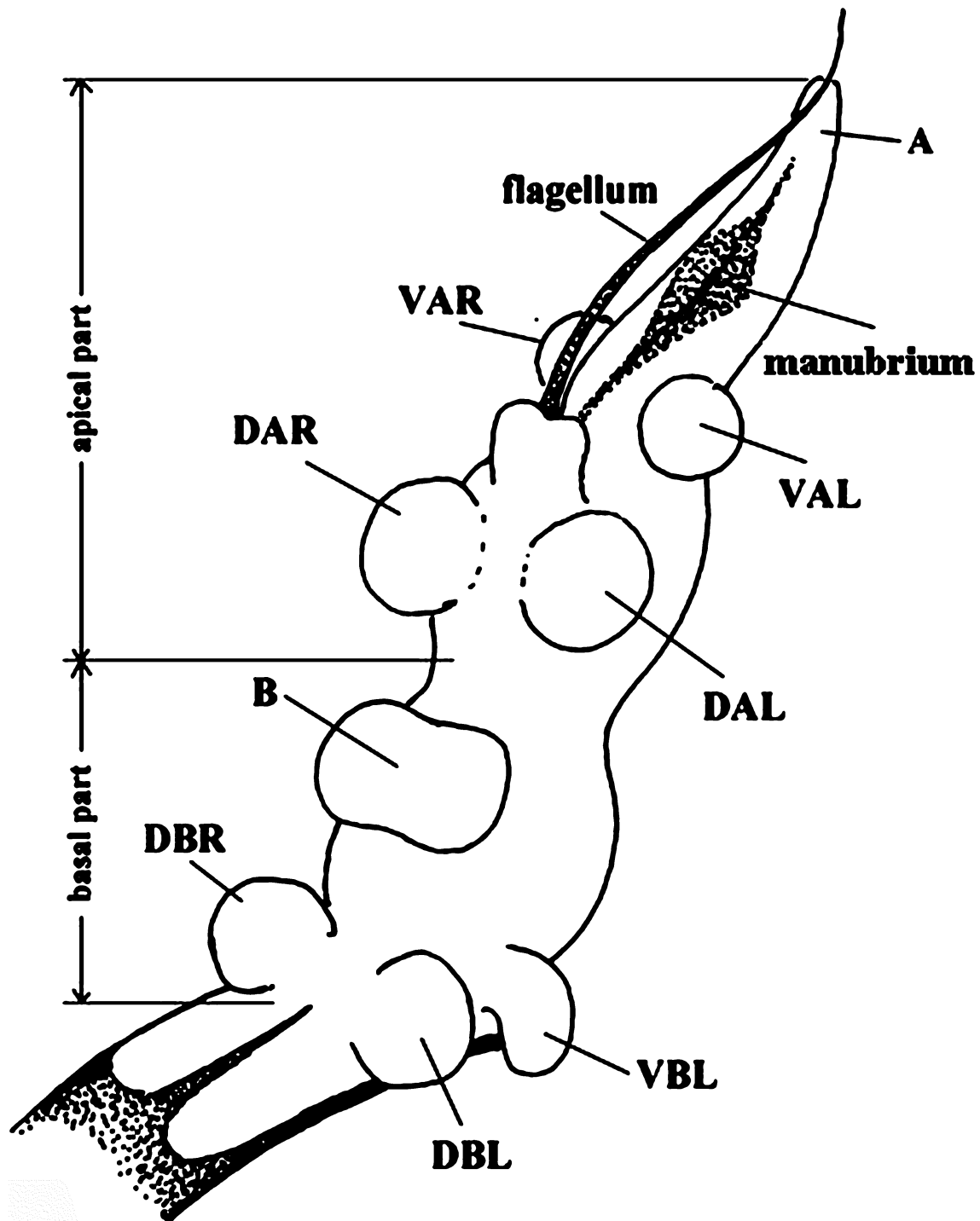


FIGURE 1. Schematic diagram of a *Timarcha* endophallus: DA = dorso-apical, DB = dorso-basal, VA = ventro-apical, VB = ventro-basal, VAL or VAR = ventro-apical left or right and VBL or VBR = ventro-basal left or right, respectively.

***T. (Metallo)timarcha hummeli* Faldermann, 1837**

Material: 2 exs.—Russia, Krasnodar reg. 6 km NE Soloniki village, Sochi National Park, 28–30.05.2009, leg. A. Anichtchenko (AAC).

Description: Endophallus has a simple globular shape (figs. 2a, b). DA large; DV short and thin; VBL and VBR small; the other protuberances indistinct.

Subgenus *Timarcha* s. str.

Type species: *T. laevigata* (Linnaeus, 1767)

Description: Sexual dimorphism clearly pronounced in body size and tarsal structure, thicker in males. Upper side usually black, less often bronzy, and rarely bluish, dark violet or green. Pronotum with marginal ridges except in a few species. Mesoventrite narrow and bulgy, more or less protuberant. Aedeagus with a short or very short apical part of tegmen. This subgenus includes also the currently invalid subgenus *Timarchostoma* Motschulsky, 1860, described on the basis of a longer marginal ridge of elytra and mid-tibia of males longer than corresponding tarsi, which cannot be substantiated neither on other morphological characters nor on the results from DNA-based phylogenies (Gómez-Zurita 2004).

“*T. tenebricosa*”—group

Description: Large species, usually > 14 mm. Pronotum rather flat, transverse, laterally rounded and feebly sloping, often heart-shaped. Mesoventrite slightly raised. Elytra very finely and densely punctated. Warchalowski (2010) quotes five species in this group. One single species studied:

***T. (T.) tenebricosa* (Fabricius, 1775)**

Material: 1 ♂: Spain, Cerler (Pyrenees of Huesca), 07. 1970, leg. X. Palaus (EPC); 1 ♂:—Spain, Port de la Bonaigua, 2000m alt., (Pyrenees of Lleida), 25.06.1970, leg. E. Petitpierre (EPC); 2 ♂: Caucasus Mts., Russia, Karachaevo—Cherkessia, Skalisty Mt. Range, Banditskoe urochische, 2–3.05.2009, leg. D. Fominhy (AAC); 1 ♂: Russia, Karachay—Cherkessia, valley of Daut river, 43°51'N 42°04'E, 20.05.2009 leg. A. Anichtchenko (ACC); 1 ♂, 1 f ♀: Ukraine, Krimea, Bahchisarai distr., 5 km ESE Sokolnoe village, Sotira Mt., 1100m, 44°32'46"N 34°01'13"E, 3.05.2015, leg. K. Makarov (AAC).

Description: Endophallus globular in shape, with VBL and VBR present; VA conical with apex slightly turned dorsally; manubrium widely divided showing two lateral parts connected by a membrane, flagellum long and well sclerotized. Shape of endophallus almost identical in the whole range of the species distribution from the Caucasus to northern Spain, and similar, but not identical to that published by Stockmann (1967) from French material. Minor differences are due to the fact that sac from the Pyrenees specimens are less inflated (figs 3a, b, c, d).

“*T. intermedia*”—group

Description: Body stout of medium or large size, 12.0—16.0 mm. Pronotum uniformly punctate, with posterior angles strongly or feebly sloping. Proventrite slightly grooved lengthwise. Mesoventrite forked. Elytra dull, sparsely and finely punctate, with or without weak costae in females.

***T. (T.) intermedia* Herrich-Schäffer, 1831**

This species is characterized by the pronotum slightly narrowed before the basal angles but with marginal ridge entire, mesosternum strongly forked, and elytra dull black with scattered shallow punctation. Body rather large and broad, 12–16 mm. While most *Timarcha* s. str. species feed on Rubiaceae and/or Plantaginaceae, the food-plants of the species in this group are always Brassicaceae (Jolivet & Petitpierre 1973; Petitpierre & Daccordi 2013). Its chorology covers Southeastern Iberia, *T. intermedia* s. str. is generally found at low altitudes in Alicante, Murcia and Almería provinces, whereas *T. intermedia carmelena* Petitpierre, 2013 **stat. nov.**, *T. intermedia kiesenwetteri* Kraatz, 1879 **stat. nov.**, and *T. intermedia lugens* Rosenhauer, 1856 **stat. nov.**, inhabit some of the isolated high

mountains in the Granada or Jaén provinces, generally over 1500 m altitude. Endophalli showing a globular apical part with a horn-like apical protuberance A, basal part fine and cylindrical with small B and almost indistinct VB and DB protuberances (figs 4–7). The four subspecies display the same type of internal sac, therefore we propose the previously quoted **stat. nov.**

T. (T.) intermedia s. str.

ehlersi Weise, 1882b: 319

sericea Fairmaire et Allard, 1873: 153

Material: 2 ♂, 1 ♀: Cabo de Gata, San José, Almería, 10.02.2004, leg. A. Anichtchenko (AAC); 2 ♂: Cabo de Gata, 6 km N San José, Almería, 12.01.2004, leg. A. Anichtchenko (AAC); 2 ♂: Faro de Santa Pola, Alicante, 06.1963 leg. E. Petitpierre (EPC); 4 ♂: isla de Tabarca, Santa Pola, Alicante, 27.12.1973, leg. E. Petitpierre (EPC).

Description: The endophalli from these three different locations are illustrated (figs 4a–f). As noticed, the basal protuberances are more or less prominent but the main features are maintained. It differs from the mountainous subspecies, *T. intermedia carmelena*, *T. intermedia kiesenwetteri* and *T. intermedia lugens*, by a less sclerotized flagellum.

***T. (T.) intermedia carmelena* Petitpierre, 2013 stat. nov.**

Material: 1 ♂: Puerto de la Sagra 1670m. alt., Sierra Guillimona, Huéscar, Granada, 12.10.2012, leg. G. Bastazo and J.M. Vela ; 2 ♂: *ibid*, 01.05.2014, leg. G. Bastazo and J.M. Vela (AAC, EPC).

Description: The shape of the endophallus (figs 5a, b) does not differ from that of *T. (T.) intermedia kiesenwetteri* Kraatz, 1879. Manubrium well sclerotized.

***T. (T.) intermedia kiesenwetteri* Kraatz, 1879 stat. nov.**

***T. sagrensis* Kuntzen, 1911**

Material: 1 ♂: Sierra Mágina, near Cortijo de los Prados 1500m. alt., Jaén, 25–28.03.2010, leg. A. Anichtchenko (AAC); 5 ♂: Sierra Mágina, 1800m. alt., Jaén, 25–28.03.2010, leg. A. Anichtchenko (AAC).

Description: Endophallus (figs 6a, b) identical to that of *T. (T.) intermedia carmelena*. Manubrium well sclerotized.

***T. (T.) intermedia lugens* Rosenhauer, 1856 stat. nov.**

Material: 1 ♂: Sierra Nevada: laguna de Aguas Verdes, 3056 m alt., Granada, 11.06.2005, leg. A. Anichtchenko (AAC); 3 ♂: Sierra Nevada: Pico Veleta, 2300 m alt., Granada, 05.07.1972, leg. E. Petitpierre (EPC).

Description: The females of this species differ from those of the remaining species of this group by having slight longitudinal costae on their elytra. Here, again, its endophallus (fig. 7) agrees with those of the former taxa

“*T. parvicollis*”—group

Description: Body with a relatively slender shape of small-medium size, 8.0–13.5 mm. Pronotum strongly narrowed at basal angles with marginal ridges erased. Proventrite deeply grooved longitudinally. Mesoventricle forked. Elytra with shallow scattered or rather dense punctuation. Bechyné (1948a, 1953) considered only one species with five subspecies from Andalusia and Portugal, in Southern Iberia: *T. parvicollis parvicollis* Rosenhauer, 1856, *T. parvicollis seidlitzii* Kraatz, 1879, *T. parvicollis coarcticollis* Fairmaire, 1873, *T. parvicollis heydeni* Weise, 1882 and *T. parvicollis paulinoi* Kraatz, 1879.

***T. (T.) parvicollis parvicollis* Rosenhauer, 1856**

Material: 1 ♂: Vélez de Benaudalla, Granada, 07.12.2003, 07.12.2003, leg. A. Anichtchenko (AAC); 1 ♂: Sierra Nevada. Rio Lanjarón 1200 m alt., Granada, 29–31. 03.2010, leg. A. Anichtchenko (AAC); 1 ♂: Sierra Mágina, 1800m alt., Jaén, 25–28.03.2010, leg. A. Anichtchenko (AAC); 1 ♂: Sierra de las Nieves, 1730—1800 m alt., Málaga, leg. A. Anichtchenko (EPC).

Description: Pronotum finely and sparsely punctate. Elytra also finely but rather densely punctate.

***T. (T.) parvicollis seidlitzii* Kraatz, 1879**

Material: Three studied specimens: 2 ♂: Sierra Nevada: Pico Veleta, 2500—2700 m alt., Granada, 08.10.2002, leg. A. Anichtchenko (AAC); 1 ♂: Sierra de Baza: prados del Rey, 2300 m alt., Granada, 04.02.2003, leg. A. Anichtchenko (EPC).

Description: Pronotum almost smooth. Elytra very finely and sparsely punctate. *T. (T.) parvicollis parvicollis* and *T. (T.) parvicollis seidlitzii* share an identical endophallus (figs. 8) without manubrium, the basal part is cylindrical, the VB and DB protuberances are indistinct, and the apical part is voluminous with only a VA horn-like protuberance.

“*T. insparsa*”—group

Description: Species vary in size from 11 to 15.5 mm, with moderate or feeble upper shining. Mesoventrite forked. Elytra loosely joined, and covered with scattered feeble punctuation. Hind tarsi of females entirely naked, with a median longitudinal groove. According to Bechyné (1948a), this group only has two species *T. (T.) marginicollis* Rosenhauer, 1856 and *T. (T.) insparsa* Rosenhauer, 1856, clearly distinct in size and shape, but Warchalowski (2010) claims that it is composed of two further species, *T. (T.) heydeni* Weise, 1882 and *T. (T.) seidlitzii* Kraatz, 1879.

***T. (T.) insparsa* Rosenhauer, 1856**

Material: 2 ♂: Sierra Nevada: laguna de Aguas Verdes, 3100 m alt., Granada, 11.06.2005, leg. A. Anichtchenko (AAC); 1 ♂: Sierra Nevada: laguna de las Yeguas, 2850 m alt., Granada, 03.09.2004, leg. A. Anichtchenko (AAC); 1 ♂: Sierra Nevada: Borreguiles, Granada, 03.08.1998, leg. A. Anichtchenko (AAC).

Description: Body of smaller size compared to *T. marginicollis* and showing convex interstices of elytra. In addition, the apex of the penis is similar to that shown by most congeneric species and does not display the two large lateral teeth of *T. marginicollis*. The endophallus of *T. insparsa* (figs 9a, b) shares with that of *T. marginicollis* a divided arrowhead manubrium but differs strongly by its shape, which is closely related to those of *T. intermedia* and *T. goettingensis* groups, because it shares the slender and straight basal part and the highly inflated apical one.

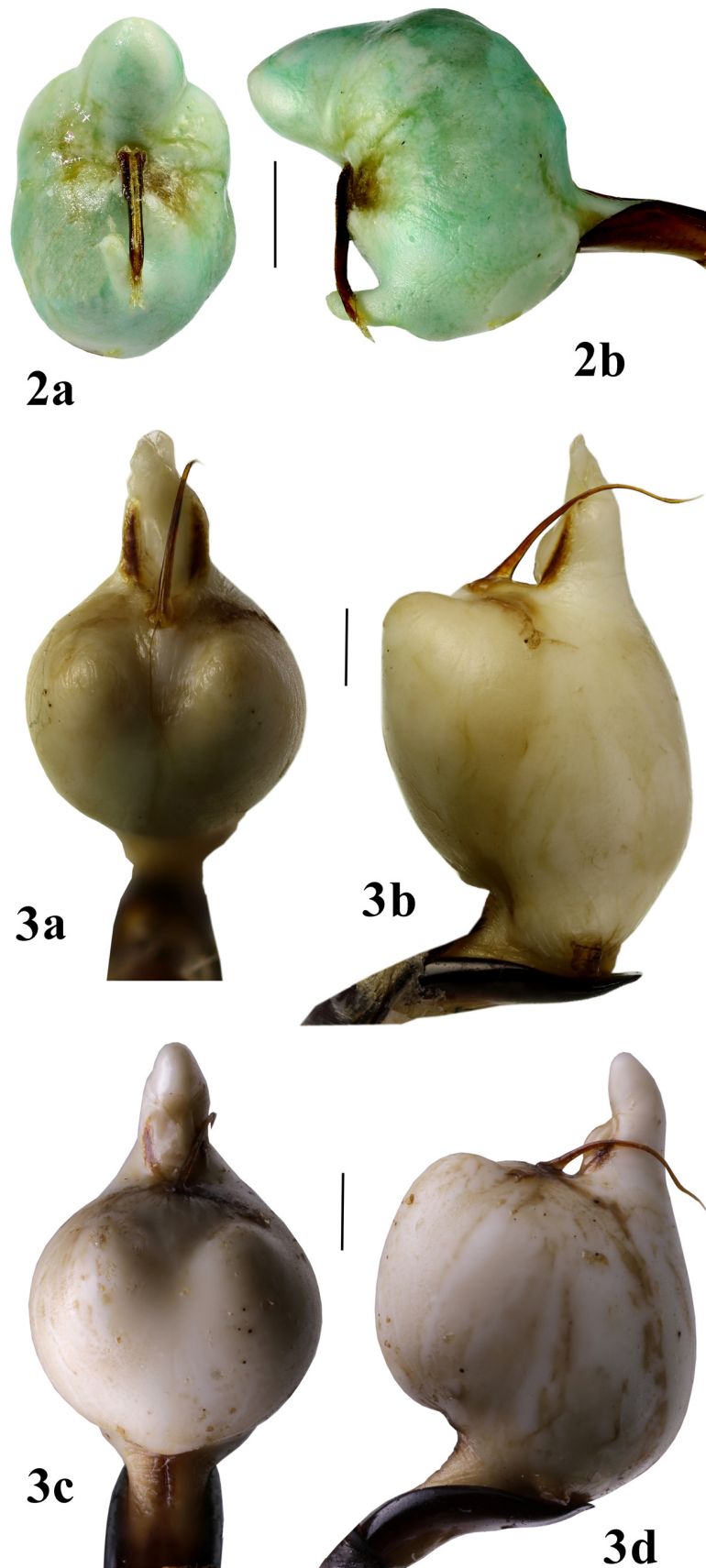
***T. (T.) marginicollis* Rosenhauer, 1856**

T. escalerae Kuntzen, 1911: 216

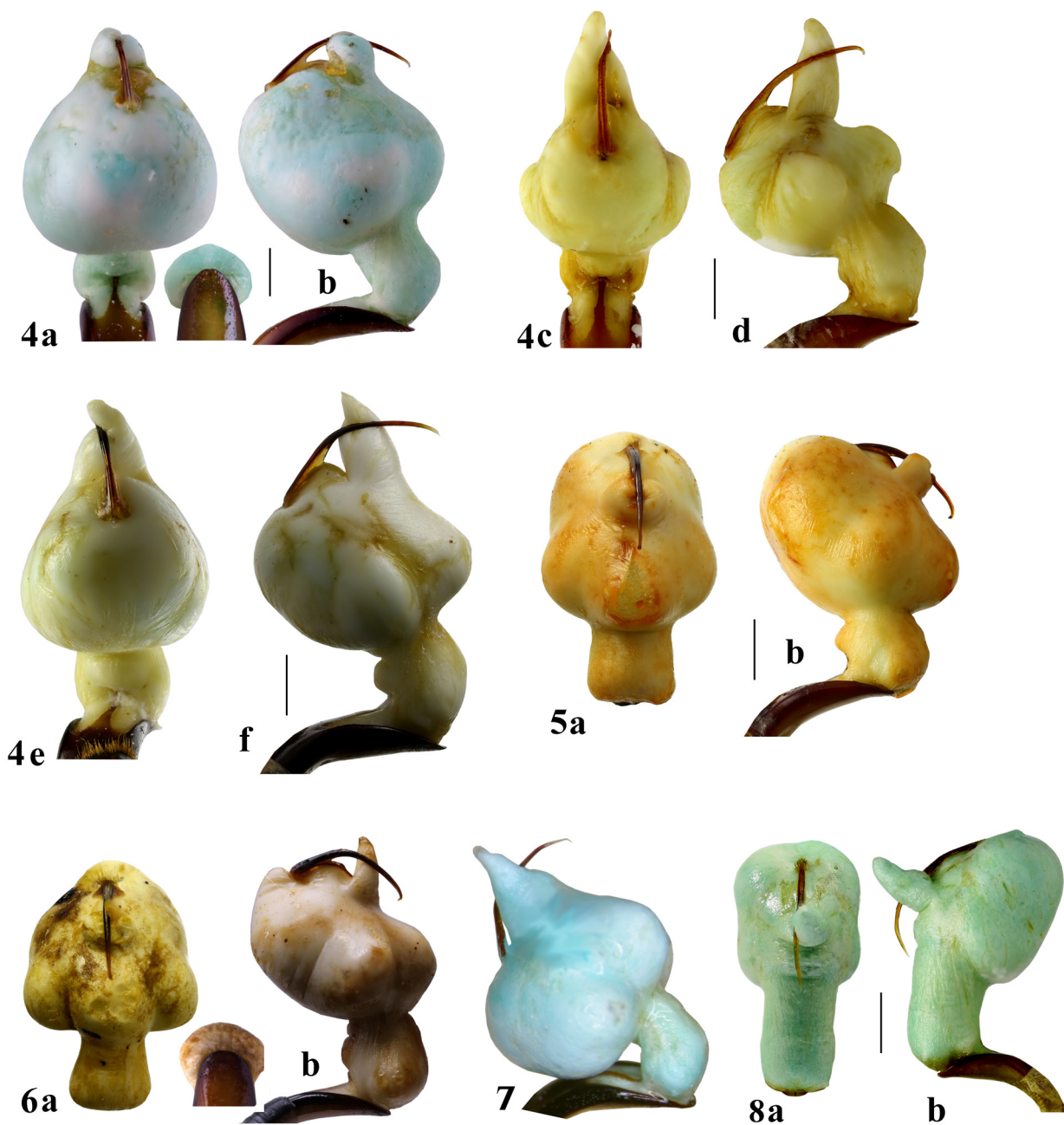
T. nevadensis Fairmaire, 1884a: 81

T. splendida Pérez Arcas, 1872: 121

Material: 2 ♂: Sierra Nevada: rio Monachil, 1600m alt., Granada, 29.04.2003, leg. A. Anichtchenko (AAC); 2 ♂: Puerto de la Ragua, 2000m alt., Granada—Almería, 26.05.2004, leg. A. Anichtchenko (AAC).

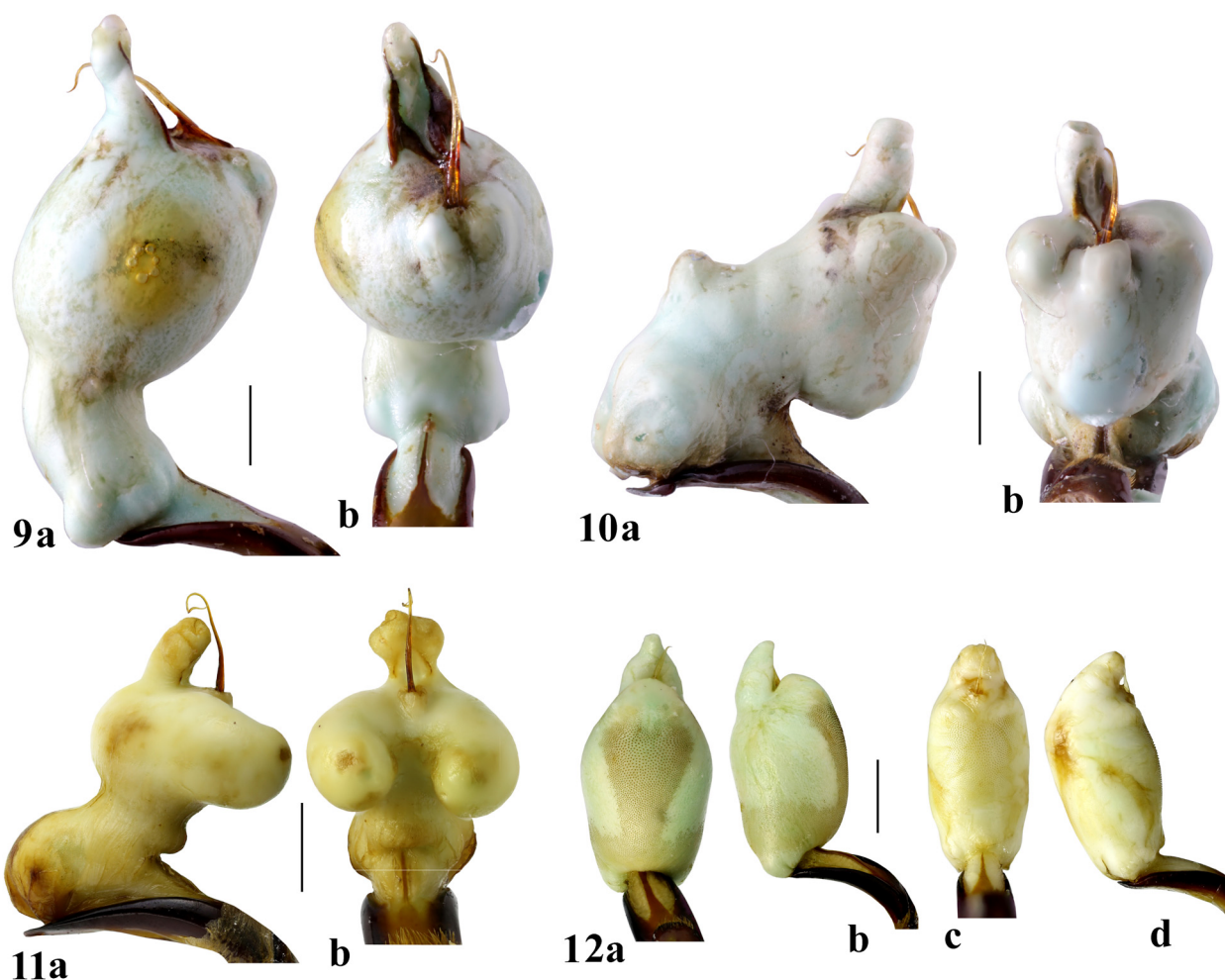


FIGURES 2–3. Endophallus structure of *Metallo timarcha* and “*T. tenebricosa*” groups: 2a, b—*T. (Metallo timarcha) hummeli*; 3a, b—*T. (T. tenebricosa)* (Pyrenees), 3c, d—*T. (T. tenebricosa)* (Caucas). Scale = 1 mm.



FIGURES 4–8. Endophallus structure of “*T. intermedia*” and “*T. parvicollis*” groups: 4a, b—*T. intermedia* s. str. (Cabo de Gata), 4c, d—*T. intermedia* s. str. (Isla de Tabarca), 4e, f—*T. intermedia* s. str. (Faro de Santa Pola); 5a, b—*T. intermedia carmelenae* (Puerto de la Sagra); 6a, b—*T. intermedia kiesenwetteri* (Sierra Mágina); 7—*T. intermedia lugens* (Sierra Nevada). 8a, b—*T. parvicollis seidlitzii* (Sierra Nevada). Scale = 1 mm.

Description: Body large (12.0–15.5 mm.) with heart-like pronotum and elongate elytra with scattered shallow punctation on flat interstices. Median lobe of aedeagus with bi-toothed apex (Petitpierre 1970a), a feature which has not been found in any other species of *Timarcha*. The endophallus shows a divided manubrium of arrowhead shape (Petitpierre 1970a; Petitpierre & Daccordi 2013), and two large VB and VA protuberances whereas the DB and DA are indistinct (figs 10a, b).



FIGURES 9–12. Endophallus structure of “*T. insparsa*”, “*T. balearica*” and “*T. strangulata*” groups: 9a, b—*T. insparsa* (Sierra Nevada); 10a, b—*T. marginicollis* (Sierra Nevada). 11a, b—*T. balearica* (Menorca); 12a, b—*T. strangulata epipleuralis* (Canfranc), 12c, d—*T. strangulata sculptipennis* (Port de la Bonaigua). Scale = 1 mm.

“*T. balearica*”—group

Description: group consisting of only one species, *T. balearica*, roughly similar in size and shape to *T. intermedia* but a bit less stout, longer legs, and differing also in the complete absence of punctation on the upper side. It displays chromatic polymorphism, although most specimens are dull dark green, sometimes even a slightly bright black colour contrary to the latter species. It is endemic of the two larger Balearic islands, Mallorca and Menorca.

T. (T.) balearica Gory, 1833

T. martini Pic, 1919a: 20

T. violacea Pic, 2019a: 20

Material: 3 ♂: Fornells, Menorca, Balearic Islands, 09.10.1973, leg. E. Petitpierre (EPC).

Description: Endophallus (figs 11a, b) complex with soft parts with a round apical horn, two prominent DA protuberances, and a divided and weakly sclerotized small manubrium, joined to a tip coiled flagellum (Petitpierre 1970a). In addition, the apex of penis is truncated instead of pointed, as it is common for most species (Petitpierre 1970a).

“*T. strangulata*”—group

Description: this is a monospecific group that only includes *T. strangulata*, a species with dorsally depressed body, erased posterior angles of pronotum, and raised but non-forked mesoventrite. Strictly **alpine species from both sides of central Pyrenees at altitudes over 1000 m**. Seven possible subspecies of *T. strangulata* have been described (Gómez-Zurita & Kippenberg 2010), and the endophalli of two of them have been analyzed.

These two subspecies display almost identical internal sacs (figs 12a, b, c, d) with strongly reduced manubrium; their endophalli are of elongate elliptic shape without clear apical protuberances, although their dorsal part is covered with small thorns.

***T. (T.) strangulata epipleuralis* Bechyné, 1953**

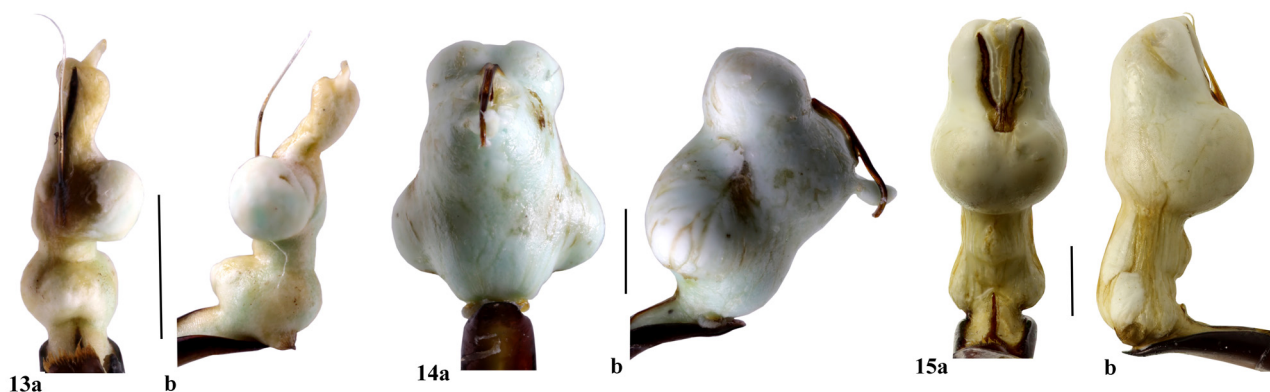
Material: 2 ♂: Canfranc: cabaña de la vuelta de Iserias, Huesca, 15.06.2005 leg. A. Anichtchenko (AAC); 2 ♂: Canfranc: barranco de Izas, 1600m alt., Huesca, 18.06.2005, leg. A. Anichtchenko (AAC).

***T. (T.) strangulata sculptipennis* Bechyné, 1948b**

Material: 1 ♂: Port de la Bonaigua, 2000m alt., Lleida, 23.08.1968, leg. E. Durall (EPC).

“*T. calceata*”—group

Description: in this monotypic group, *T. calceata* is easily recognized by the vivid red colour of their femora medially and tibiae. Body of **small–medium size (10.0–12.0 mm.)** with bright black upper side, a heart-shaped pronotum covered with a strong punctuation, mesoventrite short, notably forked, and elytra with dense and coarse punctures. Endemic species of the Iberian Northwestern area mostly.



FIGURES 13–15. Endophallus structure of “*T. calceata*”, “*T. scabripennis*” and “*T. rugosa*” groups: 13a, b—*T. calceata* (Navahermosa); 14a, b—*T. (scabripennis)* (Ceuta); 15a, b—*T. espanoli* (Jumilla). Scale = 1 mm.

***T. (T.) calceata* Pérez Arcas, 1865**

Material: 2 ♂: Galapagar, Madrid, 12.02.1989, leg. A. Anichtchenko (AAC); 1 ♂: San Sebastián de los Reyes, Madrid, 18.10.1997, leg. A. Anichtchenko (AAC); 1 ♂: Uclés, Tarancón, Cuenca, 29.03.2006, leg. A. Anichtchenko (AAC); 3 ♂: Navahermosa, Toledo, 27.02.2007, leg. A. Anichtchenko (AAC).

Description: The endophallus structure (figs 13a, b), is very similar to those of the “*T. goettingensis*”—group, but it differs from all the species known so far for this character by their asymmetric DA protuberances, namely a hypertrophic left and a reduced right protuberance. The basal part of endophallus is sphaerical, dorsally with

merged DB protuberances, and with clearly visible VB. The flagellum is weakly sclerotized contrary to the manubrium, which is long, narrow and with parallel sides.

“*T. scabripennis*”—group

Description: Body of large-medium size (12.0–14.5 mm.) with convex pronotum, laterally feebly rounded and distinctly sloping. Mesoventrite slightly raised and grooved. Elytra with undulating surface. Six or seven North African species distributed from Morocco to Tunisia (Bechyné 1948a; Warchałowski 2010). One single species studied:

***T. (T.) scabripennis* Fairmaire, 1868**

Material: 1 ♂: W Ceuta: Allabacal, NW Africa, 17.02.2004, leg. A. Anichtchenko (AAC).

Description: Pronotum with round sides, punctured laterally, and elytra dull, feebly corrugated and strongly punctured. Endophallus globular (figs 14a, b) with two large round apical expansions, plus one median and two basal enlargements, and a divided, narrow and elongate manubrium, joined to a rather thick and short flagellum.

“*T. rugosa*”—group

Description: mostly large species having a raised non-forked mesoventrite, transversally cut or emarginate, and elytra densely and coarsely punctured, strongly wrinkled. It is constituted by several North African species with only one Iberian taxon, *T. espanoli* (Warchałowski 2010).

***T. (T.) espanoli* Bechyné, 1948a**

Material: 1 ♂: Molina de Segura: La Alcaina, Murcia, SE Spain, 24.09.1989, leg. J. Serrano (EPC); 3 ♂: Jumilla: rambla del Taray, Murcia, SE Spain, 05.11.2000, leg. J.L. Lencina (AAC and EPC).

Description: Legs with tibiae and femora or only femora of red dull colour, pronotum with fine and shallow punctures in the middle, and elytra very strongly and deeply punctured, but only wrinkled at apex (Warchałowski 2010). Endophallus (figs 15a, b) with narrow and straight basal part and strongly inflated apically, manubrium divided in two sclerotized lateral lobes and flagellum relatively short and sclerotized. This endophallus reminds of that of *T. intermedia* (figs 3a, f).

“*T. goettingensis*”—group

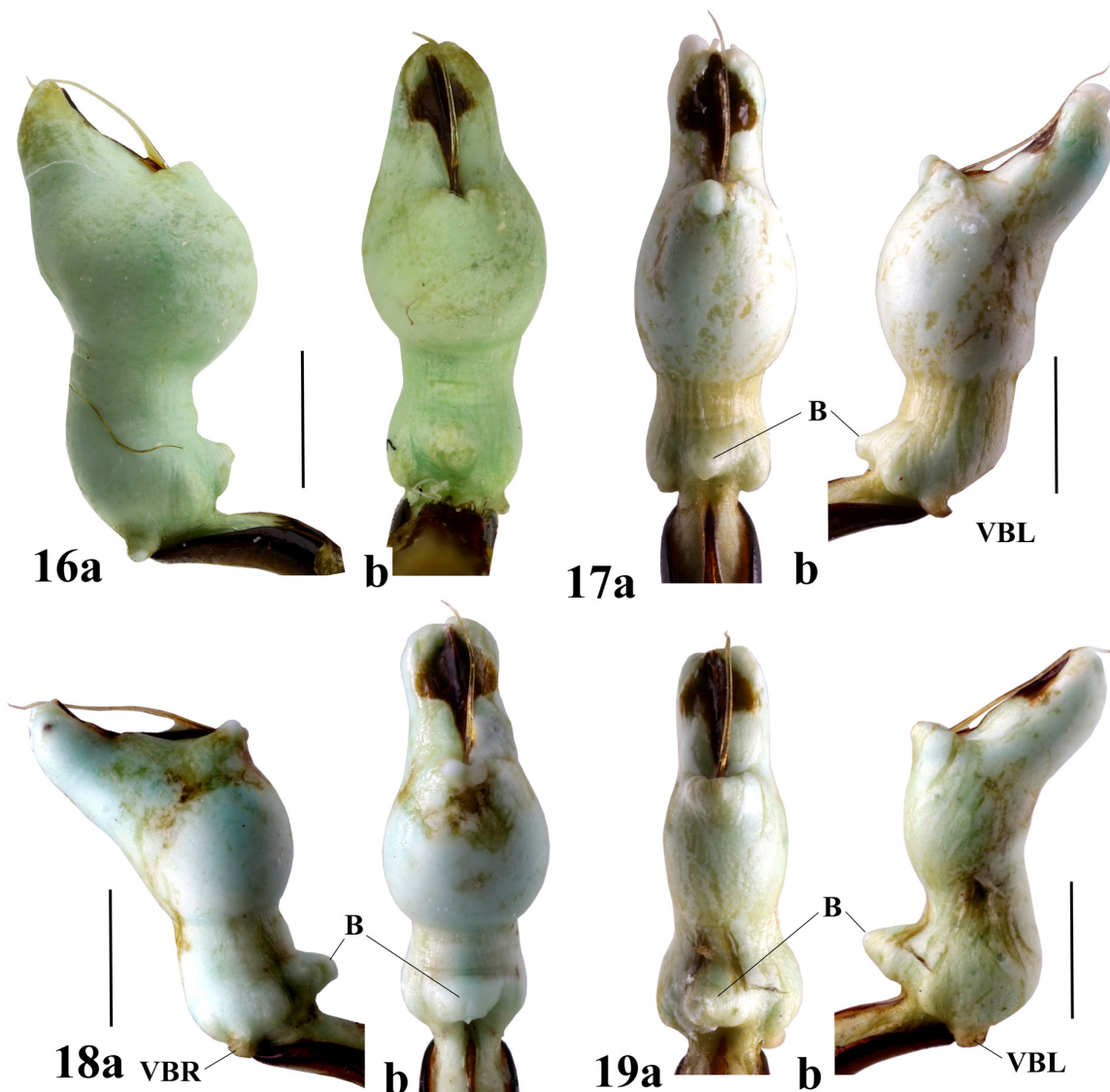
Diagnosis: We named this group (Petitpierre 1970a), for a complex of allopatric taxa resembling to the European species *T. goettingensis* (Linnaeus, 1758), with small or medium sized body, pronotum with straight or round sides, mesoventrite short, raised or almost flat but generally non-forked, and elytra densely punctured or even with coarse punctures and wrinkled. This complex, probably including at least nine of the 28 groups of Bechyné (1948a), was first substantiated based on similar adult appearance and karyotypes (Petitpierre 1970b, 1976; Gómez-Zurita *et al.* 2004), and later on with mitochondrial and nuclear phylogenies (Gómez-Zurita *et al.* 2000a, 2000b, 2000c; Gómez-Zurita & Vogler 2003; Gómez-Zurita 2004, 2008). Although many of these species have shown similar or identical endophalli, a few others have displayed highly divergent structures.

***T. (T.) geniculata* (Germar, 1824)**

***T. asturiensis* Kraatz, 1879 syn. nov.**

Material: 1 ♂: Puerto de San Glorio, 1600m alt., Cantabria—Palencia, 11.07.1997, leg. J.L. Zapata (EPC); 3 ♂: *ibid*, 31.05.1997, leg. J.L. Zapata (AAC); 2 ♂: Picos de Europa: las Colladinas, Macizo Central, 2100m alt, Asturias, 08.08.2003 leg. A. Anichtchenko (AAC); 2 ♂: Montes Cantábricos: Peña Ubiña, 2300 m alt., Cantabria, 05–06.07.2009, leg. A. Anichtchenko (AAC); 2 ♂: Sierra de Guadarrama: 2 km. E. Navacerrada, 1900 m alt., Segovia, 01.09.2002, leg. A. Anichtchenko (AAC); 2 ♂: Sierra de Guadarrama: Puerto de la Morcuera, 1796m alt., 07.04.1996, Madrid, leg. J.L. Zapata (AAC); 3 ♂: Villafuerte, Valladolid, 05.03.2002, leg. J.L. Zapata (AAC); 1 ♂: Puerto de Navalmoral, 1514 m alt., Ávila, 01.03.1997, leg. J.L. Zapata (AAC); 6 ♂: Campisabales, Guadalajara, 28.03.1998, leg. J.L. Zapata (AAC); 4 ♂: Pl. Arnau, Asturias, 02.08.1992, leg. J.L. Zapata (AAC).

Description: This taxon was described for individuals with dull red femora as the only distinctive character with respect to allied and neighboring populations. *T. geniculata* and other similar taxa share a partial or even complete absence of basal rim of pronotum, but they display rough elytra and the last male maxillary palpomeres feebly expanded apically. They are found in the NW quadrant and Central Iberia. The characteristics of endophalli shown in the three previous taxa (figs 16–19), displays a subcylindrical base, an inflated middle part which is gradually narrowed at the apex, a very thin flagellum, filiform and weakly sclerotized, whereas in most of them the arrowhead-shaped manubrium with triangular apex is well-sclerotized.



FIGURES 16–19. Endophallus of “*T. goettingensis*” group: 16a, b—*T. geniculata* (Las Colladinas); 17a, b—*T. asturiensis* (Puerto de San Glorio); 18a, b—*T. perezii* (Villafuerte), 19b, c—*T. perezii* (Puerto de Navalmoral).

***T. (T.) monticola* Dufour, 1851**

Material: 1 ♂: Gourette, Pyrénées Atlantiques, France, 21.06.1970, leg. G. Tiberghien (EPC); 1 ♂: Monte Perdido: collado de Añisclo, 2400m alt., Huesca, 16.06.2003, leg. A. Anichtchenko (AAC); 3 ♂: Canfranc: barranco de Izas, 1600m alt., 18.06.2005, leg. A. Anichtchenko (AAC); 1 ♂: Canfranc: cabaña de la vuelta de Iserias, Huesca, 15.06.2003, leg. A. Anichtchenko (AAC).

Description: Species similar to *T. geniculata*, with pronotum widest postmedially and punctured smooth elytra. Both species have a well sclerotized of rhomboid or arrowhead-shaped manubrium and a thin and weakly sclerotized flagellum. The endophalli are also similar, with a short and subcylindrical basal part, a more or less inflated middle part, and gradually narrowed apical. They differ in shape of the manubrium, i.e. rhomboid in *T. monticola* but of arrowhead-shaped in *T. geniculata*. In addition, they are distinguished by the presence or near absence of basal protuberances, respectively. Thus, the DB, VB and B are small in *T. geniculata* (figs 16–19), but VB is small whereas DB and B are almost indistinct in *T. monticola* (figs 20a, b).

***T. (T.) cyanescens* Fairmaire, 1862**

Material: 1 ♂: Noja: el Pinaruco, Cantabria, 07.09.2001, leg. J.L. Zapata (EPC); 1 ♂: Sierra de Aralar: San Miguel, 1250m alt., Navarra, 06.01.2001, leg. A. Anichtchenko (AAC); 2 ♂: Sierra de Aralar, Navarra, 03.06.2006, leg. A. Anichtchenko (AAC); 1 ♂: Bera de Bidasoa: Larrun, 850m alt., Navarra, 09–15.09.2001, leg. A. Anichtchenko (AAC); 1 ♂: Bera de Bidasoa, Navarra, 12.01.2002, leg. A. Anichtchenko (AAC); 1 ♂: Bera de Bidasoa: 3 km N, Navarra, 14.09.2002, leg. A. Anichtchenko (AAC); 2 ♂: Irún: Jaitzubia, Guipúzcoa, 23.05.2005 leg. A. Anichtchenko (AAC).

Description: Body stout with a bluish or violaceous sheen. Pronotum subquadrate, almost twice as broad as long. Elytra short and stout, with shallow and rather disperse punctures. It has several local races, six of which are considered as valid subspecies, distributed at low and median altitudes from Navarra to Eastern Asturias in Spain, and also in the Pyrenees Atlantiques of France (Bechyné 1953; Gómez-Zurita 2008).

The endophalli (figs 21a, b, c, d) of these taxa resemble to those of the previous two species although in some populations they are narrower and more slender, their manubria can be divided or simple, and their flagelli are thin and weakly or moderately sclerotized. We have tentatively considered it as a distinct species based on the external morphology but the shape of internal sacs are not clearly different from those of the previous taxa, therefore, its taxonomic category is doubtful.



FIGURES 20–21. Endophallus of “*T. goettingensis*” group: 20a, b—*T. monticola* (Canfranc); 21a, b—*T. cyanescens* (Bera de Bidasoa), 21c, d—*T. cyanescens* (Noja); 22a, b—*T. simatocollis* (Campelles); 23a, b—*T. monserratisensis* (Montseny); 24a, b—*T. interstitialis* (Canet); 25a, b—*T. piochardi* (Alió). Scale = 1 mm.

***T. (T.) sinuatocollis* Fairmaire, 1862**

Material: 2 ♂: Campelles, Pyrenees of Girona, Spain, 12.10.1970, leg. E. Petitpierre (EPC).

Description: Species of an ambiguous position because it has been classified as an independent species (Bechyné 1948a, 1948b, 1953; Petitpierre 1970b; Warchałowski 2003, 2010), a subspecies of *T. goettingensis* (Winkelman & Debreuil 2008), or a subspecies of *T. interstitialis* Fairmaire, 1861 (Jeanne 1967). It is characterized by the small size (usually less than 10.5 mm.) and the slightly sinuous or straight posterior angles of pronotum. Distributed in eastern Pyrenees of Spain and France.

Endophallus (figs 22a, b) strongly curved in a Z-shape. Basal protuberances small but distinct. Manubrium rhomboid and well sclerotized and flagellum thin and weakly sclerotized.

***T. (T.) sinuatocollis monserratis* Bechyné, 1962 comb. nov.**

T. goettingensis monserratis Bechyné, 1962: 184

Material: 1 ♂: Rupit, Barcelona, 04.1966, leg. E. Petitpierre (EPC); 1 ♂: Collformic, Montseny, 970 m alt., Barcelona, 19.07.1970, leg. Petitpierre (EPC).

Description: Very similar to the previous taxon although a bit larger (9.0–11.5 mm) and broader. Distributed in some mountains higher than 700 m altitude of middle-eastern Catalonia, NE Spain.

The endophallus structure (figs 23a, b) is almost identical to that of *T. sinuatocollis* and not to *T. goettingensis*, contrary to its description. It shows only some minor differences in the shape of manubrium, which is a little broader than in *T. sinuatocollis*.

***T. (T.) interstitialis* Fairmaire, 1884**

Material: 1 ♂: Canet: étang de Saint Cyprien, Pyrénées Orientales, France, 27.07.1972, leg. G. Tiberghien (EPC).

Description: Body similar in size to the previous species. Pronotum sides slightly but evenly round and elytra with dense but shallow punctation. Upper side black or with a feeble metallic reflex. Distributed at low altitudes in SE France and NE Catalonia.

Endophallus (figs 24a, b) with short and cylindrical base and apical part with slanted region at apex where a rhomboid manubrium and a thin, weakly sclerotized flagellum are placed.

***T. (T.) piochardi* Fairmaire, 1873 stat. nov.**

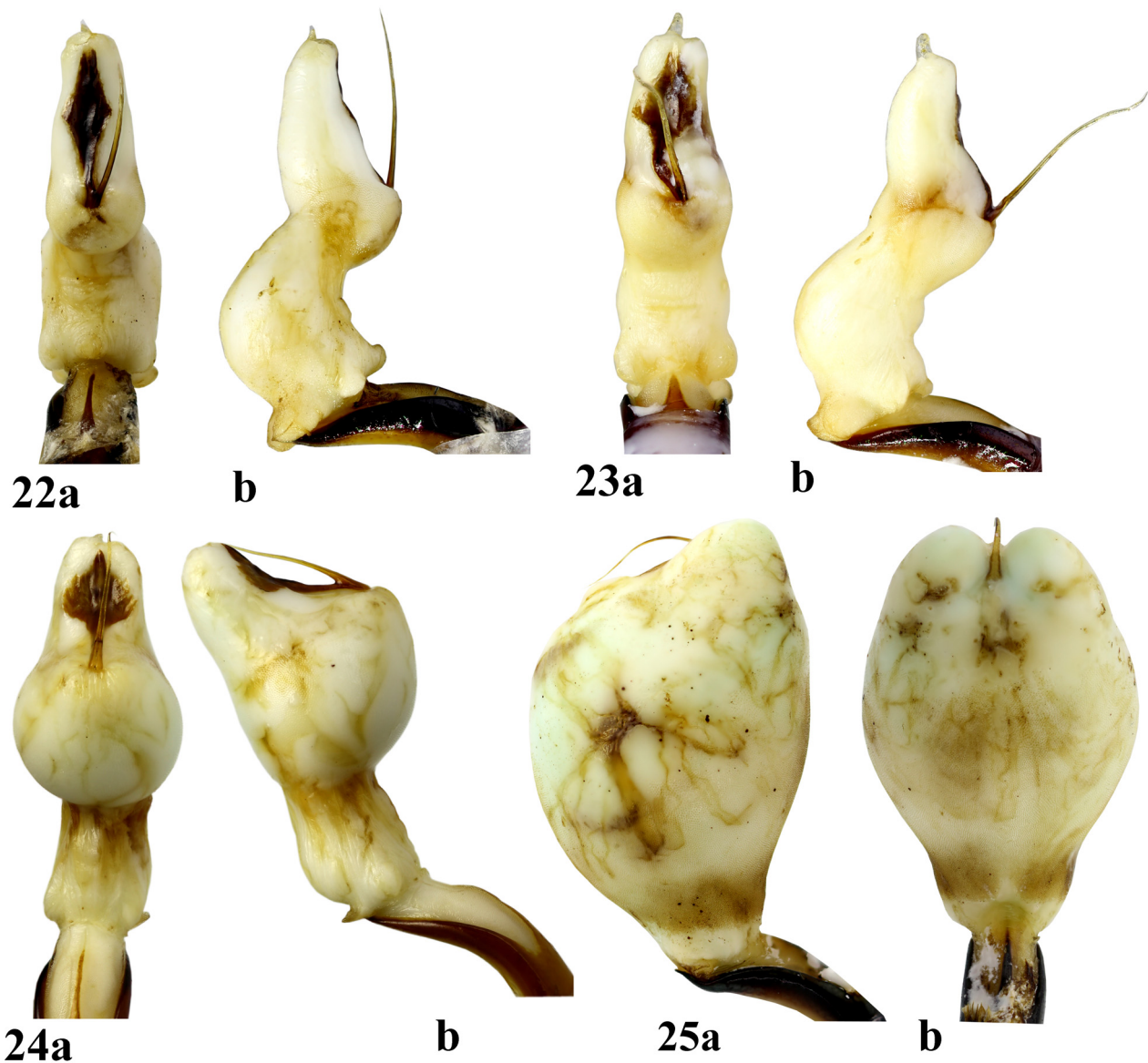
Material: 8 ♂: Alió, Tarragona, 26.11.1967, leg. E. Petitpierre (EPC); 1 ♂: Garraf, Barcelona, 04.1972, leg. E. Petitpierre (EPC); 9 ♂: Tibidabo Mt., Barcelona, around 300 m alt., 10.1966, leg. E. Petitpierre (EPC).

Description: Males and females of dull black or bronze colour. Fairmaire (1884) wrote that this species has a curved pronotum with posterior angles slightly sinuate, and a densely and finely punctured surface, with elytra also densely but coarsely punctured, with interstices very finely punctate. Bechyné (1948a) classified this taxon as a subspecies of *T. fallax* based on the pronotum slightly more shining than elytra in both sexes, and both with dense small punctures. The study of the endophallus (figs 25a, b), revealed that it is clearly different from that of *T. fallax* (figs 26a, b, c) owing to stout oviform shape with two small DA protuberances and a narrowed basal part; filiform-manubrium, divided; and flagellum weakly sclerotized. A new status for *T. piochardi* is proposed here, and the species would be tentatively placed in the “*T. goettingensis*”—group.

“*T. fallax*”—group

Description: the taxa of this group have been considered within the “*T. goettingensis*” complex based on morphological, karyological and molecular phylogenetic grounds (Petitpierre 1970a, 1970b, 1976; Gómez-Zurita

et al. 2000a, 2000b, 2000c; Gómez-Zurita 2004). However, their large body (11.5–15.0 mm.), with pronotum with more pronouncedly curved sides, mesoventrite slanted or even forked, and shallow and relatively spread punctation of elytra, allow to its diagnosis with regard to taxa in the former group. It is distributed from southern Catalonia to Murcia and in central-eastern Iberia. Furthermore, the globular and highly complex structure of most endophalli clearly substantiate this group of species.



FIGURES 22–25. Endophallus of “*T. goettingensis*” group: 22a, b—*T. sinuatocollis* (Campelles); 23a, b—*T. monserratis* (Montseny); 24a, b—*T. interstitialis* (Canet); 25a, b—*T. piochardi* (Alió). Scale = 1 mm.

***T. (T.) fallax* Pérez, 1865**

Material: Three specimens studied: 2 ♂: Paterna, Valencia, 12.1970, leg. J.M. Durall (AAC, EPC); 1 ♂: Serra, Valencia, 17.01.2006, leg. A. Anichtchenko (AAC).

Description: last palpomeres of males clearly widened apically. Pronotum heart-shaped, with sides conspicuously slanted posteriorly, disk covered with dense and rather strong punctation. Mesoventrite generally forked. Elytra with homogeneous and relatively disperse punctation. Endophallus (figs 26a, b, c) with strongly enlarged and bent apical part at right angle to the middle and basal parts, VA hypertrophied protuberances, with two additional small and widely separated protuberances at the ventral side. Manubrium long, filiform and divided, with a sawed flagellum.

***T. (T.) tortosensis* Bechyné, 1948a stat. nov.**

Material: 3 ♂: Aldea, Tarragona, 03.1969, leg. E. Petitpierre (EPC).

Description: this taxon was described as a subspecies of *T. fallax* for its bright and more densely punctured pronotum and elytra without undulated interstices, from which it can also be separated by a strikingly distinct internal sac. Endophallus (figs 27a, b) composed of three parts: basal part globular and longer than middle and apical parts combined; middle part with two double dorsal protuberances in angle; and apical part enlarged with two double lateral subspheres. Manubrium very broad and clearly differing from that of *T. fallax*. Distributed in southernmost Catalonia.

***T. (T.) aurichalcea* Bechyné, 1948a**

Material: 3 ♂: Cantavieja: Puerto del Cuarto Pelado, 1612m alt., Teruel, Aragón, 06.05.2000, leg. J.L. Zapata (AAC, EPC); 1 ♂: Puerto de Linares, 1665m alt., Teruel, Aragón, 08.05.1998, leg. J.L. Zapata (AAC); 3 ♂: Puerto de Piqueras, 1713m alt., Soria, Castilla-León, 03.06.1993, leg. A. Anichtchenko (AAC); 1 ♂: Puerto del Portillo, 1778 m alt., Teruel, Aragón, 21.05.2006, leg. A. Anichtchenko (AAC); 1 ♂: Tragacete, Cuenca, Castilla-La Mancha, 05.07.1995, leg. E. Petitpierre (EPC).

Description: Body with black metallic sheen on upper side and pronotum very finely and uniformly punctate. Distributed in several mountains of Central and Eastern Spain. Endophallus (figs 28a, b, c) with very enlarged apical part, bent at right angle dorsally as in *T. fallax*, but easily distinguishable by the long DA protuberances and the low large and fused ones. Manubrium divided in two narrow and filiform halves, and flagellum widened medially.

***T. (T.) aitanae* Petitpierre et Anichtchenko sp. nov.**

Material: Holotype, ♂, Serra d'Aitana, Alicante, 01.11.1985, leg. M. Beltrán (MNCN); Paratypes: 1 ♂: Tarbeba: coll de Rates, 750m alt., Alicante, 07.12.1984, leg. J.L. Lencina (EPC); 1 ♂, 1 ♀: Alcolexa: port de Tudons, 1000 m alt., Alicante, 01.11.1985, leg. J.M. Beltrán (AAC, J.L. Lencina coll.).

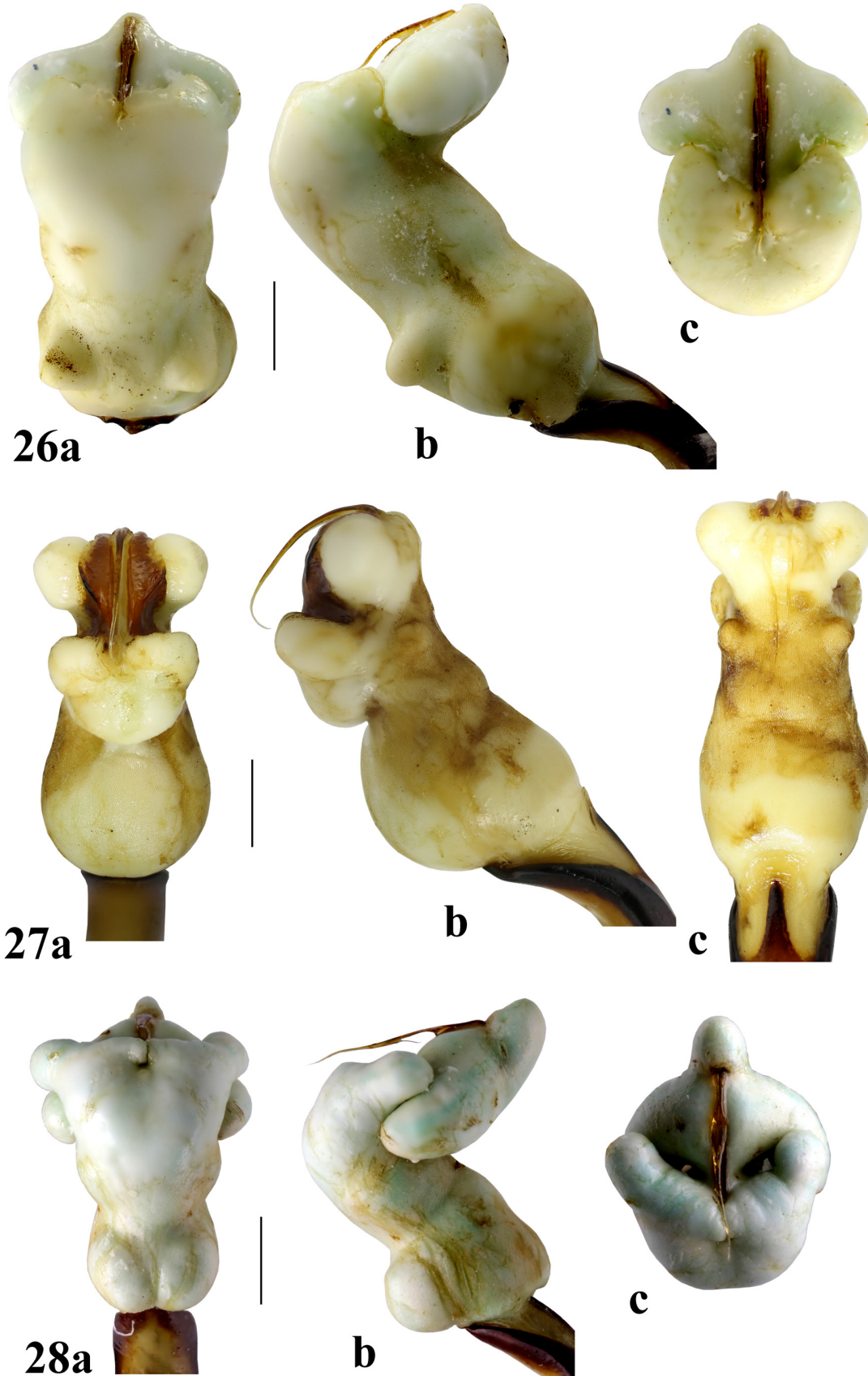
Description: Males 10.0–12.0 mm. Female: 12.5 mm.

Body convex, of a dull black or black-brown colour. Head with fine puncturation denser on clypeus than on vertex, maxillary palpi of males with last palpomere strongly widened apically, less so in females; antennae rather thin, as long as body length in males, shorter in females. Pronotum transverse, twice as wide as long, with projecting anterior angles, sides regularly curved, slightly more at basis, widest slightly before middle, lateral rims complete and visible at their whole extension from above, and tegument covered with fine, moderately dense and uniform punctation, larger at anterior angles. Legs black, with protarsi and mesotarsi of males strikingly expanded, narrow with glabrous longitudinal sole in females. Elytra globose, widest just before middle, covered with fine but coarser punctation than pronotum, and very fine and sparse punctures on some interstices, both in males and females. Proventrite almost flat; mesoventrite short, clearly forked; metaventrite flat, covered with fine puncturation, subequal to puncturation of abdominal ventrites. Endophallus (figs 29a, b, c) with three sections; basal part at a right angle to apical two parts; apical region with two large lateral lobes and finger-like appendix at apex, manubrium divided, narrow and elongated and flagellum short moderately sclerotized.

Derivatio nominis: from Serra d'Aitana, the highest mountain in the Alicante province, Valencia, Eastern Spain.

***T. (T.) oblongula* Fairmaire, 1880**

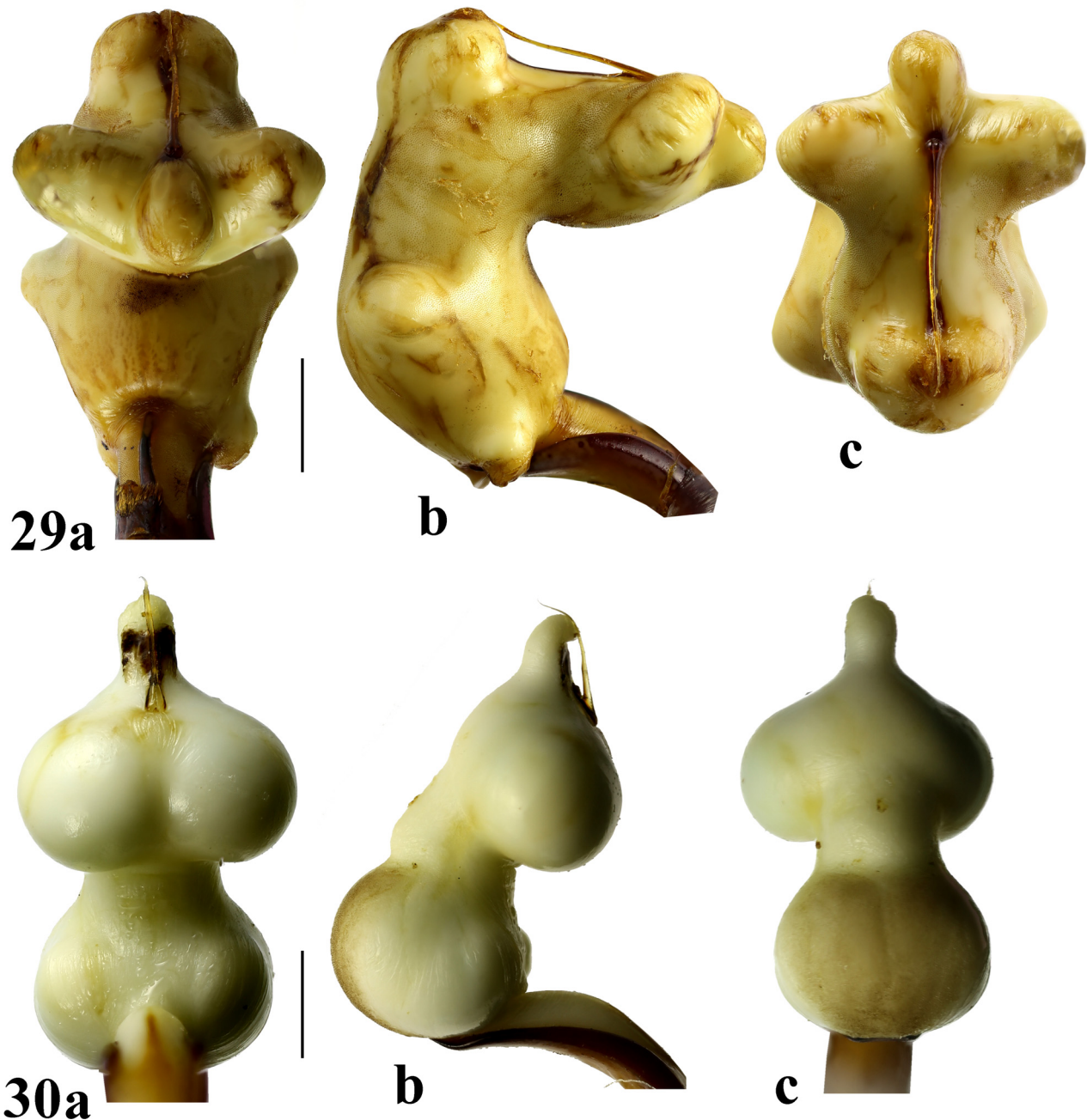
Material: 2 ♂: Camarena de la Sierra, Sierra de Javalambre, Teruel, Aragón, 15.05.1993, leg. J.L. Zapata (AAC, EPC); 2 ♂: Morella, Castelló, Valencia, 19.04.1993, leg. J.A. Alcover & G. Pons (MNCN ex. 8542); 1 ♂: Alcoi: la Font Roja, Alicante, 16.10.1995, leg. E. Petitpierre (EPC); 2 ♂: Jumilla: Sierra del Carche, Murcia, SE Spain, 28.10.1985, leg. J.L. Lencina (EPC); 1 ♂: Jumilla: los Gavilanes, Murcia, SE Spain, 04.09.2002, leg. J.L. Lencina (JLC).



FIGURES 26–28. Endophallus structure of “*T. fallax*” group: 26—*T. fallax* (Paterna); 27—*T. tortosensis* (Aldea); 28—*T. aurichalcea* (Tragacete)

Description: body middle-sized (9.0–10.5 mm) and elongate, with a slightly shining black colour. Last palpomere of males subovoid, not widened at the tip. Head and pronotum with extremely fine punctures, the latter with regularly curved sides, and elytra with shallow and disperse coarse punctures. Proventrite broad and clearly furrowed. Mesoventricle almost flat and straight at apex.

Endophallus (figs 30a, b, c) with two globular parts and a finger-like apex, with extremely enlarged and merged DB and VB protuberances. Manubrium broad and strongly sclerotized, with truncate apex and long retracted lateral lobes. Flagellum short and moderately sclerotized.



FIGURES 29–30. Endophallus structure of “*T. fallax*” group: 29—*T. aitanae* sp. nov. (Serra d’Aitana); 30—*T. oblongula* (Morella). Scale = 1 mm.

“*T. hispanica*”—group

Description: Body roundish or oval. Pronotum with slightly curved sides and covered with dense, strong or

moderate punctures. Mesoventrite forked. Elytra with strong but disperse punctures or wrinkled with furrows separating rugosities. The group is composed of few endemic species from the Iberian Peninsula. It is the sister group to the *T. goettingensis* complex based on the molecular phylogeny of *Timarcha* (Gómez-Zurita 2004).

***T. (T.) hispanica* Herrich-Schäffer, 1838**

Material: 2 ♂: Sagres, Cabo de Sao Vicente, Algarve, Portugal, 18.03.1997, leg. E. Petitpierre (EPC).

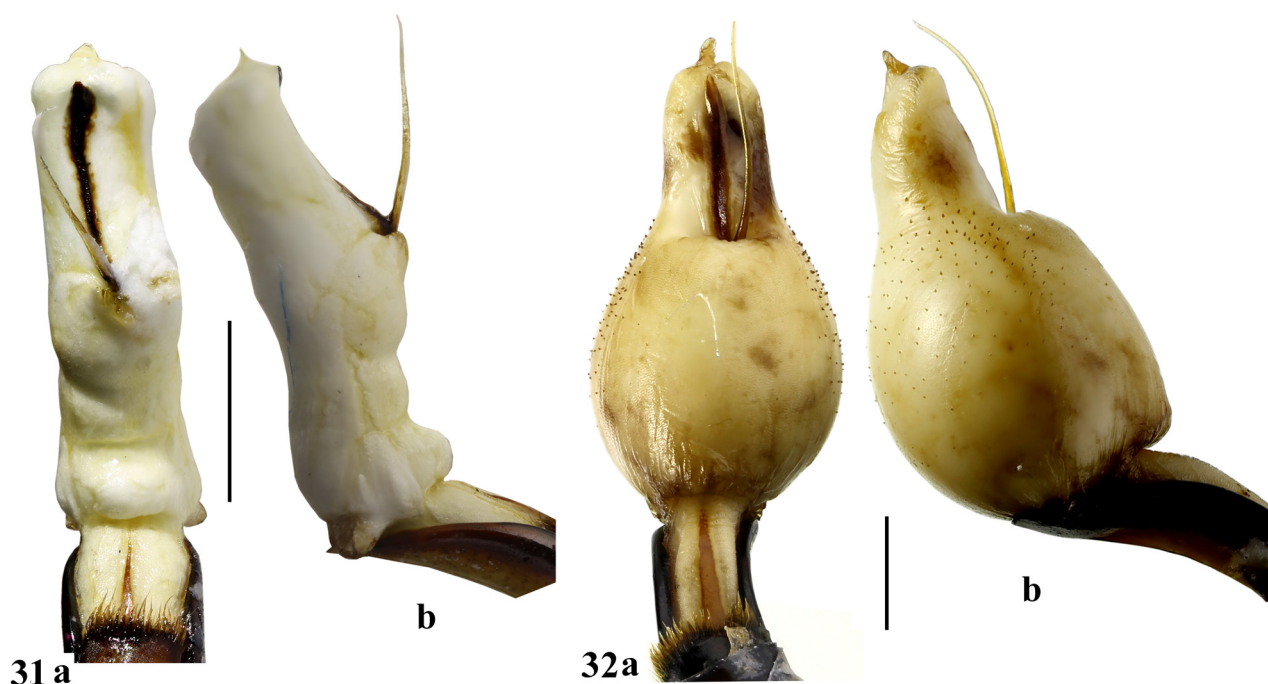
Description: Species with several isolated subspecies and populations, quite different in habitus, which are found in Central and Southern regions of the Iberian Peninsula (Warchałowski 2010). Endophallus (figs 31a, b) with simple outline made of two parts, the basal one expanded dorsally with two ventral protuberances and the apical one slanted dorsally to hold a narrow manubrium and a short flagellum, ending in a round tip. This structure resembles to that of *T. geniculata*, but it is enlarged at the basis unlike the apical part which is more complex.

***T. (T.) granadensis* Bechyné, 1948a**

Material: 1 ♂: Huéscar, Puerto de la Sagra, 1600m alt., Sierra Guillimona, Granada, Andalusia, 14.05.2007, leg. E. Petitpierre (EPC).

Description: Body shining black, small (less than 10.0 mm), with strongly punctured head, pronotum heart-shaped with sparse punctation and elongate, coarsely punctate and rough elytra.

Endophallus (figs 32a, b) very characteristic in its simple structural plan, with a large globular basis laterally covered with small spines and ending in a short narrowed apex. Manubrium long, with a well sclerotized central shaft and short and weakly sclerotized lateral triangular lobes. Flagellum weakly sclerotized.



FIGURES 31–32. Endophallus structure of “*T. hispanica*” group: 31—*T. hispanica*; 32—*T. granadensis*. Scale = 1 mm.

Discussion and conclusions

Our results on the endophalli of 32 taxa of *Timarcha* have provided a worthy source of taxonomic information to separate cryptic species and to check the reliability of morphological groups by Bechyné (1948a), in agreement mainly with DNA phylogenies, but also on chromosomes and host-plant affiliations.

The discrimination between the subgenus *Metallotimarcha* and some *Timarcha* s. str. is slight because the endophallus of *T. (Metallotimarcha) hummeli* is of a simple globular shape as found in a few *Timarcha* s. str., although most of the latter are much more complex with at least two well-characterized parts. However, the tegmen apex of the *Metallotimarcha* species is larger than in any of the *Timarcha* s. str. (Khnzorian 1966; Petitpierre 1970a), and this aedeagus trait together with the slight sexual dimorphism of adults (Bechyné 1948a; Jolivet 1948; Warchałowski 2010), agree with the basal position of *Metallotimarcha* in DNA phylogenies (Gómez-Zurita 2004), and the high diploid number of $2n = 38$ chromosomes in *T. metallica* (Petitpierre 2016), higher than in any *Timarcha* s. str., all of whose species have lower chromosome numbers (Petitpierre 1976; Gómez Zurita *et al.* 2004).

Conversely, the endophalli of the species formerly placed in the subgenus *Timarchostoma* Motschulsky, 1860 do not show any distinctive trait that could give support to this subgenus, in agreement also with DNA phylogenies and cytogenetic findings (Petitpierre 1976; Gómez-Zurita *et al.* 2000a, 2000b, 2000c; Gómez-Zurita 2004), as well as on the opinion of several authors based on the external morphology of adults only (Jeanne 1967; Daccordi 1994; Winkelman & Debreuil 2008; Gómez-Zurita & Kippenberg 2010; Tiberghien 2014).

Four taxa of *Timarcha* with a forked mesoventrite and scattered shallow punctuation on elytra, those of “*T. intermedia*” and “*T. lugens*” groups of Bechyné (1948a), *T. intermedia* s.str., *T. intermedia lugens* **stat. n.**, *T. intermedia kiesenwetteri* **stat. n.** and *T. intermedia carmelenae* **stat. n.**, share almost identical endophalli made of a globular structure with a narrow basal part and an asymmetric horn at apex. Although they might be considered as the same species with a subspecies rank, we should prefer waiting for a complete molecular phylogeny of the four taxa, because only the two first mentioned have been analyzed using these data (Gómez-Zurita 2004). Nevertheless, their near relationships are also based on their shared unusual trophism on Brassicaceae plants (Jolivet & Petitpierre 1973; Petitpierre 1970b, 1976; González-Megías *et al.* 2003), plus similar geographic distributions in the Southeastern Iberian area.

Two further morphological groups of Bechyné (1948a) with a forked mesoventrite and scattered shallow punctuation on elytra, that of “*T. insparsa*” with two species, *T. insparsa* and *T. marginicollis*, and that of “*T. parvicollis*” with five subspecies, both inhabiting the south of the Iberian Peninsula (Bechyné 1948a), also have globular endophalli but more complex than in the previous taxa, and clearly distinctive to each other. In addition, the endophallus of *T. insparsa* resembles more that of *T. parvicollis* than that of *T. marginicollis*, both of the former having a greatly enlarged apical part, a trait which is correlated with closest vicinity of *T. parvicollis* and *T. insparsa*, contrary to either of them with *T. marginicollis* as shown in the molecular phylogeny of the genus (Gómez-Zurita 2004). *T. marginicollis* displays a median lobe with a bi-toothed apex (Petitpierre 1970a), a trait which has not been found in any other congeneric species. This character and the phylogenetic position of *T. marginicollis* within a clade including many North African species of *Timarcha* (Gómez-Zurita 2004), gives further support to its separation from either *T. insparsa* or *T. parvicollis* morphological groups of Bechyné (1948a).

T. strangulata and *T. calceata*, although very distinct morphologically in adults and belonging to clearly separated morphological groups (Bechyné 1948a; Warchałowski 2010), show endophalli without any structural similarity, very simple and fusiform in *T. strangulata* whereas it has three distinct parts in straight line in *T. calceata*, as in a few other taxa of the *T. fallax* group. However, they appear in the same clade in the combined DNA phylogeny (Gómez-Zurita 2004), and both have high diploid chromosome numbers, $2n = 28$ and $2n = 30$ chromosomes, respectively (Petitpierre 1970b, 1976; Gómez-Zurita *et al.* 2004). Therefore, the observed differences in their endophalli correspond with adult morphology but not with their phylogeny and cytogenetic data.

T. balearica, the only species with uniformly flat elytra, has also an endophallus made of three parts, somehow reminds that of *T. intermedia*, though much more exaggerated. In the molecular phylogeny of *Timarcha*, *T. balearica* is located as an independent clade from that of *T. intermedia* and *T. lugens* (Gómez-Zurita 2004). These three species share a stout body, a heart-like pronotum and a forked mesoventrite but neither molecular phylogeny nor their karyotypes could provide any clue in concern with their origin.

The North African *T. scabripennis*, its Iberian relative *T. espanoli*, and the European *T. tenebricosa* share a divided manubrium and a globular endophallus, with or without large round basal expansions. These three species despite of their belonging to three different adult morphological groups (Bechyné 1948a; Warchałowski 2010) and showing diverse chromosome numbers, $2n = 28$, $2n = 26$ and $2n = 22$, respectively, (Petitpierre 1970b; Gómez-Zurita *et al.* 2004), belong to the same large clade in the molecular phylogeny of *Timarcha* (Gómez-Zurita 2004). Thus, their endophalli show a better agreement with molecular phylogeny than external morphology or karyotypes.

In the *T. goettingensis* complex (Petitpierre 1970b; Gómez-Zurita *et al.* 2000a, 2000b), *T. geniculata*, *T. asturiensis*, *T. perezii*, *T. monticola* and *T. cyanescens* display endophalli with a short subcylindrical basal part, a more or less inflated middle part gradually narrowed at apex, with a rhomboid or arrowhead-shaped manubrium and a thin weakly sclerotized flagellum. The endophalli of the first three are identical and the molecular phylogeny of different populations of *T. geniculata* and *T. perezii* interspersed within the same subclade (Gómez-Zurita *et al.* 2000a, 2000b, 2000c; Gómez-Zurita 2004), both data pointing out to their synonymy. However, the slightly different endophalli of *T. monticola* and *T. cyanescens* with regard to the former species deserves keeping their species status. All these species share partial or complete absence of pronotum basal rim and belong to the Western clade of the *T. goettingensis* complex mainly distributed in the central Spain and northeastern Iberian area (Gómez-Zurita *et al.* 2000b, 2000c, Gómez-Zurita 2008). Although Bechyné (1948a) and Warchałowski (2010) classified these species in three different groups, those of *T. geniculata*, *T. perezii* and *T. monticola*, they are not substantiated because they belong to the same group in Western clade within the *T. goettingensis* complex.

Most taxa closely related to the European *T. goettingensis*, such as *T. interstitialis*, *T. sinuatocollis* and *T. monserratisensis*, but not *T. piochardi*, share endophalli similar to the previous group of species, with two distinctive parts, the basal short and cylindrical and the apical slanted, a rhomboid manubrium and a weakly sclerotized flagellum. Since those of *T. sinuatocollis* and *T. monserratisensis* do not differ in their structures, and some populations of the former and the only one of the latter have shown a hybrid origin, derived from the same maternal mtDNA (*cox2*) and paternal nDNA (*its2*) sources (Gómez-Zurita 2008), these findings allow us to propose their synonymy but keeping a subspecies rank for *T. monserratisensis*. Moreover, the endophallus of *T. interstitialis* is similar but not identical to those of *T. sinuatocollis*, which supports their taxonomic separation, and the enlarged spindle-shaped of *T. piochardi* is completely different from any of the previous species. The molecular phylogeny of this group of species is complicated because one population of *T. interstitialis* is close to another of *T. sinuatocollis*, while a second of *T. interstitialis* is located near to a German population of *T. goettingensis* (Gómez-Zurita 2004). Later, a population of *T. interstitialis* and one of *T. piochardi* (sub *T. fallax*), geographical neighbours, showed also a hybrid origin with the maternal mtDNA (*cox2*) from an Eastern and the paternal nDNA (*its2*) from a Western source (Gómez-Zurita & Vogler 2006; Gómez-Zurita 2008). However, the conspicuous differences between their endophalli would exclude a possible conspecificity. As a good example of debatable taxonomic opinions in this group, based on morphological grounds, Jeanne (1967) claimed that *T. sinuatocollis* is a subspecies of *T. interstitialis* and more recently Winkelman and Debreuil (2008) put both of them as subspecies of *T. goettingensis*. Nevertheless, the reticulated evolution demonstrated in several examples of introgressive hybridizations within *T. goettingensis* complex (Gómez-Zurita & Vogler 2006), would need more insightful analyses before reaching a general taxonomic agreement.

The group of *T. fallax* (Bechyné 1948a; Warchałowski 2010), classified within the *T. goettingensis* complex group of species (Petitpierre 1970b; Gómez-Zurita *et al.* 2000), display the most divergent endophalli structures, including a simple ovoid shape in *T. piochardi* **stat. nov.**, species with uncertain position in this group of species, and complex arrangements as seen in *T. tortosensis* **stat. nov.**, *T. fallax*, *T. aitanae* **sp. nov.**, *T. aurichalcea* and *T. oblongula*. Their endophalli consist of two or three aligned parts or the basal one at right angle with the others, with peculiar distinctive protuberances. Although their adult morphological characteristics are quite similar, their endophalli show great differences which substantiate new taxonomic status in some of them. In addition, *T. aurichalcea* has the lowest chromosome number in the genus $2n = 18$, and a unique male sex-chromosome system (Gómez-Zurita *et al.* 2004). It seems that the differentiation in isolation of ancestral populations and in some cases followed by hybridization upon secondary contacts of these differentiated forms in Central Eastern mountains, from South Catalonia and Teruel in Aragón to Alicante and Murcia in Southeastern Spain could have contributed to the high phenotypic diversity, most presumably restricted to small geographic areas, except in the case of *T. aurichalcea* and *T. oblongula*.

The “*T. hispanica*” group includes two species, *T. hispanica* and *T. granadensis* with very different endophalli. Both show two parts as found in *T. geniculata* and its subspecies, *T. cyanescens*, *T. sinuatocollis* and *T. interstitialis*, but they can be separated from them because in that of *T. hispanica* these parts are of similar width while in that of *T. granadensis* the basal part is more inflated and covered with small spines, contrary to the other group of species except *T. interstitialis* whose basal part is even larger than in *T. granadensis*. The karyotype of *T. hispanica* with $2n = 20$ chromosomes separates this species from the one in most species of the “*T. goettingensis*” group, all with $2n = 20$ chromosomes, but the former with two instead of three large autosome pairs (Petitpierre

1976; Gómez-Zurita *et al.* 2004), and that of *T. granadensis* shows $2n = 22$ chromosomes, again with only two large autosome pairs but with eight small ones, instead of the seven found in *T. hispanica* (Gómez-Zurita *et al.* 2004). On the other hand, the molecular phylogeny of *Timarcha* shows *T. hispanica* and *T. granadensis* in the same clade (Gómez-Zurita 2004), supporting morphological and cytological inferences.

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