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Revised classification and phylogeny of an Afrotropical species group based on molecular and morphological data, with the description of a new genus (Coleoptera: Scarabaeidae: Onthophagini)

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26 **Revised classification and phylogeny of an Afrotropical species group based on**
27 **molecular and morphological data, with the description of a new genus**
28 **(Coleoptera: Scarabaeidae: Onthophagini)**

29

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37

38 **Abstract**

39 The worldwide distributed *Onthophagus* genus comprises at present more than 2,000
40 species, that often show a complicated and uncertain systematic history. In particular,
41 the many Afrotropical species included in this genus have never been entirely reviewed
42 after the division into 32 species-groups proposed by d'Orbigny in 1913, although
43 subsequent research focussing on some of these species suggested that *Onthophagus*
44 constituted a not monophyletic taxon. In order to highlight their phylogenetic
45 relationships, the various Afrotropical species-groups of d'Orbigny must thus be
46 examined, and it would be advisable to study them separately to avoid
47 misunderstanding. In this framework, the taxonomic position of the three species
48 currently included in the 21st d'Orbigny group was examined. Both morphological and
49 biomolecular analyses contributed in confirming that these species (i.e., *Onthophagus*
50 *caffrarius* d'Orbigny, 1902, *O. quadriceps* Harold, 1867 and *O. signatus* Fåhraeus,
51 1857) constituted a well-defined monophyletic group that cannot be maintained within
52 the genus *Onthophagus*. Therefore, the *Kurtops* **gen.n.** is here described to
53 accommodate these Afrotropical species, that are nevertheless always included within
54 the Onthophagini tribe. On the basis of the phylogenetic relationships here elucidated, it
55 was also emphasized that the new genus is strictly related to *Digitonthophagus* and
56 *Phalops*, thus it was proposed to include the three genera into a single clade of
57 suprageneric rank naming it as *Phalops* complex.

58

59 **Keywords.** *Onthophagus*; new genus; *Phalops* complex; molecular analysis;

60 morphological analysis; phylogeny; geometric morphometrics

61

62 **Introduction**

63 The widespread genus *Onthophagus* Latreille, 1802 comprises more than 2,000 species
64 and is thus one of the largest genera in the world (Emlen et al. 2005). It was
65 hypothesized that these dung beetles originated in Africa during the Oligocene (23-33
66 MYA) concurrently with the expansion of grassland habitats and the radiation of
67 mammals (Ahrens et al. 2014). They quickly spread from Africa, and now can be found
68 in all continents, with species living in a wide range of exceedingly different habitats
69 and feeding on every kind of dung (Emlen et al. 2005). Such a high biological
70 diversification corresponds to an extreme systematic complexity, that is exemplified by
71 the troublesome taxonomic history not only of the *Onthophagus* genus, but also of the
72 whole Onthophagini tribe.

73 The more than 700 Afrotropical *Onthophagus* species currently known are still divided
74 (for the most part) into the 32 species-groups proposed by d'Orbigny (1913), who
75 developed a system of dichotomous keys entirely based on characters of external
76 morphology for species recognition. The monophyly of the *Onthophagus* species-
77 groups was not expressly supported by the d'Orbigny compendium, and some of these
78 groups had to be removed from *Onthophagus*, and must be regarded as new entities
79 whose taxonomic rank requires a careful evaluation.

80 Over the years, a number of new taxa were described in order to accommodate some of
81 those species previously included in *Onthophagus*. A good example is the case of
82 *Digitonthophagus* Balthasar, 1959 that was described (together with others) as a
83 subgenus of *Onthophagus* (Balthasar 1959, 1963) and later elevated to generic rank
84 (Zunino 1981). Yet again in recent years more controversial classifications within the
85 Afrotropical *Onthophagus* d'Orbigny groups was developed (Moretto 2009; Tagliaferri
86 et al. 2012), but a lot remains unresolved due to the well-known species richness and
87 complexity of this megadiverse genus. As a result, not only the *Onthophagus* genus, but
88 the entire d'Orbigny classification system of Afrotropical Onthophagini is now under
89 scrutiny.

90 Within this framework, we focused on the 21st group, that includes only three small
91 species, recorded from the Southern Africa subregion: *Onthophagus caffrarius*
92 d'Orbigny, 1902, *O. quadriceps* Harold, 1867 and *O. signatus* Fåhræus, 1857. The
93 species-group was defined by a set of characters related to external morphology, that are

94 not exclusive to this group (d'Orbigny 1913), as the base of pygidium with a transversal
95 carina, or the pronotum covered by granules or granulate points which can both be
96 found in the majority of *Onthophagus* groups (d'Orbigny 1913).

97 The question about the ambiguous taxonomic position of the 21st group has been
98 recently raised in the context of studies dealing with the review of phylogenetic
99 relationships within Scarabaeinae by the use of a biomolecular approach. In their
100 phylogenetic review of the Madagascar dung beetles Wirta *et al.* (2008) placed *O.*
101 *signatus* (a species of 21st d'Orbigny group) very close to *Phalops wittei* (Harold, 1867)
102 and *Digitonthophagus gazella* (Fabricius, 1787), all these species being however well-
103 separated by both Oniticellini and other Onthophagini. The latter was thus regarded as
104 not monophyletic, with at least two distinct clades recognized within this tribe. In
105 addition, Monaghan *et al.* (2007) and, more recently, Mlambo *et al.* (2015) showed that
106 the clade *Digitonthophagus* and *Phalops* Erichson, 1848 are sister to all the other
107 Onthophagini, although neither of them included the species of the 21st d'Orbigny group
108 in the analysis. Based on this research, it was hypothesized that *Phalops* and
109 *Digitonthophagus* constitute a separate clade from the other Onthophagini previously
110 examined, and were closely related. However, the taxonomic position of the 21st
111 species-group of *Onthophagus* was not verified in those studies.

112 The uncertain taxonomic position of *Phalops* and *Digitonthophagus* within
113 Onthophagini was also highlighted by studies in which various morphological
114 characters were analyzed and discussed. The male genitalia (formed by the aedeagus
115 and endophallus) have been recently examined in various Onthophagini groups
116 (Tarasov and Solodovnikov 2011; Medina *et al.* 2013; Tarasov and Génier 2015),
117 giving remarkable results especially in defining the endophallus sclerites, although the
118 homologies of *Digitonthophagus* and *Phalops* were not fully defined (see the online
119 Supplementary Material for further details). Other internal morphological structures that
120 have not been employed till now (for instance the female genitalia and the epipharynx)
121 could bear phylogenetic signals, and surely deserve a careful examination, to determine
122 their usefulness to solve major taxonomic and phylogenetic problems within the
123 Onthophagini.
124

125 The aim of the present paper was to evaluate the taxonomic position of the species of
 126 the *Onthophagus* 21st group within Onthophagini and verify the suggested hypothesis of
 127 its close relationships to *Phalops* and *Digitonthophagus*, according to former findings.
 128 The present research employed both molecular (COI sequences) and morphological
 129 (external and internal anatomical traits) approaches, focusing also on the recognition of
 130 novel structures useful in the assessment of the phylogenetic relationships among these
 131 taxa.

132

133

134

135 **Material and Methods**

136 A diversified approach was chosen to evaluate the hypothesis that the species included
 137 in the *Onthophagus* 21st group constituted a monophyletic and separate taxon, more
 138 closely related to *Phalops* and *Digitonthophagus* than to the other *Onthophagus* taxa.

139 The results obtained from the different methods (i.e., biomolecular taxonomic distance
 140 analysis, morphological phylogeny and geometric morphometrics analysis) were then
 141 compared.

142 A dataset was established that included *Phalops*, *Digitonthophagus*, *Onthophagus* 21st
 143 species-group, and some other representatives of *Onthophagus* from Afrotropical and
 144 Palearctic regions. The Oriental species *Serrophorus seniculus* (Fabricius, 1781),
 145 belonging to the *Proagoderus* complex (Tarasov and Kabakov 2010) was chosen as the
 146 outgroup taxon in the phylogenetic analyses.

147 In detail, the following species were examined: *Digitonthophagus bonasus* (Fabricius,
 148 1775); *D. gazella* (Fabricius, 1787); *Euonthophagus flavimargo* (d'Orbigny, 1902);
 149 *Onthophagus caffrarius* d'Orbigny, 1902; *O. quadraticeps* Harold, 1867; *O. signatus*
 150 Fåhraeus, 1857; *O. nigriventris* d'Orbigny, 1902; *O. (Onthophagus) illyricus* (Scopoli,
 151 1763); *O. (Palaeonthophagus) coenobita* (Herbst, 1783); *O. (Palaeonthophagus)*
 152 *medius* (Kugelann, 1792); *O. (Palaeonthophagus) nuchicornis* (Linnaeus, 1758); *O.*
 153 *(Palaeonthophagus) ovatus* (Linnaeus, 1767); *O. interstitialis* (Fåhraeus, 1857); *O.*
 154 *bituberculatus* (Olivier, 1789); *O. depressus* Harold, 1871; *Phalops ardea* (Klug, 1855);
 155 *P. boschas* (Klug, 1855); *P. prasinus* (Erichson, 1843); *P. rufosignatus* van Lansberge,
 156 1885; *P. wittei* (Harold, 1867).

157

158

159 ***Molecular analysis***

160 The molecular analysis focused on mitochondrial cytochrome oxidase I (COI), a
161 powerful tool for characterizing taxa (Hebert et al. 2003, 2004; King et al. 2008; Dincă
162 et al. 2013) commonly employed for species identification at a molecular level, and the
163 core of an integrated taxonomic system (i.e., the DNA barcoding, see Casiraghi et al.
164 2010; Dincă et al. 2015; Vodă et al. 2015). COI sequences of various Onthophagini
165 species collected from GenBank were employed to provide a dataset comprising 21
166 sequences from 14 species (see Table 1 for the list of species employed in the analysis,
167 their acronyms and accession codes).

168 Multiple sequence alignment was performed using the MUSCLE method as
169 implemented in MEGA v6 (Tamura et al. 2013), then the alignment of sequences was
170 checked manually. All positions containing gaps and missing data were eliminated
171 during the subsequent analyses, that were made using MEGA v6, except when
172 otherwise stated.

173 To test the genetic divergence among these taxa, a distance matrix was calculated
174 employing the Kimura 2 parameter (K2P) correction, claimed as the best DNA
175 substitution model for low genetic distances (Nei and Kumar 2000; Casiraghi et al.
176 2010), and commonly used to evaluate the barcode gap among taxa. Standard error
177 estimates were obtained by the bootstrap procedure (Nreps = 1,000). The threshold
178 value between intra and interspecific distances (i.e., the barcode gap) was established at
179 1%, which is commonly used as the level of separation in most previous studies of
180 animals (Ratnasingham and Hebert 2007, 2013; Chevasco et al. 2014; Del Latte et al.
181 2015).

182 Phylogenetic reconstruction *via* Nearest-Neighbor-Interchange (NNI) was applied to
183 generate an automatically-computed NJ tree using the Tamura-Nei (TN93) parameter
184 substitution model (Nei and Kumar 2000) with all positions containing gaps and
185 missing data eliminated from the dataset (complete deletion option). This initial tree
186 was set as default for phylogenetic reconstruction *via* the Maximum Likelihood (ML)
187 method coupled with bootstrapping reliability tests (Nreps = 1,000). Support for
188 internodes was assessed by bootstrap percentages.

189 The branch supports were evaluated by both approximate likelihood ratio test (SH-like
190 aLRT) and non-parametric bootstrap (Nreps = 1,000) methods (Simmons 2014), as
191 implemented in PhyML 3.1 (Guindon and Gascuel 2003; Guindon et al. 2010), applying
192 the same settings of the former ML analysis (single initial BioNJ tree; TN93nucleotide
193 substitution model; no discrete gamma model; equilibrium frequencies optimised; NNI
194 tree topology search).

195 To test the monophyly of clades, the MUSCLE-aligned matrix was analyzed by
196 phylogenetic networks analysis (PNA) as implemented in SplitsTree 4.14.2 (Huson and
197 Bryant 2006). Constant (N = 166), gapped (N = 286) and non-parsimony informative (N
198 = 336) sites were excluded from the analysis. Monophyly of the lineages was assessed
199 by the Neighbor-Net (splitstransform = EqualAngle) method (Bryant and Moulton
200 2004), whereas bootstrapping estimates (1,000 runs) were employed to support the
201 splits.

202

203

204 ***Morphological analysis***

205 More than 1,500 specimens were examined to determine morphological characters that
206 support inter and intraspecific differences among the Onthophagini taxa, with a special
207 focus on the *Onthophagus* group 21 species and related groups.

208 The material examined was loaned from the following Museum collections:

209 MHNL – Musée des Confluences, Lyon, France

210 NMEG – Naturkundemuseum, Erfurt, Germany

211 MNHN – Muséum National d'Histoire Naturelle, Paris, France

212 We also examined material from private collections of E. Barbero (EBCT - Torino,
213 Italy), and P. Moretto (PMCT - Toulon, France).

214

215 Various external and internal morphological traits were carefully examined, according
216 to the suggestions of the most recent literature (Tarasov and Solodovnikov 2011;
217 Tarasov and Génier 2015) that emphasized the necessity to find novel morphological
218 characters to elucidate phylogenetic relationships within the Scarabaeoidea.

219 The mouthparts and genitalia of both sexes were dissected and treated following the
220 methods usually employed to prepare slides (Barbero *et al.* 2003). The images of the

221 internal and external structures were then captured using a Leica® DMC4500 digital
222 camera connected to a stereoscopic dissecting scope (Leica® Z16Apo).

223 The nomenclature of the anatomical traits adopted in this study follows those used in
224 Tarasov and Solodovnikov (2011), Tarasov and Génier (2015) and Roggero *et al.*
225 (2015).

226 The datasets obtained by observation of the various structures have been employed to
227 carry out two different analyses, a morphological phylogeny and a geometric
228 morphometric analysis.

229

230 Among the various structures examined, some were selected to build the matrix for the
231 subsequent phylogenetic analysis (see the characters list below), although others were
232 discarded. In particular, the antenna was not used in the present analysis since it proved
233 to be very complicated structurally and difficult to interpret. Although the cavity
234 identified by Tarasov and Solodovnikov (2011) can be easily detected on the 12th and
235 13th antennal segments (Fig. 1A-C) of the species studied here, it is apparently
236 extremely variable and can appear as either a more or less concave or convex area. The
237 shape of this area is not constant even in the same species (Fig. 1D-E). Although the
238 antennal cavity is an extremely interesting structure, its functions have to be studied
239 further in detail.

240 Male genitalia are currently employed in the systematics of Onthophagini, but their
241 features remain to be fully elucidated. They are constituted by an aedeagus and an
242 inflatable endophallus which extends into the female bursa copulatrix during copulation
243 (House and Simmons 2003). On the inside membrane of the endophallus there are
244 various sclerites, that were recently examined and named by Tarasov and Solodovnikov
245 2011 (see online Supplementary Material for further details).

246 Unlike the male genitalia, widely employed in insect systematics for many years, the
247 female genitalia are much less studied, despite the hypothesized co-evolution among
248 these structures. As pointed out in evolutionary biology studies, male and female
249 genitalia are subject to a stabilizing selection to enforce mate recognition and
250 reproductive isolation at a specific level (Eberhard 1992; Gilligan and Wenzel 2008;
251 Mikkola 2008; Masly 2012; Wojcieszek and Simmons 2013). As female genitalia must
252 co-evolve in concert with those of males to allow coupling, phylogenetic signals of

253 genitalia must follow the same trend in both sexes (Simmons and Garcia-Gonzales
254 2011). The female genitalia in Onthophagini are structurally relatively simple. They
255 consist of a membranous sac-like vagina, carrying a more or less sclerotized support
256 area (the infundibular wall, variously shaped), and a receptaculum seminis for the
257 storage of sperm, connected to the vagina by the infundibular tube (House and Simmons
258 2005; Pizzo et al. 2006, 2008).

259 The epipharynx constitutes the upper part of the mouth, with the function of food
260 filtration. It is an extremely complex structure formed by a membranous part and a
261 sclerotized part with a support role. Due to extreme diversification of features, the
262 epipharynx has proved a very useful tool to generate separation of groups at different
263 taxonomic levels, giving often highly meaningful results as regards phylogenetic signals
264 (Barbero et al. 2003; Roggero et al. 2015).

265

266

267 *Phylogenetic analysis*

268 The selected structures (i.e., head, pronotum, elytra, legs, mentum, epipharynx, and
269 genitalia of both sexes) were employed to build a matrix of 35 binary and multistate
270 characters (Table 2, and see the online Supplementary Material for a detailed discussion
271 of the endophallus sclerites).

272 The character list can be found in the Supplementary Material.

273

274 The matrix of 35 morphological characters (set as unordered and equally weighted) was
275 analyzed by Maximum Parsimony Analysis (heuristic search) in PAUP 4.0b.10
276 (Swofford 2002) using the software default settings (stepwise addition with simple
277 addition sequence, tree bisection – reconnection branch swapping, ACCTRAN
278 character-state optimization). The multistate characters were interpreted as
279 “uncertainty”, and the gaps treated as “missing”. The MaxTrees limit was set to
280 automatically increase from the initial setting. Trees were rooted by the outgroup
281 method, and the strict consensus was calculated. After the first run, the characters were
282 reweighted by the rescaled consistency index (successive weighting) and heuristic
283 searches were performed until the character weights no longer changed and trees with
284 identical length were found in three consecutive searches (stability in the trees). The

285 Newick output trees obtained in the former analysis were visualized with FigTree v1.4.2
286 (Rambaut 2014).

287 Statistical support for each branch was assessed by PAUP using the non-parametric
288 bootstrap method (Felsenstein 1985), with the same heuristic search settings as above,
289 and 100,000 replications.

290 The morphological dataset was also analyzed using TNT (Goloboff et al. 2003, 2008).
291 Both Implicit Enumeration and Traditional Search options were employed using the
292 default settings with the Implied Weighting set to ON. The synapomorphies common to
293 all trees were mapped onto the resulting trees. Tree statistics were calculated using a
294 TNT script (stats.run). Relative support values were calculated within TNT by
295 symmetric resampling, bootstrap standard and jackknife with 1,000 iterations (Sharkey
296 et al. 2012).

297 The Bayesian inference of phylogeny (Markov chain Monte Carlo simulations, or
298 MCMC) was used to approximate the posterior probabilities of trees and parameters, as
299 implemented in MrBayes v3.2 (Huelsenbeck et al. 2001; Ronquist and Huelsenbeck
300 2003; Ronquist et al. 2011). The analysis was initiated with a random starting tree and
301 run for 2,500,000 generations (two runs, eight chains), sampling trees every 100
302 generations, with rate heterogeneity modelled by an equal distribution. Posterior clade
303 probabilities were used to assess nodal support. The trees sampled during the burn-in
304 phase (i.e. before the chain had reached its apparent target distribution) were discarded
305 (25% of the total). The remaining trees were summarized in the Bayesian Majority Rule
306 consensus trees, and the topologies of the two runs were compared to detect differences.
307 For the graphic exploration of MCMC convergence in Bayesian phylogeny, TRACER
308 v1.6 (Rambaut et al. 2013) was then employed to analyze the results obtained from
309 Bayesian MCMC runs. Trends that might suggest problems with MCMC convergence
310 were checked and the lnL probability plot was examined for stationarity.

311 The consensus tree obtained in the former analysis was visualized with FigTree v1.4.2
312 (Rambaut 2014).

313 The distances between the taxa and the monophyly of clades were analyzed by
314 phylogenetic networks analysis (PNA) as implemented in Splits Tree 4.14.2 (Huson and
315 Bryant 2006). The monophyly of the lineages was assessed with the Neighbor-Net

316 (splitstransform = EqualAngle) method (Bryant and Moulton 2004), and the
317 bootstrapping estimate (1,000 runs) was employed to support divisions.

318

319 *Geometric morphometrics analysis*

320 The geometric morphometrics semilandmark method was applied to capture the overall
321 shape variation of the epipharynx (or labrum) since this structure can provide a detailed
322 survey of the more complicated relationships among the taxa (Tocco et al. 2011;
323 Roggero et al. 2015). On the basis of the former biomolecular and morphological
324 analyses (see above), two main issues were identified. One comprised the overall
325 epipharynx shape variation within the whole dataset to assess the reciprocal
326 relationships among all the taxa. The other comprised a more precise characterization of
327 the shape variation patterns that distinguish *Phalops*, *Digitonthophagus* and
328 *Onthophagus* 21st group.

329

330 The configuration of points (Fig. 2) was chosen to capture the overall shape variation of
331 the epipharynx, and was sampled using tpsDig2 v2.20 (Rohlf 2015a) and tpsUtil v1.64
332 (Rohlf 2015b). The same points configuration was employed to examine the patterns of
333 shape variation in both datasets (see above) applying the same protocol. This comprised
334 Principal component analysis (a.k.a., Relative warps analysis), Canonical variate
335 analysis and Multivariate tests of significance (Roggero et al. 2013).

336 Reciprocal relationships among the species were evaluated for both datasets ($N_1 = 84$
337 and $N_2 = 62$) using tpsSmall v1.33 (Rohlf 2015c) and tpsRelw v1.54 (Rohlf 2015d).
338 Relative warp values (RWs) and the aligned configurations (AL) were retained for
339 further analyses.

340 Canonical Variates analysis (CVA) on the RWs values was employed to test the
341 proposed taxa classifications as implemented in IBM[®] SPSS[®] Statistics v22 (IBM Corp.
342 2013). This procedure applied the Mahalanobis distance method and the leave-one-out
343 option on the whole dataset of the RWs values to account for 100% of the overall shape
344 variation.

345 The goodness of group assignments was examined by tpsRegr v1.42 (Rohlf 2015e),
346 employing the aligned configurations gained from the PCA (see above) to test the
347 proposed classifications through a taxa comparison. For the analysis, a design matrix

348 was chosen (Rohlf 2015e) to represent the current experimental design for the study of
349 specimens classification. The significance of the classification was tested by
350 Permutation tests (N reps=100,000) as implemented in tpsRegr.

351

352

353

354 **Results**

355 ***Taxonomic revision***

356 The species formerly included in *Onthophagus* 21st group are separated as a new genus,
357 *Kurtops* **gen.n.**, that was included in the *Phalops* complex (see online Supplementary
358 Material for further details)

359

360 ***Kurtops* Roggero, Barbero and Palestrini gen.n.**

361 (Figs 3, 4, 5 and 6)

362 *Type species.* *Onthophagus signatus* Fåhraeus, 1857: 304.

363 *Included species.* At present, the three species that formerly constituted the
364 *Onthophagus* 21st group (Fåhraeus 1857; Harold 1867; d'Orbigny 1902, 1913) are
365 included in the new genus. A detailed description of the species included in the genus
366 can be found in the online Supplementary Material.

367

368 *Description.* Length 0.50-1.00 cm. Head squared, without horns or laminar extensions,
369 covered by a thick, whitish pubescence; rounded and slightly protruding genae; small
370 superior portion of eyes. Pronotum covered by thick rasping points, with a long, light
371 yellow pubescence thicker on sides. Marked elytral striae, with points as large as the
372 striae. Pygidium with deep, irregular points, and slightly larger in males. Legs
373 characterized by testaceous femurs, and darker tibiae; fore tibia stouter in males than in
374 females, with an evident tooth only in males.

375 Epipharynx (Fig. 3). Fore margin only slightly notched, sickle-shaped in *K. caffrarius*
376 and *K. quadriceps*, more squared in *K. signatus*; corypha constituted by a well-
377 developed tuft of setae; the triangular sclerotized area below the haptomerum almost
378 reaching the coripha, narrow at base in *K. signatus*, and larger in *K. quadriceps* and *K.*
379 *caffrarius*; apotormae always present, more or less developed; hollow area below the

380 haptolachus (i.e., the plegmatic area) narrowed (*K. quadraticeps*) or inapparent (*K.*
381 *caffrarius* and *K. signatus*); reduced and thick pternotormae; very short and rounded
382 laeotorma and the dextiotorma. On the whole, the epipharynx features of *Kurtops* are
383 well-differentiated from those of *Digitonthophagus* and *Phalops* (Fig. 7).

384 Male genitalia (Figs. 4D-F, 5). Aedeagus parameres rounded and slightly tapering at
385 apex, with a well-developed inward expansion (triangular in *K. signatus*, and beak-
386 shaped in *K. quadraticeps* and *K. caffrarius*). Phallobase twice as long as the parameres,
387 slightly inward curved. Well-differentiated endophallus sclerites, but lamella copulatrix
388 absent; accessory lamellae well developed, sharing a similar pattern to
389 *Digitonthophagus* and *Phalops* ones (Fig. 8); FLP always well-developed, the apical
390 part expanded, rounded and less sclerotized, carrying many small teeth, and the basal
391 part expanded into a lamina more or less developed, but always well sclerotized; FLP
392 carrying also a lateral part (here named EC) triangular shaped and well-developed;
393 conspicuous BSC sclerite near the base of the FLP sclerite; C-shaped and tightly
394 connected A and SA sclerites positioned laterally to FLP; SRP sclerite present, more or
395 less developed.

396 Female genitalia (Fig. 6). The females are known only for *K. quadraticeps* and *K.*
397 *signatus*, that show a similar pattern, analogous to that already seen in *Phalops* and
398 *Digitonthophagus* (Fig. 9). Moderately sclerotized infundibular wall, triangular-shaped
399 in *K. quadraticeps*, and more clearly mushroom-shaped in *K. signatus*. Receptaculum
400 seminis well sclerotized, slender, elongate, tapering to the sharp apex, with the
401 glandular tube opening very near the point of insertion of the infundibular tube.

402 *Etymology.* The new genus was named after the characteristically rounded pronotum,
403 employing the Greek word *kurtos* that means convex.

404 *Distribution.* The genus is known from the whole Southern African subregion (Fig. 10).

405 *Remarks.* According to the results of biomolecular and morphological analyses, these
406 species constitute a distinct monophyletic taxon that is closely related to
407 *Digitonthophagus* and *Phalops*. They were thus removed from *Onthophagus* and raised
408 to generic level. Although these three species show similar features, they can be easily
409 identified from each other. *Kurtops caffrarius* differs greatly from *K. signatus* on the
410 basis of the size and general appearance. It differs from *K. quadraticeps* essentially by
411 the pronotum, that is evenly covered by granulate small points in *K. caffrarius*, and with

412 granulate larger points which are smaller only on hind central half in *K. quadraticeps*.
 413 The rasping points and the simple points are mixed in the *K. signatus* pronotum. The
 414 yellowish ochreous elytra in *K. quadraticeps* and *K. signatus* carry darker patches,
 415 while they are evenly ochreous in *K. cafferarius*.
 416 The epipharynx (Fig. 3) fore margin is rounded in *K. quadraticeps* and *K. cafferarius*,
 417 squared in *K. signatus*; the proplegmatium is narrow in *K. signatus*, but thicker in the
 418 two other species; the apotormae are linear shaped in *K. signatus*, more developed and
 419 almost reaching the proplegmatium in *K. cafferarius*, while in *K. quadraticeps* lengthens
 420 beyond the proplegmatium line.
 421 In males the parameres apices (Fig. 4) are triangular-shaped in *K. signatus*, hook-shaped
 422 in *K. quadraticeps* and *K. cafferarius*, although they are far more developed in the latter
 423 species; the endophallus lamellae are very differently shaped in the three species (Fig.
 424 5).
 425 In females (Fig. 6) the infundibular wall in *K. signatus* and *K. quadraticeps* is very
 426 differently shaped, in accordance with what has already been seen in *Phalops* and
 427 *Digitonthopagus* (Barbero et al. 2003).

428

429

430 ***Molecular analysis***

431 The pairwise distance matrix is shown in Table 3 (Supplementary Material). Distances
 432 were mostly >0.1 except for *O. ovatus/O. coenobita*, *O. nuchicornis/O. medius* and *O.*
 433 *ovatus/O. nuchicornis* that had a distance value <0.1. These lower distance values were
 434 found only within some Palearctic *Onthopagus*, and are likely due to recent speciation
 435 events. Two major groups were clearly identified. In one, pairwise distance values were
 436 always > 0.6-0.8, corresponding to a group comprising only *Onthopagus* species. The
 437 second group comprises *Onthopagus interstitialis* and other genera.
 438 The ML trees showed two major clades. One comprised *Phalops* + *Digitonthopagus* +
 439 *Kurtops*. The second was divided into two further clades. One includes the
 440 *Onthopagus s.l.*+*O. interstitialis* species while the other comprised *Euonthopagus*
 441 *flavimargo* + *Onthopagus depressus*. Small differences were shown among the species
 442 within each clade, but the support values were homogeneous in all the computed trees.
 443 Both SH-like aLRT and bootstrap gave congruent support values for the major clades.

444 High bootstrap (100%) and SH-like aLRT (1) values were shown for separation of the
445 *Onthophagus* clade in the ML tree (TN93 BIC = 8793.309, Fig. 11), although the
446 support values were frequently lower within the clade. This result was expected since
447 only a fraction of the many *Onthophagus* species were considered in the present
448 research, thus the intrageneric relationships surely could not be fully elucidated. The
449 position of *O. interstitialis*, *O. depressus* and *E. flavimargo* could not be resolved,
450 although the results showed closer relationships to *Onthophagus s.l.* than to the *Phalops*
451 + *Digitonthophagus* + *Kurtops* clade (the latter one showing bootstrap = 27%, but SH-
452 like aLRT = 0.775). Within the last clade, the support values were high for
453 *Digitonthophagus* and *Kurtops* **gen.n.**, but for *Phalops* the intrageneric relationships
454 were not fully supported. The particularly low value shown for *Phalops* may depend on
455 the fact that only two out of the 38 known species have been used in the analysis, and
456 the two species belong to two distinct clades within *Phalops*.
457 The tree generated by phylogenetic networks analysis (see online Supplementary
458 Material) showed significant recomputed fit values (fit = 98.744, LS fit 99.983 and
459 stress = 0.013). Significant bootstrap values of 100% were shown for the two major
460 clades and all included species groups (see online Supplementary Material).

461

462

463 ***Morphological analysis***

464 *Phylogenetic analysis*

465 The first heuristic search performed on the matrix of unordered and equal weight
466 characters (Table 2) generated six trees (length = 111, CI = 0.594, HI = 0.405, RI =
467 0.750, RC = 0.445, not shown here). Successive weighting analysis was then applied to
468 generate a single tree (Fig. 12A, length = 49.130, CI = 0.775, HI = 0.224, RI = 0.887,
469 and RC = 0.687) where two major clades were identified. In the first clade, two groups
470 were distinguished, one including *Onthophagus bituberculatus* and *O. depressus*, the
471 other comprising *Phalops*, *Digitonthophagus* and *Kurtops* **gen.n.** In the second major
472 clade all the other species were included.

473 Implicit Enumeration and the Traditional Search (with Implied Weighting set to ON) as
474 implemented in TNT gave analogous results. By both methods a single tree (length =
475 115, CI = 0.595, RI = 0.750) was produced, that was identical to the one from maximum

476 parsimony analysis in PAUP. The standard bootstrap, jackknife and symmetric
477 resampling methods generated congruent support values at a generic level, with the
478 average group support equal to 48.1, 51.5 and 51.7 respectively. The support statistics
479 from TNT were congruent to the ones from the Bootstrap in PAUP (see Fig. 12A).
480 The majority rule 50% consensus tree (Fig. 12B) produced by the Bayesian Inference
481 method was not fully resolved. While the genera were well-defined, having a good
482 credibility value, the reciprocal relationships among the genera were not clearly
483 established, and the nodes were collapsed. The chain swap information for the two runs
484 generated equal results for proportion of successful state exchanges between chains.
485 TRACER confirmed the correctness of the Bayesian Inference by the analysis of the
486 statistics of the two runs.

487 The resulting network splits tree (Fig. 12C) from the Phylogenetic Networks analysis
488 (NeighborNet Equal Angle algorithm) had a recomputed fit = 95.18, and LS fit = 99.62.
489 The Resampling by the bootstrap method confirmed the proposed groups, as already
490 shown in the former analyses. The support values of the genera were marked onto the
491 tree (Fig. 12C). The close relationships among *Phalops*, *Digitonthophagus* and *Kurtops*
492 **gen.n.** were assessed, as well as those within the *Onthophagus* species. *Euonthophagus*
493 *flavimargo* is isolated from the other species, and not related to the *Onthophagus*
494 species (see Moretto 2009 for further details). Also *Onthophagus bituberculatus* and *O.*
495 *depressus* constituted a distinct clade secluded from the others, and these species are
496 currently under review based on the results obtained by this research.

497

498 *Geometric morphometrics analysis*

499 In the analysis on the whole dataset of Onthophagini, the correlation value of the
500 tangent distances against the Procrustes distances obtained by tpsSmall was 1.000, thus
501 the amount of variation in shape in the present dataset was small enough to permit the
502 subsequent GM analysis.

503 In the principal component analysis (PCA, as implemented in tpsRelw), forty out of the
504 forty-six obtained RWs were enough to account for 100% of the overall shape variation,
505 thus the last six RWs were discarded from the following analysis. Each of the first four
506 RWs gave a percent value of explained variance greater than 5%. These RWs accounted

507 together for almost 75% of the overall shape variation, being approximately 50% of the
508 overall shape variation represented by the two first RWs (plots not shown here).
509 The deformation grids of the RWs 1-4 axes (Fig. 13) were examined separately, and
510 marked differences were displayed. In RW_1 the main changes involved the fore
511 margin, that can be more or less notched, the width of the proplegmatium, the length of
512 the triangular sclerotized medial area below the haptomerum, and the more or less
513 accentuate curvature of the chaetopariae. RW_2 represents variation in the fore margin
514 together with marked differences in development of the crepis. RW_3 accounted mainly
515 for the shape variation of the hollow area which is located at the base of the anterior
516 epitorma, and can be more or less expanded. Variations of the fore margin, and length
517 of the medial sclerotized area were summarized by RW_4.
518 Due to the great number of RWs obtained from the PCA, these variables cannot be (as
519 usual) examined in pairs by means of graphics to furnish a full representation of the
520 overall shape variation. The taxa classification was tested for all the variables that gave
521 together 100% of explained variance (i.e., forty RWs) using CVA.
522 CVA analysis of variation in shape of the epipharynx defined four well-separated
523 groups (Fig. 14A) that were consistent with taxonomic classification (Fig. 12). High
524 goodness of fit was confirmed by cross validation (98.8%, Table 4, Supplementary
525 Material). Figure 14A shows that the species of *Onthophagus* group 21 are more closely
526 related to *Digitonthophagus* and *Phalops* than to *Onthophagus s.l.* Figure 14B shows
527 that group 21 is, nevertheless, separate from *Digitonthophagus* and *Phalops* thus
528 justifying its status as the new genus *Kurtops*.
529 In the tpsRegr analysis, the Multivariate tests of significance gave significant values
530 (Hotelling-Lawley trace = 25.469, $F_{(184, 130.0)} = 4.499$, $p < 0.0001$). The Generalized
531 Goodall F-test also gave a significant result ($F = 11.1477$, $df = 184, 3634$, and $p =$
532 0.0000). The results of the Permutation tests, based on 100.000 replications, are in
533 agreement with the former findings (see above), being the percent of Goodall F values \geq
534 observed equal to the significant value of 0.001% (small percentages imply
535 significance).
536

537 Also for the second analysis, the amount of variation in shape obtained by tpsSmall was
538 small enough (1.000) to permit the subsequent GM analysis of the *Phalops* complex
539 dataset.

540 From the principal component analysis (PCA), forty out of the forty-six obtained RWs
541 accounted for 100% of the overall shape variation, thus the last six RWs were discarded
542 from the following analysis. About 54% of the overall shape variation was represented
543 by the two first RWs, and each of the first four RWs gave a percent value of explained
544 variance greater than 5%, accounting together for almost 72% of the overall shape
545 variation. The three genera are clearly distinguishable in the scatterplot of RW 1 and 2
546 (the plots of the RWs in pairs are not showed here).

547 The CVA testing the taxa classification at generic level (Table 5, Supplementary
548 Material) gave 100.0% of cases correctly classified for *Phalops*, *Digitonthophagus* and
549 *Kurtops*, and 98.4% after the cross validation. In the CV 1 and 2 plot (Fig. 14B), the
550 three genera were well-differentiated, *Digitonthophagus* and *Kurtops* **gen.n.** seemingly
551 being more closely related among themselves than to *Phalops*.

552 The Multivariate tests of significance by the tpsRegr analysis gave a significant value of
553 the Hotelling-Lawley trace (60.374 , $F_{(184, 42.0)} = 3.445$, $p < 0.0001$). The Generalized
554 Goodall F-test gave a significant result ($F = 6.6993$, $df = 184, 2622$, and $p = 0.0000$).
555 Also, the results of the Permutation tests based on 100,000 replications were significant,
556 with the percent of Goodall F values \geq observed equal to the significant value of
557 0.001%.

558

559

560

561 **Discussion**

562 The study was aimed mainly at evaluating the taxonomic position of the 21st
563 *Onthophagus* species-group within the Onthophagini. The present findings indicate that
564 the group does not belong in *Onthophagus s.l.*, and must be raised to generic rank as
565 *Kurtops* **gen.n.** Furthermore, it was confirmed that *Onthophagus* as currently defined is
566 not a monophyletic taxon, which concurs with recent findings (Monaghan et al. 2007;
567 Wirta et al. 2008; Mlambo et al. 2015).

568 When looking at the results of both biomolecular and morphological analyses of
569 *Kurtops* **gen.n.**, *Phalops* and *Digitonthophagus*, there was a homogenous pattern that
570 was not evident in the *Onthophagus s.l.* species, thus excluding any relationship between
571 the former three genera and the latter genus. Therefore, it was hypothesized that the
572 three genera might constitute a distinct taxonomic group separate from the other
573 Onthophagini.

574 Herein, we recommend to include *Kurtops* **gen.n.**, *Phalops* and *Digitonthophagus* into a
575 *Phalops* complex of genera distinct from *Onthophagus* in order to further mark its
576 separation from the other Onthophagini, as was previously suggested for the
577 *Serrophorus* complex (Tarasov and Kabakov 2010; Tarasov and Solodovnikov 2011),
578 until the systematic position of all the taxa currently within this tribe (especially, the
579 *Onthophagus*) can be fully elucidated (see online Supplementary Material for further
580 details).

581 High pairwise distance values from the COI sequence identified two main distinct
582 groups, one including the *Onthophagus* species and the other comprising the *Phalops*
583 complex together with *Euonthophagus flavimargo*, *Onthophagus depressus* and *O.*
584 *interstitialis*. An ancient separation was accounted for in the taxa from the Afrotropical
585 Region, whilst the Palearctic *Onthophagus* species showed lower pairwise values, thus
586 indicating a more recent speciation than the Afrotropical taxa. The seclusion of
587 *Onthophagus s.l.* was also confirmed by other biomolecular analyses (ML and PNA). It
588 is noteworthy that the *Phalops* complex constituted a distinct clade from all the other
589 taxa, in both trees. Furthermore, *O. interstitialis* was never linked to the *Onthophagus*
590 species, confirming it as a separate clade whose taxonomic status must surely be
591 reviewed.

592 Consistent results were obtained from the morphological phylogenetic analyses,
593 confirming the presence of two distinct clades for the *Onthophagus s.l.* and the *Phalops*
594 complex, although ostensibly also *Euonthophagus flavimargo* and *Onthophagus*
595 *bituberculatus* + *O. depressus* were identified as distinct clades. The hypothesis of a far
596 greater taxonomic complexity than is currently believed within the Onthophagini was
597 thus corroborated.

598 The highlighted differentiation of these taxa was also confirmed by the geometric
599 morphometrics analysis, in which the epipharynx was adequate by itself to identify the

600 same four groups already classified by the phylogenetic analyses founded on both
601 morphological and (partly) biomolecular data.

602 To summarize the results, it was found that *Digitonthophagus*, *Phalops* and *Kurtops*
603 **gen.n.** are both closely related, and are characterized by extremely differentiated
604 external features, quite different epipharynx (Figs. 3 and 7) and markedly similar
605 genitalia (Figs. 4-6 and 8-9) patterns (See below for a thorough review of the *Phalops*
606 complex, with an in-depth discussion of the epipharyngeal and genitalic features).

607

608 The combination of biomolecular and morphological analyses has definitely contributed
609 in solving the question of the taxonomic position of the three species formerly included
610 in d'Orbigny 21st group, confirming again that *Onthophagus s.l.* is not a monophyletic
611 taxon. Past and present results clearly indicate the need for an urgent review of the
612 classification of each group currently included in this genus, both to define in detail the
613 phylogenetic relationships among these Afrotropical taxa, and to increase the systematic
614 delineation of the whole Onthophagini tribe.

615

616

617

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629

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631

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- 818
- 819

820 **Tables**

821

822 **Table 1.** List of the COI sequences with the GENBANK accession number.

823

species	GenBank accession	distribution	acronym
<i>Digitonthophagus gazella</i> (Fabricius, 1787)	EF188213.1	Worldwide	GAZ_1
<i>Digitonthophagus gazella</i> (Fabricius, 1787)	EF188212.1	Worldwide	GAZ_2
<i>Euonthophagus flavimargo</i> (d'Orbigny, 1902)	EF188209.1	Afrotropical	FLA_1
<i>Euonthophagus flavimargo</i> (d'Orbigny, 1902)	EF188210.1	Afrotropical	FLA_2
<i>Onthophagus depressus</i> (Harold, 1871)	EF188207.1	Afrotropical	DEP
<i>Onthophagus coenobita</i> (Herbst, 1783)	KM445555	Palaearctic	COE
<i>Onthophagus illyricus</i> (Scopoli, 1763)	HQ954129	Palaearctic	ILL_1
<i>Onthophagus illyricus</i> (Scopoli, 1763)	KM450900	Palaearctic	ILL_2
<i>Onthophagus interstitialis</i> (Fahraeus, 1857)	JN804624.1	Afrotropical	INT_1
<i>Onthophagus interstitialis</i> (Fahraeus, 1857)	JN804625.1	Afrotropical	INT_2
<i>Onthophagus medius</i> (Kugelann, 1792)	KM447997	Palaearctic	MED
<i>Onthophagus nigriventris</i> d'Orbigny, 1905	EU162459.1	Afrotropical	NIG
<i>Onthophagus nuchicornis</i> (Linnaeus, 1758)	HQ954131	Palaearctic	NUC
<i>Onthophagus ovatus</i> (Linnaeus, 1767)	HQ954130	Palaearctic	OVA
<i>Onthophagus signatus</i> (Fahraeus, 1857)	EF188216.1	Afrotropical	SIG_1
<i>Onthophagus signatus</i> (Fahraeus, 1857)	EF188215.1	Afrotropical	SIG_2
<i>Phalops ardea</i> (Klug, 1855)	AY131935.1	Afrotropical	ARD
<i>Phalops rufosignatus</i> Lansberge, 1885	JN804662.1	Afrotropical	RUF_1
<i>Phalops rufosignatus</i> Lansberge, 1885	JN804660.1	Afrotropical	RUF_2
<i>Phalops rufosignatus</i> Lansberge, 1885	JN804661.1	Afrotropical	RUF_3
<i>Serrophorus seniculus</i> (Fabricius, 1781)	EF188225.1	Oriental	SEN

824

825

826

827 **Table 2.** Matrix of the 35 morphological characters used in the phylogenetic analysis.

species	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18
<i>S. seniculus</i>	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
<i>D. gazella</i>	0	1	0	1	1	0	0	2	0	0	0	0	0	1	0	1	0	0
<i>D. bonasus</i>	0	1	0	1	1	0	0	2	0	0	0	0	0	1	0	1	0	0
<i>P. ardea</i>	0	2	0	1	1	0	0	2	0	2	0	0	0	0	1	0	2	0
<i>P. rufosignatus</i>	1	2	0	1	0	0	0	2	0	2	0	0	0	0	1	0	2	0
<i>P. wittei</i>	1	2	0	1	0	0	0	2	0	2	0	0	0	0	1	0	2	0
<i>K. signatus</i>	1	1	1	0	1	2	1	1	1	1	2	1	1	2	0	1	0	0
<i>K. quadraticeps</i>	2	1	1	0	0	0	0	1	1	1	2	1	0	0	0	1	0	1
<i>K. cafrarius</i>	2	1	1	0	0	0	0	1	1	1	2	1	0	0	0	1	0	1
<i>E. flavimargo</i>	1	4	1	1	2	1	2	1	1	3	2	1	2	0	0	1	1	0
<i>O. nuchicornis</i>	1	0	2	1	1	0	1	2	1	0	0	0	0	1	0	0	0	0
<i>O. coenobita</i>	1	1	2	1	1	0	1	2	0	0	0	1	0	1	0	0	1	0
<i>O. illyricus</i>	1	2	0	1	1	0	1	1	0	0	1	1	0	0	0	0	1	0
<i>O. medius</i>	1	0	2	1	1	0	1	2	0	0	0	1	0	1	0	0	1	0
<i>O. nigriventris</i>	1	0	0	1	0	0	1	2	0	0	1	1	0	1	0	1	0	0
<i>O. ovatus</i>	1	1	2	1	1	2	1	2	1	0	0	0	1	1	0	0	1	0
<i>O. bituberculatus</i>	0	3	1	1	2	1	1	0	0	0	0	2	0	2	0	0	1	1
<i>O. depressus</i>	0	1	1	1	2	1	1	0	0	0	0	2	1	2	0	0	1	1

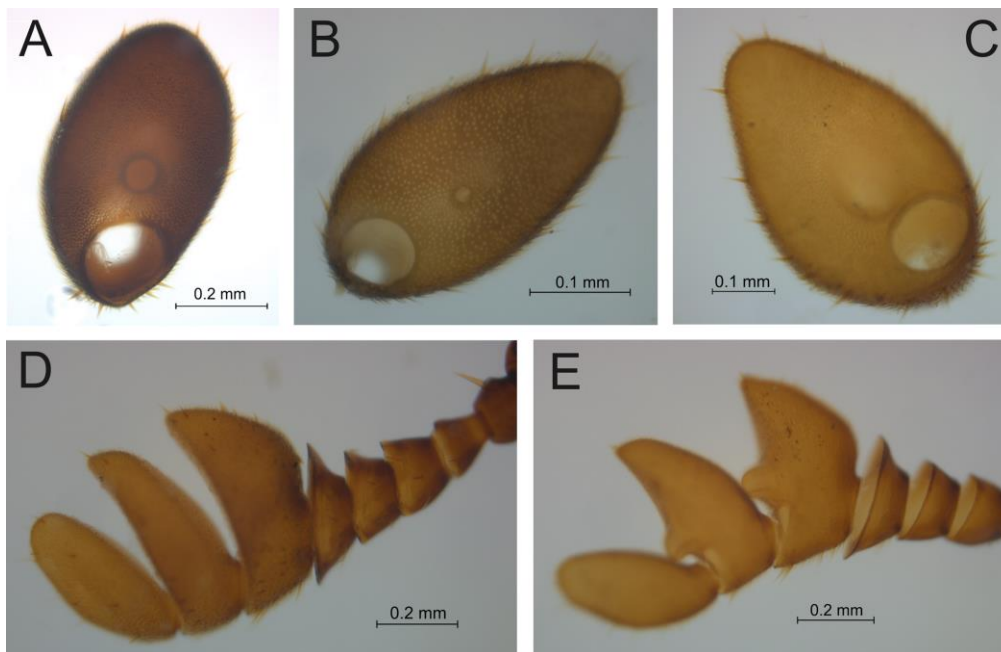
species	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35
<i>S. seniculus</i>	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
<i>D. gazella</i>	0	0	0	0	0	1	-	0	0	2	1	1	0	2	2	0	2
<i>D. bonasus</i>	0	0	0	0	0	1	-	0	0	2	1	1	0	2	2	0	2
<i>P. ardea</i>	0	1	1	0	0	1	-	0	0	2	1	1	0	2	2	0	1
<i>P. rufosignatus</i>	1	1	1	0	0	1	-	0	0	2	1	1	0	2	2	0	1
<i>P. wittei</i>	1	1	1	0	0	1	-	0	0	2	1	1	0	2	2	0	1
<i>K. signatus</i>	1	0	0	0	2	1	-	0	0	2	1	1	0	2	2	0	2
<i>K. quadraticeps</i>	1	1	0	0	2	1	-	0	0	2	1	1	0	2	2	0	2
<i>K. cafrarius</i>	1	1	0	0	2	1	-	0	0	2	1	1	?	?	?	?	?
<i>E. flavimargo</i>	1	0	0	0	1	0	1	1	1	0	0	0	3	3	0	1	0
<i>O. nuchicornis</i>	1	0	0	0	1	0	2	0	0	1	0	0	1	1	0	1	0
<i>O. coenobita</i>	1	0	0	0	1	0	2	0	0	1	0	0	1	1	0	1	2
<i>O. illyricus</i>	1	0	0	1	1	0	0	1	0	4	0	0	1	0	0	2	1
<i>O. medius</i>	1	0	0	0	1	0	2	0	0	1	0	0	1	0	0	1	0
<i>O. nigriventris</i>	2	0	0	1	1	0	0	1	0	4	0	0	1	0	0	2	1
<i>O. ovatus</i>	1	0	0	0	1	0	2	1	1	1	0	0	1	1	0	2	2
<i>O. bituberculatus</i>	1	1	0	0	3	1	-	0	0	3	0	2	2	2	1	0	2
<i>O. depressus</i>	1	1	0	0	3	1	-	0	0	3	0	2	2	2	1	0	2

828

829

830 **Figures**

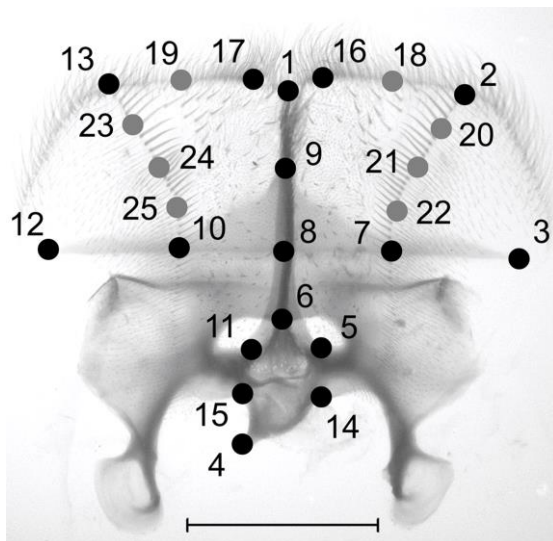
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832

833 **Figure 1.** Antennal scape, central cavity of: A) *Phalops ardea*; B) *Kurtops signatus*; C)
 834 *Digitonthophagus gazella*; D-E) Different expansions of the central part is shown in two
 835 antennae of *Digitonthophagus gazella*.

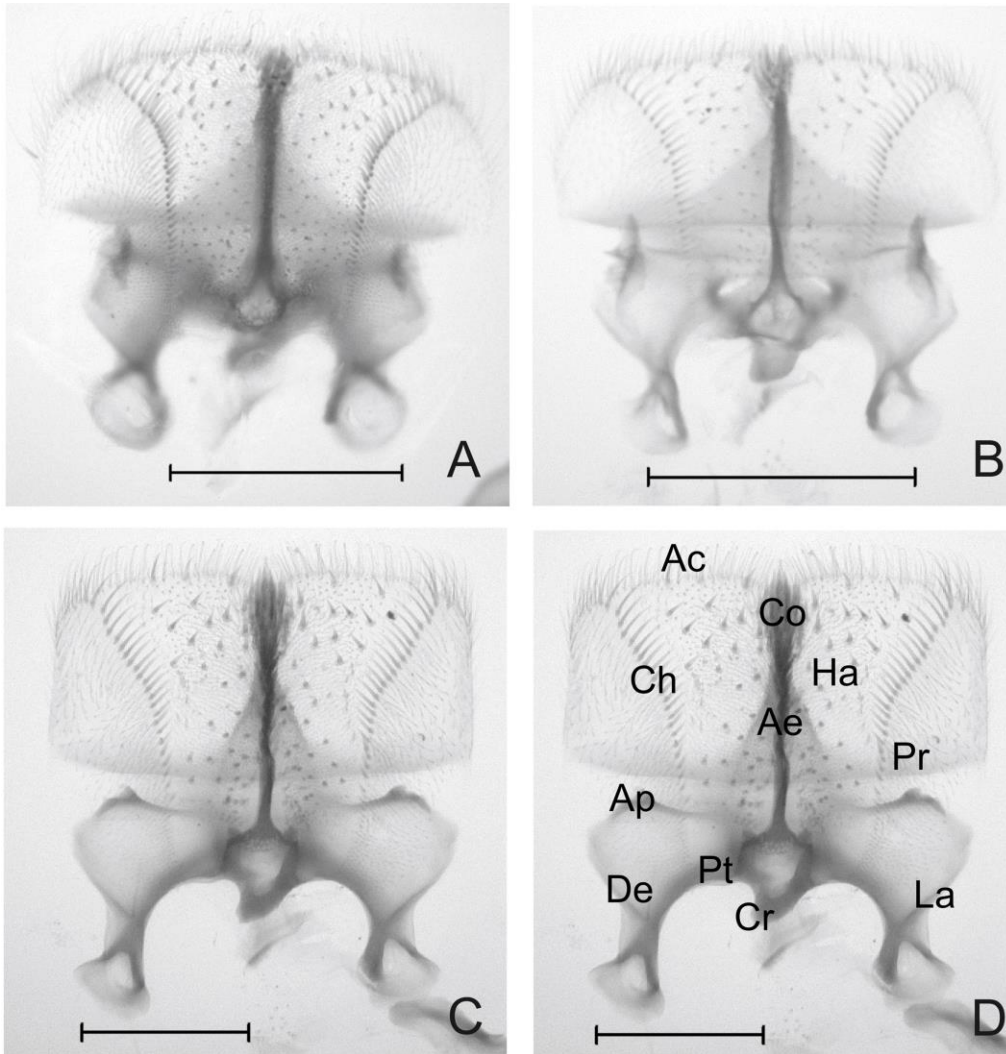
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838 **Figure 2.** Points configuration for the geometric morphometrics analysis of the
 839 epipharynx, with the landmarks marked in black and the semilandmarks in dark grey.
 840 Scalebar = 0.5 mm.

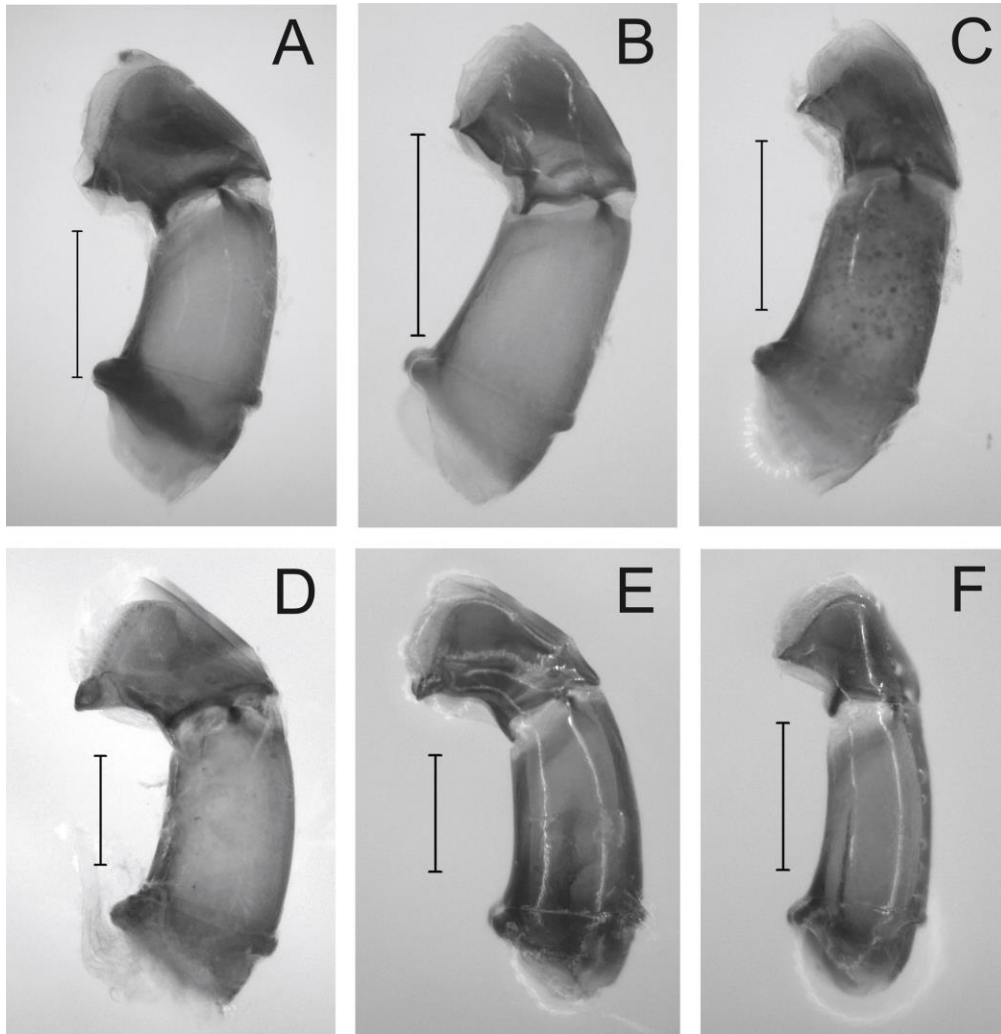
841



842
 843 **Figure 3.** Epipharynx of A) *Kurtops caffrarius* (scalebar = 0.5 mm); B) *K. quadraticeps*
 844 (scalebar = 0.5 mm); C) *K. signatus* (scalebar = 0.2 mm); D) Scheme of the various
 845 parts of the epipharynx: Ac = Acropariae; Co = Coripha; Ha = Haptomerum; Ch =
 846 Chaetopariae; Ae = Anterior epitorma; Pr = Proplegmatium; Ap = Apotormae; Pt =
 847 Pternotormae; Cr = Crepis; De = Dexiotorma; La = Laeotorma.

848

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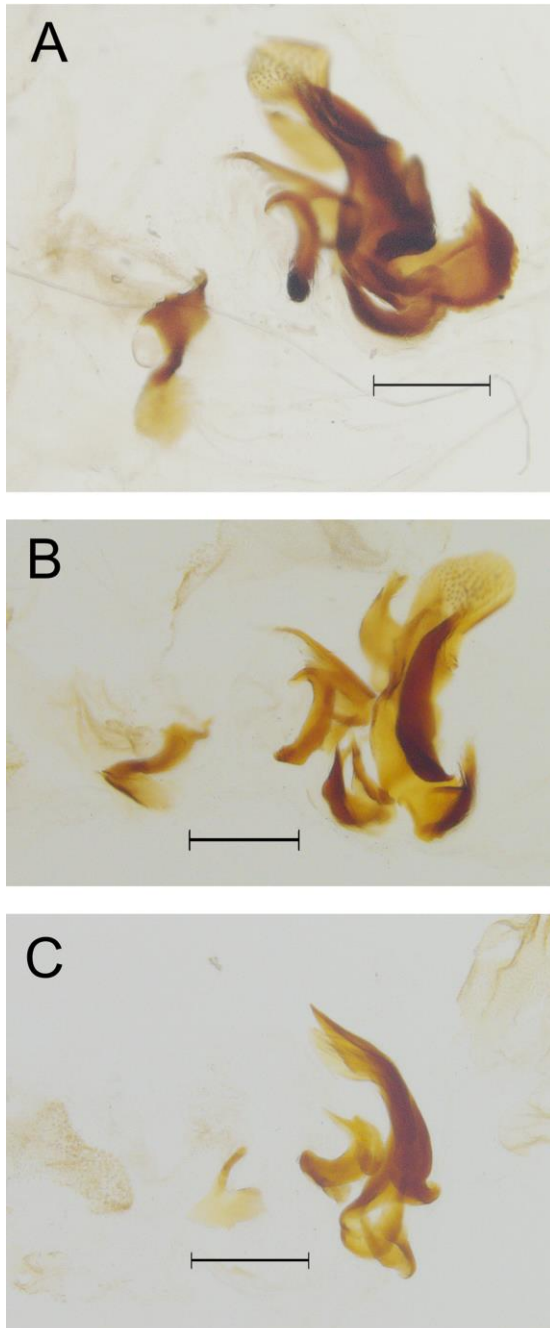


850

851 **Figure 4.** Aedeagus of A) *Digitonthophagus bonasus* (scalebar = 1.0 mm); B) *D.*
 852 *gazella* (scalebar = 1.0 mm); C) *Phalops ardea* (scalebar = 1.0 mm), D) *Kurtops*
 853 *caffrarius* (scalebar = 0.5 mm); E) *K. quadraticeps* (scalebar = 0.5 mm); F) *K. signatus*
 854 (scalebar = 0.5 mm).

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856

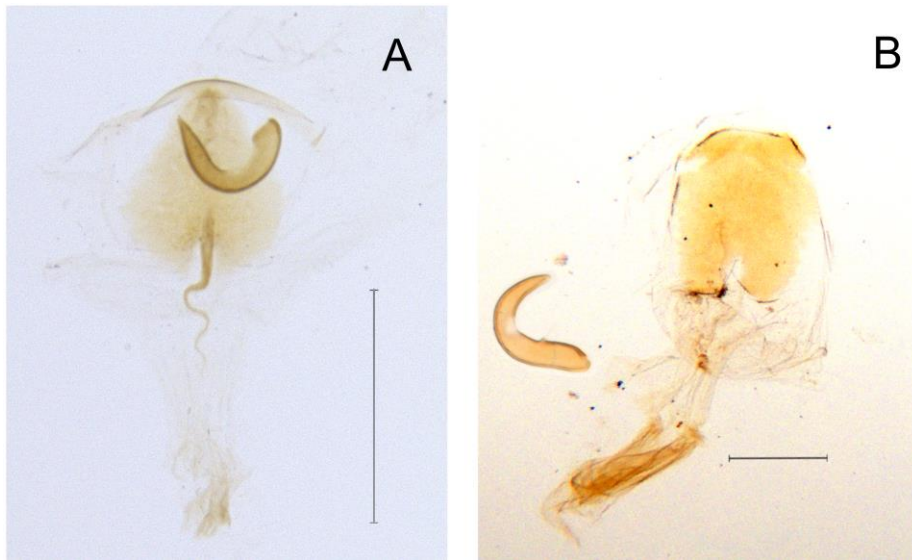


857

858 **Figure 5.** The endophallus sclerites of A) *Kurtops cafferarius*; B) *K. quadraticeps*; C) *K.*859 *signatus*. Scalebar = 0.2 mm.

860

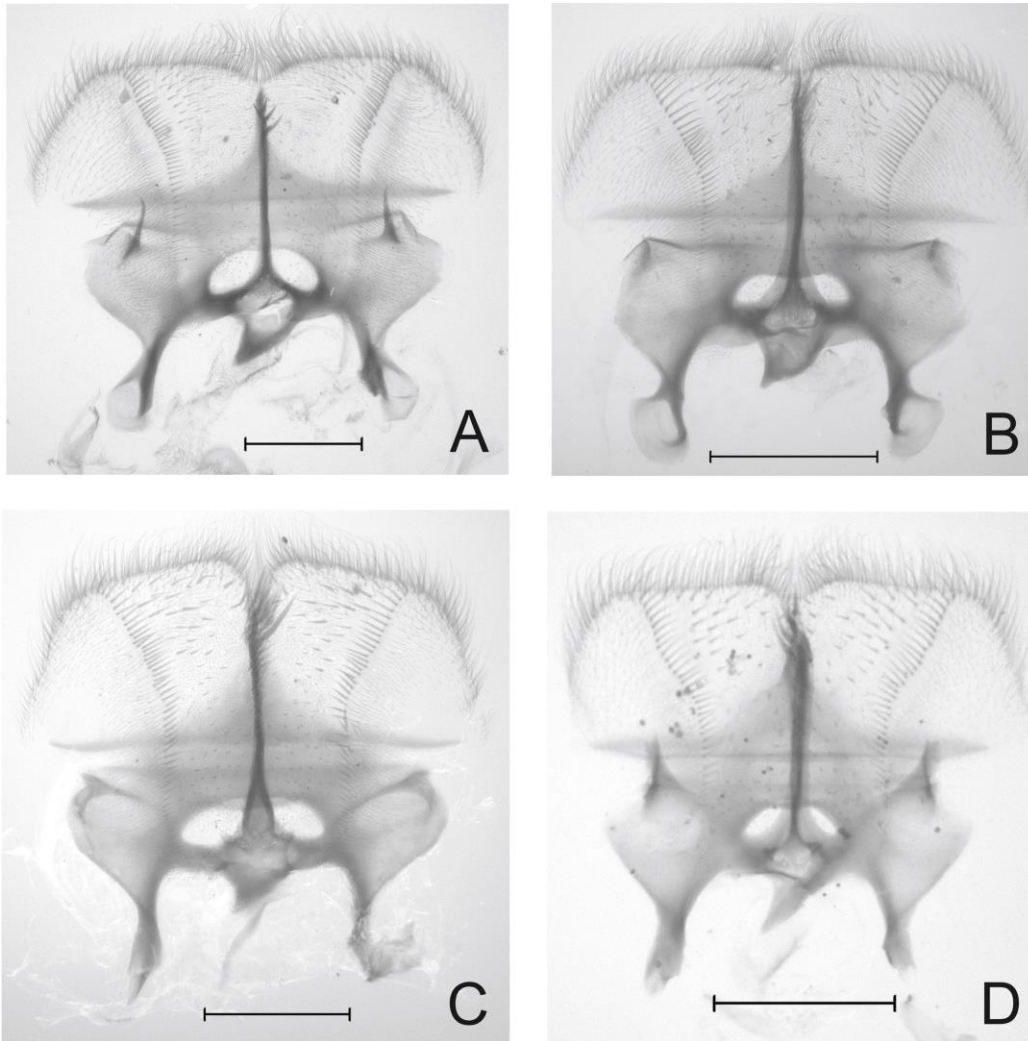
861



862
863 **Figure 6.** Vagina and receptaculum seminis of A) *Kurtops quadraticeps*, scalebar = 0.5
864 mm; B) *K. signatus*, scalebar = 0.2 mm.

865

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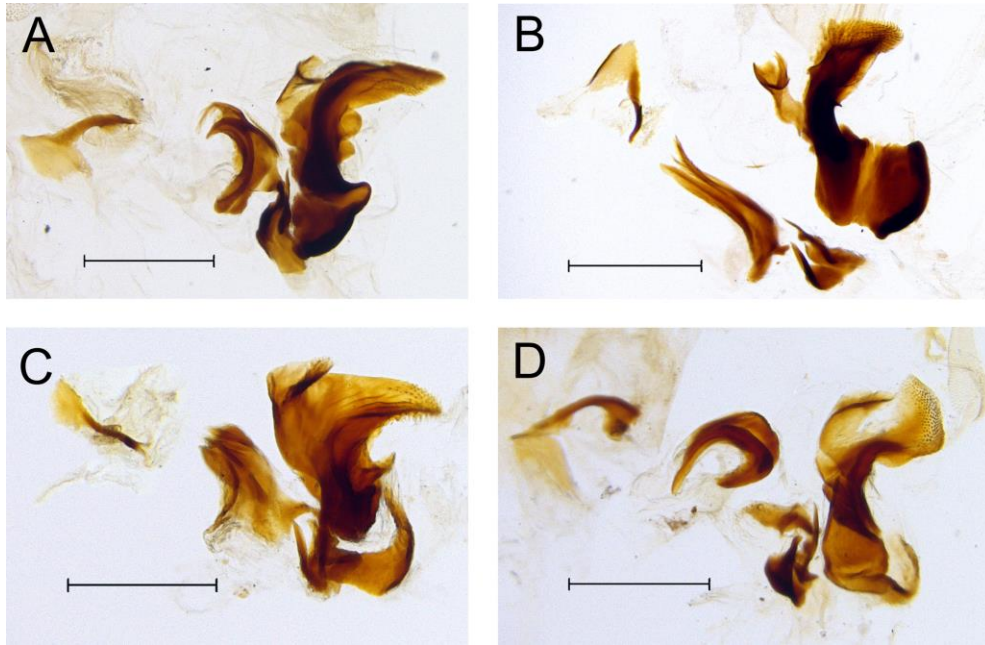


867

868 **Figure 7.** Epipharynx of A) *Digitonthophagus bonasus*; B) *D. gazella*; C) *Phalops*
869 *ardea*; D) *P. wittei*. Scalebars = 0.5 mm.

870

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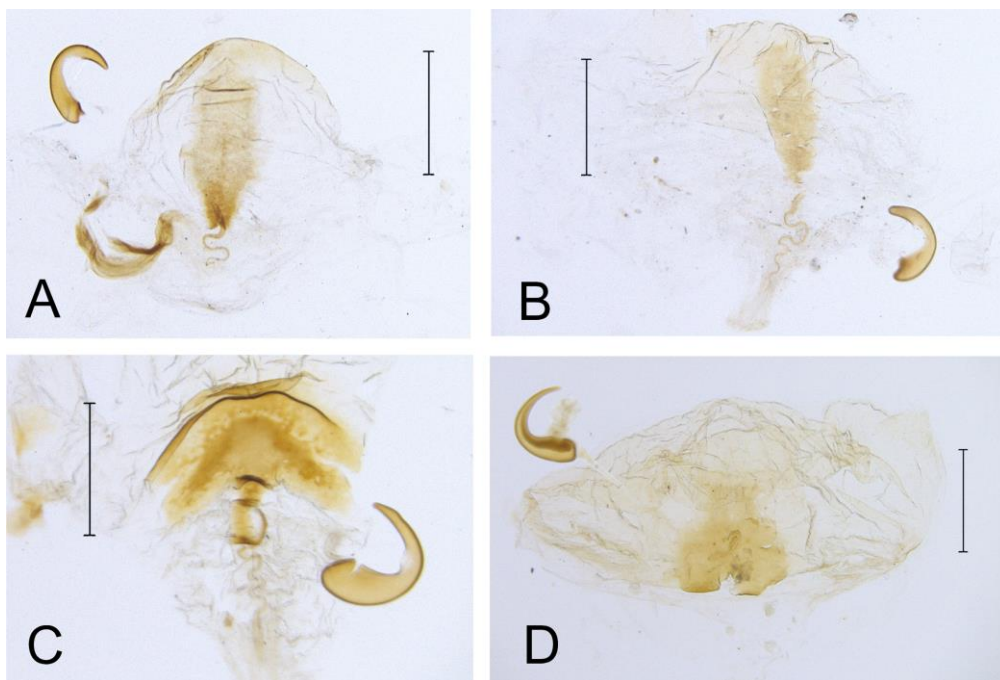


872

873 **Figure 8.** The endophallus sclerites of A) *Digitonthophagus bonasus*; B) *D. gazella*; C)874 *Phalops ardea*; D) *P. wittei*. Scalebar = 0.5 mm.

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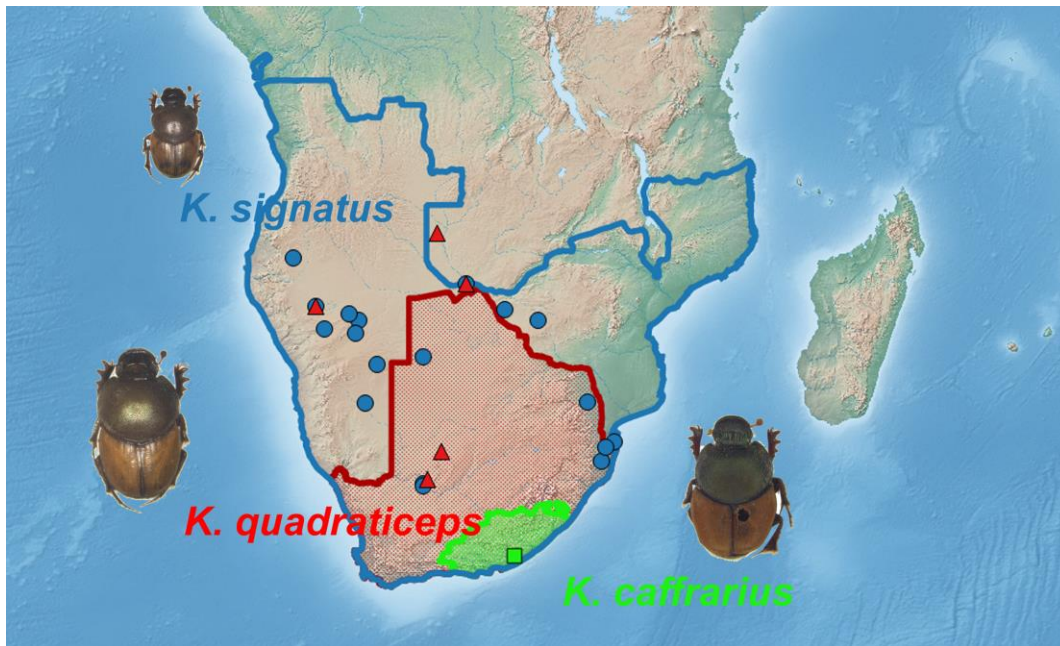


877

878 **Figure 9.** Vagina and receptaculum seminis of A) *Digitonthophagus bonasus*; B) *D.*879 *gazella*; C) *Phalops ardea*; D) *P. wittei*. Scalebar = 0.5 mm.

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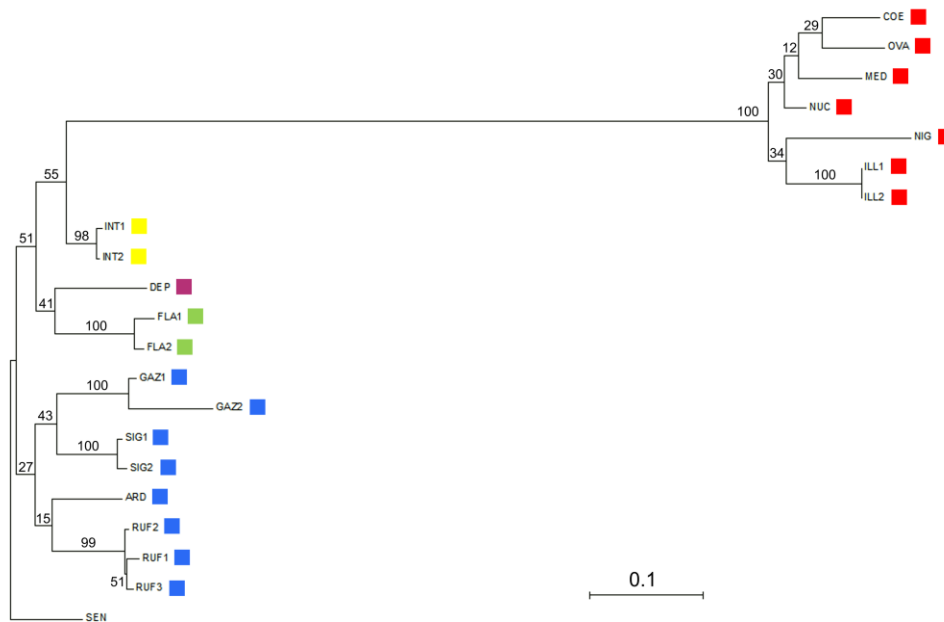
882

883 **Fig. 10.** Distribution map and photos of *Kurtops caffrarius* (green), *K. quadriceps*884 (red) and *K. signatus* (blue).

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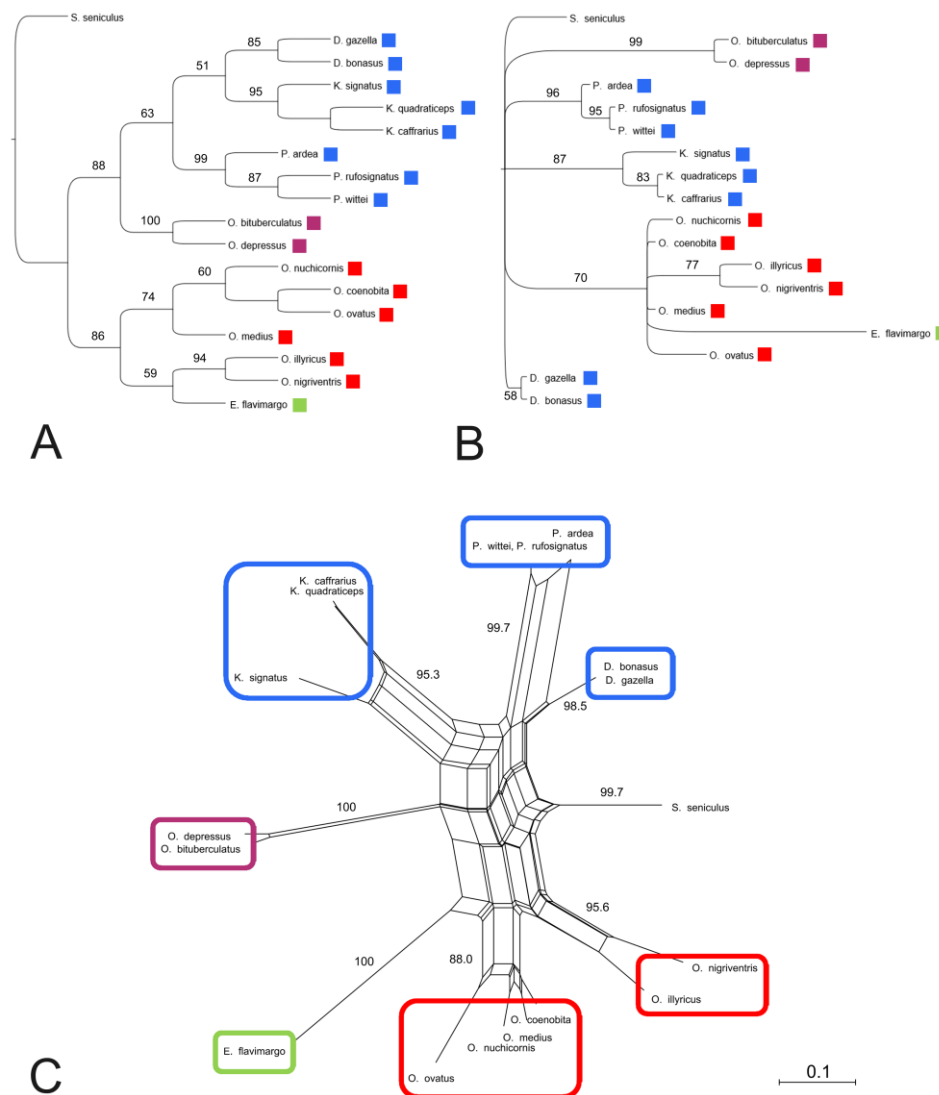


888

889 **Fig. 11.** Maximum Likelihood tree from TN93 method (uniform rates) showing the
 890 bootstrap support values on branches. On the tree, *Onthophagus s.l.* are marked in red,
 891 *O. depressus* in purple, *O. interstitialis* in yellow, *Euonthophagus flavimargo* in green,
 892 and *Phalops*, *Digitonthophagus* and *Kurtops gen.n.* in blue. The acronyms are the same
 893 as in Table 1: SEN = *Serrophorus seniculus*; GAZ = *Digitonthophagus gazella*; SIG =
 894 *Kurtops signatus*; FLA = *Euonthophagus flavimargo*; DEP = *Onthophagus depressus*;
 895 COE = *O. coenobita*; ILL = *O. illyricus*; INT = *O. interstitialis*; MED = *O. medius*; NIG
 896 = *O. nigriiventris*; NUC = *O. nuchicornis*; OVA = *O. ovatus*; ARD = *Phalops ardea*,
 897 RUF = *P. rufosignatus*.

898

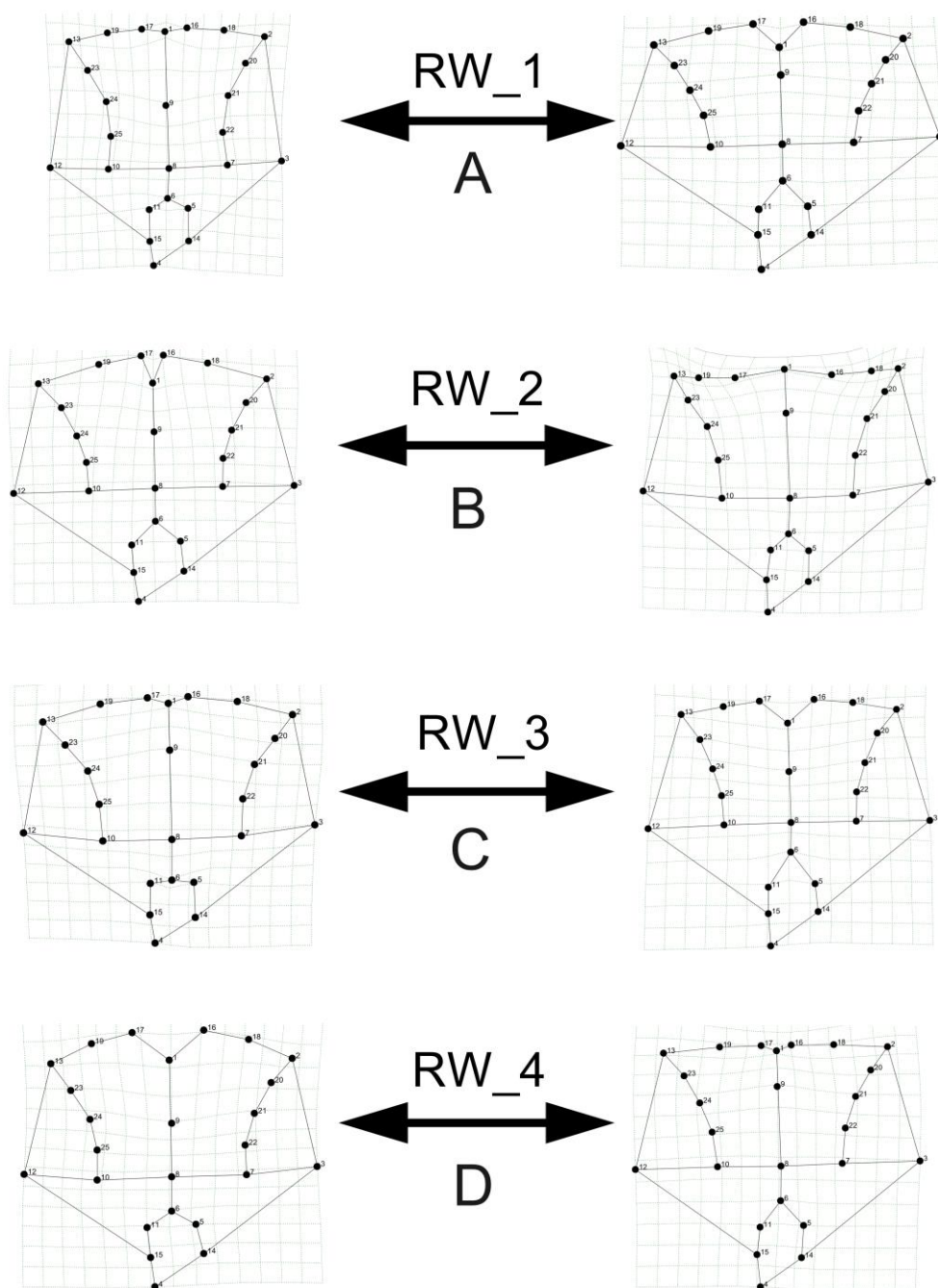
899



900
 901 **Fig. 12.** A) The single tree obtained from maximum parsimony analysis with successive
 902 weighting option (Length = 49,130, CI = 0.775). The Bootstrap support values (majority
 903 rule 50%) from PAUP are shown above the branches, the resampling from TNT
 904 (bootstrap standard, symmetric resampling, and jackknife respectively) gave analogous
 905 results (not shown here); B) 50% majority rule consensus tree from Bayesian inference
 906 analysis, with the support values shown on branches; C) splits tree by neighbor-net
 907 method, with the bootstrap support values for each group shown on branches. In each
 908 tree *Onthophagus* are marked in red, *Euonthophagus flavimargo* in green, *Onthophagus*
 909 *bituberculatus* and *O. depressus* in purple, and *Phalops*, *Digitonthophagus* and *Kurtops*
 910 **gen.n.** in blue.

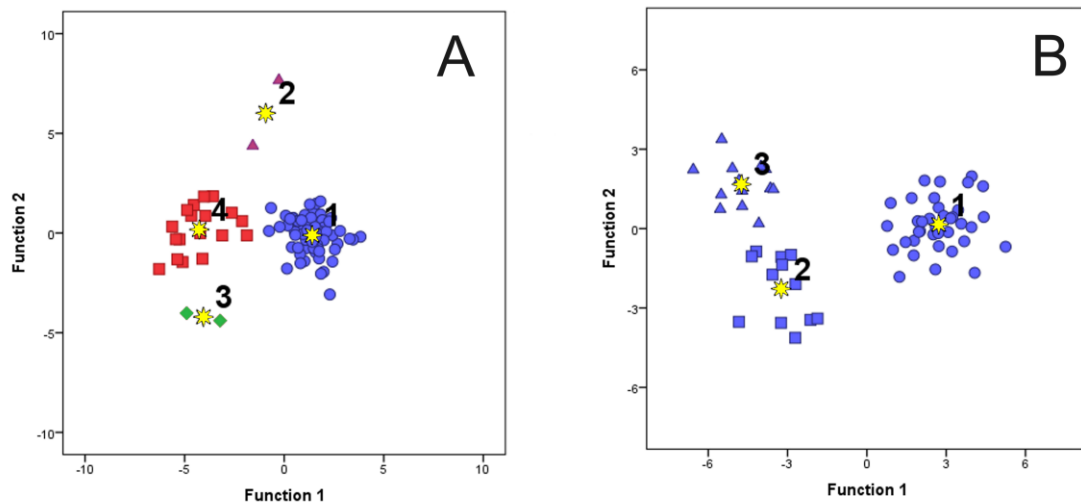
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913

914 **Fig. 13.** The extreme deformation grids obtained by each axis of the RWs 1-4, that have
 915 percent values of explained variance greater than 5%, namely A) RW_1 = 37.08%, B)
 916 RW_2 = 16.81%, C) RW_3 = 11.92%, and D) RW_4 = 9.43%.



917

918 **Fig. 14.** CVA ordination plots derived from analysis of morphometric data for the
 919 epipharynx in which yellow stars represent group centroids. A. Four groups defined for
 920 20 species of Onthophagini: (1) *Phalops*, *Digitonthophagus* and *Kurtops* (blue circles);
 921 (2) *Onthophagus bituberculatus* and *O. depressus* (purple triangles); (3) *Euonthophagus*
 922 *flavimargo* (green rhombus); (4) *Onthophagus s.l.* (red squares). B) Three groups
 923 defined for genera of the *Phalops* complex (1) *Phalops* (circles); (2) *Digitonthophagus*
 924 (squares); (3) *Kurtops* **gen.n.** (triangles).

925

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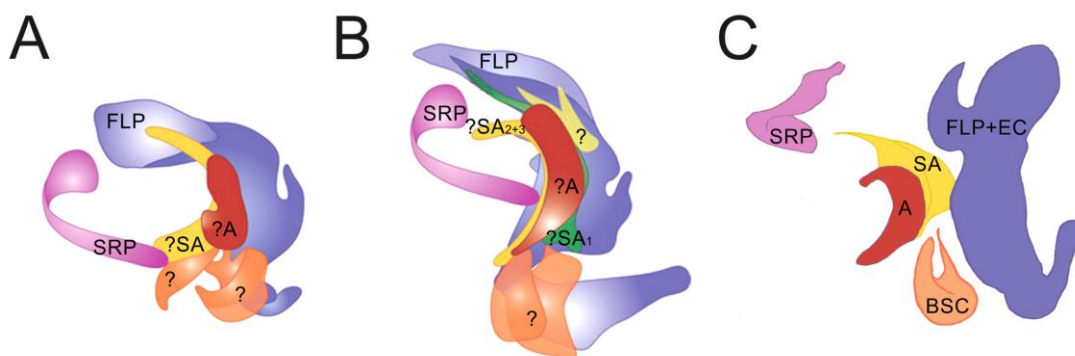
928 **Supplementary Material**

929

930 **Supplementary material 1 - The endophallus sclerites in Onthophagini**

931 The homologies among the various parts that constitute the extremely complicated
 932 endophallus sclerites have been recently highlighted by Tarasov and Solodovnikov
 933 (2011) for many Onthophagini. Besides, in *Phalops* and *Digitonthophagus* only the
 934 fronto-lateral peripheral (FLP) and the superior right peripheral (SRP) sclerites were
 935 definitely recognized, the other sclerites being marked as “unknown” since they were
 936 extremely different from those of other taxa examined (Tarasov and Solodovnikov
 937 2011). Subsequently, the homologies of the sclerites within Scarabaeinae were
 938 evaluated and assessed by Tarasov and Génier (2015), but neither of the two taxa were
 939 included in the analysis. The basal semicircular (BSC), the axial (A) and subaxial (SA)
 940 sclerites of some Scarabaeinae may be considered homologous to the “unknown” ones
 941 of both *Phalops* and *Digitonthophagus* (Tarasov and Solodovnikov 2011), but also to
 942 those of *Kurtops* **gen.n.**

943 While the BSC sclerite was clearly identifiable as a distinct entity in the examined
 944 species, the A and SA sclerites, as hypothesized by Tarasov and Genier (2015), cannot
 945 be differentiated in these taxa and must be considered as a single entity. The “unknown”
 946 sclerite marked in light yellow by Tarasov and Solodovnikov (2011) cannot be
 947 considered as separate from FLP, as is clearly shown in the examined taxa (*Phalops*,
 948 *Digitonthophagus* and *Kurtops* **gen.n.**), and is here named EC (external claw) of FLP.
 949



950

951 **Figure A1.** The scheme of the endophallus sclerites of the three genera: A) *Phalops*
 952 *laminifrons*, B) *Digitonthophagus bonasus* (both modified from Tarasov &
 953 Solodovnikov 2011), and C) *Kurtops quadraticeps*. The acronyms of the different parts

954 were reported on the figures, and correspond to FLP = fronto-lateral peripheral sclerite,
955 SRP = superior right peripheral sclerite, A = axial sclerite, SA = subaxial sclerite, and
956 BSC = basal semicircular sclerite.

957

958

959 **References**

960 Tarasov, S.I. & Génier, F. (2015) Innovative Bayesian and Parsimony Phylogeny of
961 Dung Beetles (Coleoptera, Scarabaeidae, Scarabaeinae) Enhanced by Ontology-
962 Based Partitioning of Morphological Characters. *PlosOne*, *10*, e0116671.
963 doi:10.1371/journal.pone.0116671.

964 Tarasov, S.I & Solodovnikov, A.Y. (2011) Phylogenetic analyses reveal reliable
965 morphological markers to classify mega-diversity in Onthophagini dung beetles
966 (Coleoptera: Scarabaeidae: Scarabaeinae). *Cladistics*, *27*, 1–39.

967

968

969 **Supplementary material 2 - Characters list**

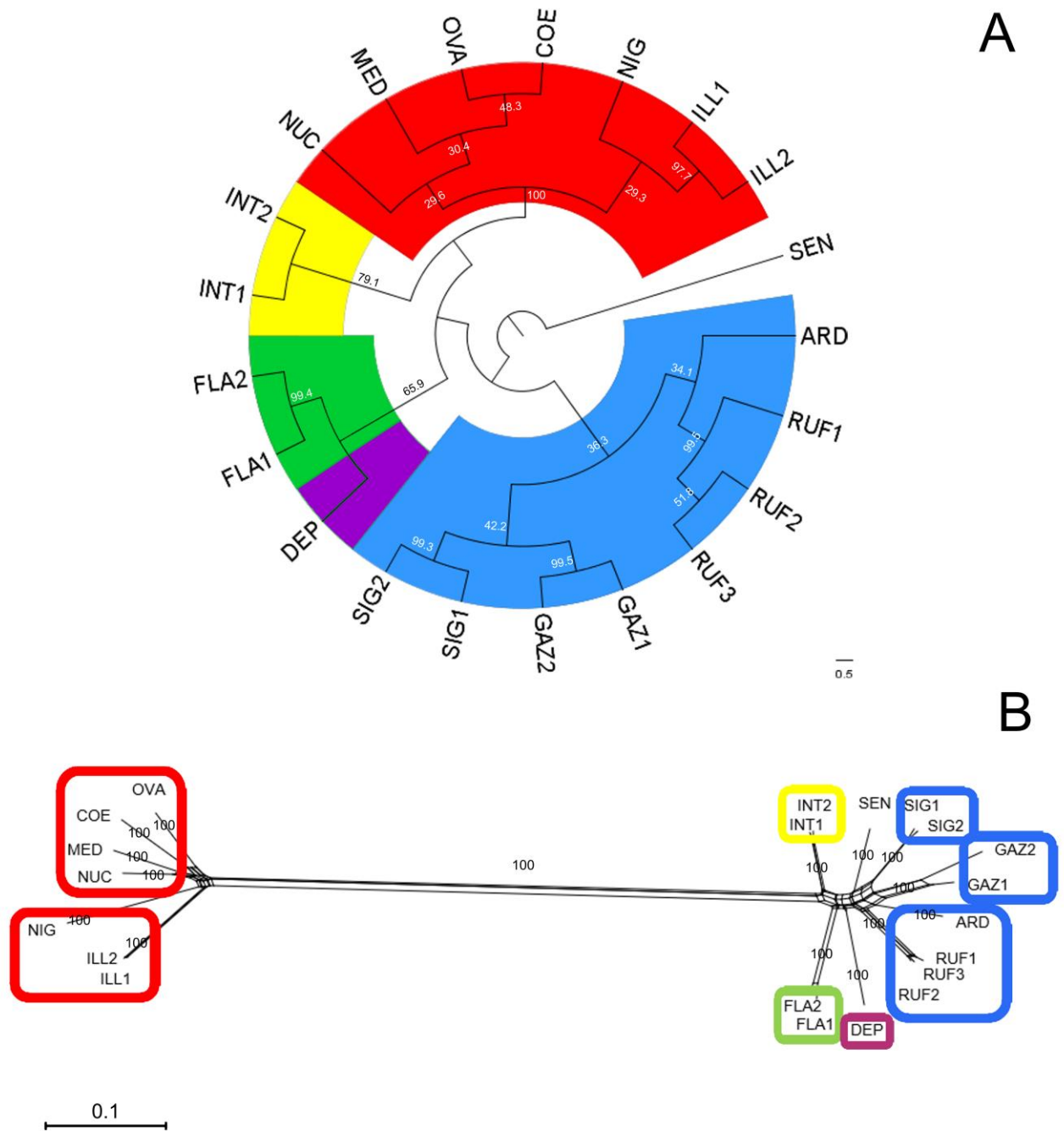
- 970 1. *Epipharynx, the fore half in front of the proplegmatium*: (0) subtrapezoidal; (1)
971 subrectangular; (2) sickle-shaped.
- 972 2. *Epipharynx, the fore margin*: (0) rectilinear; (1) only slightly sinuate in the middle;
973 (2) deeply and narrowly notched; (3) largely notched; (4) convex.
- 974 3. *Epipharynx, corypha*: (0) absent, only a row of few, sparse and long setae is present
975 along the anterior epitorma; (1) present, constituted by a thick tuft of short setae; (2)
976 present, constituted by short and thin setae.
- 977 4. *Epipharynx, anterior epitorma*: (0) never reaching the fore margin; (1) reaching the
978 fore margin.
- 979 5. *Epipharynx, anterior epitorma*: (0) thick; (1) narrow; (2) very narrow.
- 980 6. *Epipharynx, the fore triangular sclerotized area of haptomerum*: (0) large and
981 lowered; (1) narrow and lowered; (2) narrow and elongate.
- 982 7. *Epipharynx, proplegmatium position*: (0) in the anterior third of the epipharynx; (1)
983 in the medial part of epipharynx surface; (2) in the posterior third of the epipharynx.
- 984 8. *Epipharynx, chaetopariae*: (0) subrectilinear; (1) angulate; (2) arched.
- 985 9. *Epipharynx, apex of the crepis left turned and*: (0) sharp; (1) blunt.
- 986 10. *Epipharynx, laeotorma and dexiotorma distal part*: (0) medium length, with the
987 insertion to mandibles area drop-like; (1) very short and markedly rounded at level of
988 insertion to mandibles; (2) very thin and often elongate, with the insertion to mandibles
989 very narrow; (3) short and spatulate.
- 990 11. *Epipharynx, pternotormae*: (0) short and thick; (1) longer and narrower; (2) greatly
991 reduced.
- 992 12. *Mentum, fore margin*: (0) a large and rounded notch; (1) deeply and triangular
993 notched; (2) a large and triangular notch.
- 994 13. *Mentum, the basal margin*: (0) triangular notched; (1) sinuate; (2) rectilinear.
- 995 14. *Head, clypeus fore margin*: (0) not-incised; (1) only feebly sinuate; (2) deeply V-
996 notched.
- 997 15. *Head, genae*: (0) not especially developed; (1) protruding.
- 998 16. *Pronotum, on the whole*: (0) oval; (1) rounded.
- 999 17. *Pronotum hind margin*: (0) angulate; (1) rounded; (2) straight.

- 1000 *18. Pronotum, fore angles:* (0) more or less developed, blunt and always rectilinear,
 1001 facing forward; (1) well-developed, sharp, and outward turned.
- 1002 *19. Legs, fore tibia:* (0) markedly dimorphic in the two sexes, being narrow and inward
 1003 arched in male; (1) almost identical in both sexes, but showing differences in the tooth
 1004 shape; (2) showing no sexual dimorphism.
- 1005 *20. Elytra, 7th stria:* (0) sinuate; (1) rectilinear.
- 1006 *21. Elytra, 8th stria:* (0) absent; (1) present but incomplete, and distally fused to 7th
 1007 one.
- 1008 *22. Male genitalia, phallobase/parameres ratio:* (0) reaching almost 2:1; (1) about 1:1.
- 1009 *23. Male genitalia, parameres:* (0) quadrangular symmetrical, carrying laminar
 1010 expansions ventrally, the apices rounded, with a digitiform expansion subapically; (1)
 1011 simple, symmetrical, without laminar expansion on ventral side, the apices sharp but
 1012 short, largely triangular-shaped; (2) arched, the apices large and sharp, carrying a
 1013 laminar, rounded expansion subapically; (3) triangular-shaped ventrally, pointed at
 1014 apex, and flat apically.
- 1015 *24. Male genitalia, lamella copulatrix:* (0) present; (1) absent.
- 1016 *25. Male genitalia, lamella copulatrix present and :* (0) constituted by a globular
 1017 expansion and a rectilinear part, comma-shaped; (1) well-developed, globose, C-shaped;
 1018 (2) cupoliform, almost globose, well-sclerotized.
- 1019 *26. Male genitalia, endophallus carrying apically:* (0) many small diffusely-arranged
 1020 scales, sometimes almost effaced; (1) diffusely-arranged scales, and two well-defined,
 1021 ventral areas with more thickened scales; reduced scales, but a large transversal ridge
 1022 with well-developed scales.
- 1023 *27. Male genitalia, raspula:* (0) absent; (1) present.
- 1024 *28. Male genitalia, FLP (= fronto-lateral peripheral) sclerite:* (0) laminar, double, with
 1025 projections encircling SA+A (= subaxial + axial) sclerite; (1) band-shaped, encircling
 1026 the SA+A; (2) large, well-developed, with evident expansions apically and basally; (3)
 1027 linked to SA+A; (4) well sclerotized, and pluridigitate.
- 1028 *29. Male genitalia, SA+A sclerites:* (0) rod-shaped, usually separate; (1) C-shaped,
 1029 connected.
- 1030 *30. Male genitalia, BSC (= basal semicircular) sclerite:* (0) absent, (1) present, well-
 1031 developed; (2) present, very reduced.

- 1032 31. *Female genitalia, infundibular wall*: (0) carrying a large longitudinal sclerotization
1033 subrectangular or mushroom-shaped; (1) supported by a thin, "arched" sclerotization;
1034 (2) with a sinuate, asymmetrical and folded sclerotization; (3) complex sclerotization,
1035 with two pillar-shaped lateral sclerotizations and a key-hole central sclerotization.
- 1036 32. *Female genitalia, vagina*: (0) enlarged; (1) elongate and wrinkled; (2) rounded; (3)
1037 extremely narrow and elongate.
- 1038 33. *Female genitalia, infundibular tube*: (0) very sclerotized, orthogonal to infundibular
1039 wall; (1) non sclerotized, lowered at floccular level; (2) sigmoidal, placed below the
1040 well-developed sclerotization of the infundibular wall.
- 1041 34. *Female genitalia, receptaculum seminis*: (0) elongate, tapering to often sharp apex;
1042 (1) elongate, subequal on the whole length, the apex slightly narrowed, but never sharp;
1043 (2) short and tough, rounded at apex.
- 1044 35. *Female genitalia, receptaculum seminis at base*: (0) cone-shaped, carrying a
1045 sclerotized portion on infundibular tube; (1) gently rounded; (2) truncated.
1046
1047

1048 **Supplementary material 3 – Further molecular results**

1049



1050

1051 **Figure A3.** A) Maximum Likelihood tree from TN93 method (uniform rates) showing
 1052 on branches non-parametric bootstrap support values, similar to the SH like aLRT
 1053 values (not shown). B) Splits tree by neighbor-net method, with the bootstrap support
 1054 values for each taxon showed on branches. On each tree, *Onthophagus s.l.* are marked
 1055 in red, *Euonthophagus flavimargo* in green, *O. depressus* in purple, *O. interstitialis* in

1056 yellow, and *Phalops*, *Digitonthophagus* and *Kurtops* **gen.n.** in blue. The acronyms are
1057 the same than in Table 1: SEN = *Serrophorus seniculus*; GAZ = *Digitonthophagus*
1058 *gazella*; SIG = *Kurtops signatus*; FLA = *Euonthophagus flavimargo*; DEP =
1059 *Onthophagus depressus*; COE = *O. coenobita*; ILL = *O. illyricus*; INT = *O.*
1060 *interstitialis*; MED = *O. medius*; NIG = *O. nigriventris*; NUC = *O. nuchicornis*; OVA =
1061 *O. ovatus*; ARD = *Phalops ardea*, RUF = *P. rufosignatus*.

1062

1063

1064 **Supplementary material 4 - The *Kurtops* species**

1065 The species currently included in *Kurtops* **gen.n.** are here described in detail. The
1066 figures quoted here are in

1067

1068 ***Kurtops caffrarius* (d'Orbigny, 1902: 171)**

1069 (Figs 3A, 4D, 5A)

1070 *Type material.* 1 male, holotype. SOUTH AFRICA: Caffraria. Muséum National
1071 d'Histoire Naturelle (MNHN, Paris, France).

1072 *Description.* Length 0.92 cm. Head blackish green, covered by a light yellow, thin and
1073 long pubescence; clypeus largely rounded, and genae only slightly expanded; vertex and
1074 frontal carinae large, well-developed, blade-shaped; thick and dense granules on the
1075 whole surface, antennae ochreous. Pronotum markedly rounded, very dark olive green,
1076 entirely covered by small, thick and dense granules and a light yellow, thin and long
1077 pubescence. Elytra ochreous and opaque, the striae narrow and the interstriae covered
1078 by dense, small setigerous points. Pigidium dark brown entirely covered by large and
1079 deep setigerous points, and a dense, light yellow, thin and long pubescence. Very dark
1080 brown legs and body lower side. Metasternal disc with large, deep, and rade points.
1081 Epipharynx. The fore margin rounded; acropariae and acanthopariae long and thick;
1082 acanthopedia covered by a dense pubescence; corypha constituted by a well-developed
1083 tuft of setae; chaetopariae angulate, with the setae short and dense; proplegmatium
1084 thick; laeotoema and dexiotorma short and stout, pternotormae very small and thick;
1085 crepis well-sclerotized and large, with the apex blunt.

1086 Male genitalia. Phallobase of aedeagus twice as long as the paramers, slightly arched,
1087 with the diameter subequal on the whole length; symmetrical paramers with a rounded
1088 and just a little protruding superior part, the apices large and sharp, slightly hook-
1089 shaped, carrying a laminar and rounded expansion subapically. Endophallus entirely
1090 lacking a copulatrix lamella, the accessory lamellae well-developed, comprised of
1091 various parts (SRP+FLP/EC+SA+A+BSC), similarl to those of *Phalops* and
1092 *Digitonthophagus* (Fig. 8).

1093 *Distribution.* The species is known only from the type locality in SE Eastern Cape
1094 province, formerly designed as Kaffraria (Fig. 10).

1095 *Remarks.* The species at present is known only from a single specimen, the male
1096 holotype from Caffraria. The female is unknown.

1097

1098

1099 ***Kurtops quadriceps* (Harold, 1867: 52)**

1100 (Figs 3B, 4E, 5B, 6A)

1101 *Type material.* SOUTH AFRICA: Orange Free State. Museum für Naturkunde der
1102 Humboldt-Universität (ZMHB, Berlin, Germany).

1103 *Description.* Length 0.60-1.00 cm. Head dark green, covered by a light yellow, thin and
1104 long pubescence; clypeus largely rounded, and genae only slightly expanded; vertex and
1105 frontal carinae large, well-developed, blade-shaped; thick and dense granules on the
1106 whole surface, antennal scape reddish brown, lamellae ochreous. Pronotum markedly
1107 rounded, dark green, entirely covered by a light yellow, thin and long pubescence, with
1108 small, thick and dense rasping points reducing from the fore to hind margin, and larger
1109 points with smaller granules in the hind central half. Elytra yellowish ochreous often
1110 carrying brown and elongate patches more or less developed till covering almost the
1111 whole surface, the striae narrow and the interstriae covered by equally spaced, small
1112 setigerous points. Pigidium blackish brown, entirely covered by large and deep
1113 setigerous points, and a dense, light yellow, thin and long pubescence. Very dark brown
1114 legs and body lower side. Metasternal disc with large, deep, and scattered points.
1115 Epipharynx. The fore margin rounded; acropariae and acanthopariae long and thick;
1116 acanthopedia covered by a dense pubescence; corypha constituted by a well-developed
1117 tuft of setae; chaetopariae angulate, with the setae short and dense; proplegmatium
1118 thick; laeotoema and dexiotorma short and stout, pternotormae very small and thick;
1119 crepis squared and large.

1120 Male genitalia. Phallobase of aedeagus twice as long as the paramers, slightly arched,
1121 with the diameter subequal along the whole length; symmetrical paramers with a
1122 rounded and just a little protruding superior part, the apices small and sharp, slightly
1123 hook-shaped, carrying a laminar and rounded expansion very near the apices.

1124 Endophallus entirely lacking a copulatrix lamella, the accessory lamellae well-
1125 developed, comprised of various parts (SRP+FLP/EC+SA+A+BSC), showing the same
1126 general pattern to those *Phalops* and *Digitonthophagus* (Fig. 8).

1127 Female genitalia. Infundibular wall triangular-shaped, with the basal part rounded;
 1128 infundibular tube well-sclerotized, plurisinate and tapering distally. Receptaculum
 1129 seminis large, C-shaped, the apex sharp, almost entirely sclerotized, the proximal part to
 1130 infundibulum shorter than the distal one.

1131 *Distribution.* The species is known from South Africa (where is diffusely reported from
 1132 Orange Free state, Cape Colony, Natal, Western Cape [Twee Rivieren]), and Botswana
 1133 (Fig. 10).

1134 *Remarks.* It is noteworthy that (unlike *K. caffrarius*) this species has a very wide
 1135 distribution and is known throughout Southern Africa.

1136

1137

1138 ***Kurtops signatus* (Fåhraeus, 1857: 304)**

1139 (Figs 3C, 4F, 5C, 6B)

1140 *Type material.* MOZAMBIQUE: Limpopo river. Naturhistoriska Riksmuseet (NHRS,
 1141 Stockholm, Sweden).

1142 *Synonymy.* *O. junodi* d'Orbigny, 1902: 223. Muséum National d'Histoire Naturelle
 1143 (MNHN, Paris, France).

1144 *Description.* Length 0.50-0.60 cm. Head shiny black, covered by a rade, light yellow,
 1145 thin and long pubescence; clypeus deeply V-notched in the middle, and genae only
 1146 slightly expanded; vertex and frontal carinae large, well-developed, blade-shaped; dense
 1147 setigerous points on the whole surface, antennae reddish brown. Pronotum rounded,
 1148 shiny black, with thick and dense setigerous granulate points mixed with smaller simple
 1149 points and a light yellow, short and thin pubescence. Elytra ochreous with black dots,
 1150 the striae as large as the striae points, and the interstriae covered by 2 rows of small
 1151 setigerous points. Pigidium shiny black, covered by superficial points mixed with
 1152 smaller ones. Very dark brown body lower side and legs, except the ochreous femura.
 1153 Metasternal disc with large, deep, and scattered points.

1154 *Epipharynx.* The fore margin squared; acropariae long and thick; acanthopariae short
 1155 and thin; acanthopedia covered by a pubescence short and evenly distributed; corypha
 1156 comprising a well-developed tuft of setae; chaetopariae angulate, with the setae long in
 1157 the anterior half, far shorter in the posterior half; proplegmatium very thin and slightly

1158 arched; laeotoema and dexiotorma short and stout, pternotormae very small and thick;
1159 short crepis with the apex blunt.

1160 Male genitalia. Phallobase of aedeagus more than twice the length of the paramers,
1161 slightly arched, with the diameter subequal along the whole length; symmetrical
1162 paramers with a rounded and just a little protruding superior part, the apices large and
1163 sharp, subtriangular, the ventral laminar expansion almost not apparent. Endophallus
1164 entirely lacking a copulatrix lamella, the accessory lamellae well-developed, comprised
1165 of various parts (SRP+FLP/EC+SA+A+BSC), similar to the model already evidenced in
1166 *Phalops* and *Digitonthophagus* species (Fig. 8).

1167 Female genitalia. Infundibular wall mushroom-shaped, with the basal part far more
1168 developed than the apical part; infundibular tube plurisinate. Receptaculum seminis
1169 large, J-shaped, the apex sharp, almost entirely sclerotized, the proximal part to
1170 infundibulum longer than the distal one.

1171 *Distribution.* The species is known from Angola, Botswana, Mozambique, Namibia,
1172 South Africa, and Zimbabwe (Fig. 10).

1173 *Remarks.* *O. junodi* d'Orbigny, 1902 (from Mozambique) was synonymized to *O.*
1174 *signatus* by d'Orbigny (1913).

1175

1176

1177 **References**

1178 d'Orbigny, H. (1913) Synopsis des Onthophagides d'Afrique. *Annales de la Société*
1179 *Entomologique de France*, 82, 1–742.

1180

1181

1182 **Supplementary material 5 - The *Phalops* complex**

1183 The comparison of *Kurtops* **gen.n.**, with three species, *Phalops* Erichson, with 38
1184 species (Barbero et al. 2003; Genier 2013), and *Digitonthophagus* (Balthasar) with two
1185 species (Balthasar 1959, 1963; Zunino 1981) led to the identification of the *Phalops*
1186 complex in accord to that already suggested for the *Serrophorus* complex (Tarasov and
1187 Kabakov 2010; Tarasov and Solodovnikov 2011).

1188 These three genera are characterized by extremely differentiated external features that
1189 are very useful as identification characters. The evident sexual dimorphism present in
1190 *Digitonthophagus* and *Phalops* is reduced to the variation of the fore tibiae in *Kurtops*.
1191 The male head carries more or less developed horns in *Digitonthophagus*, and laminar
1192 projections in *Phalops*, but is unarmed in *Kurtops*. The pronotum in *Phalops* and
1193 *Kurtops* has a dense granulation on the whole surface, while in *Digitonthophagus* it is
1194 smooth with sparse, large simple points (*D. gazella*) or few granulate points (*D.*
1195 *bonasus*). The pronotum hind margin is straight only in *Phalops*, and the pubescence is
1196 far thicker and longer in *Kurtops* than in the two other genera. The 8th elytral stria is
1197 absent and the 7th stria sinuate in *Digitonthophagus* and *Kurtops*, while in *Phalops* the
1198 8th stria is distally fused to 7th stria, that is rectilinear.

1199 Also the epipharynx (Figs. 3 and 7) allows to clearly distinguish these taxa (see also the
1200 results of the geometric morphometrics analysis above for more details), e.g. the fore
1201 margin is more deeply notched in *Phalops* and *Digitonthophagus* than in *Kurtops*, the
1202 laeotorma and dexiotorma markedly differ in the three genera, and the apotormae are
1203 characteristically more or less developed in the three genera.

1204 These genera share instead a highly similar pattern for both male and female genitalia,
1205 that confirms the marked proximity among them. In males, the aedeagus is
1206 characterized by short paramers (Fig. 4); in the endophallus the lamella copulatrix (LC)
1207 is absent (while in *Onthophagus s.l.* is always present), and the accessory sclerites (FLP,
1208 SRP, BSC, and A+SA) are conspicuous, and show an analogous and very characteristic
1209 developmental model in the three genera (Figs 5 and 8). In *Onthophagus*, the accessory
1210 sclerites features are markedly different from those of the *Phalops* complex, never being
1211 as developed. Furthermore, the A+SA sclerites are usually straight and not C-shaped,
1212 the BSC sclerite is not present, and the FLP sclerite usually encircles the others
1213 (Tarasov and Solodovnikov 2011).

1214 In females, the infundibular wall of the vagina is sub-rectangular or mushroom-shaped,
1215 and always well-sclerotized, the infundibulum is short and plurisinate, and the
1216 receptaculum seminis is usually elongate, tapering to a sharp apex, with a very reduced
1217 non-sclerotized medial area (Figs 6 and 9). In the *Onthophagus* species here examined,
1218 the infundibular wall support is usually constituted by a narrow and (more or less) H-
1219 shaped sclerotization, the infundibular tube is well-sclerotized and C-shaped, and the
1220 receptaculum seminis has an even diameter along the whole length, the apex rounded,
1221 and a very large non-sclerotized medial area.

1222 On the whole, the *Phalops* complex has a worldwide distribution. Its original
1223 distribution extends in Palearctic (*Phalops* and *Digitonthophagus*), Afrotropical
1224 (*Phalops*, *Kurtops* and *Digitonthophagus*) and Oriental (*Phalops* and *Digitonthophagus*)
1225 regions, but was also introduced in Nearctic, Neotropical and Australian regions (only
1226 *D. gazella*).

1227 The genus *Phalops* was described by Erichson in 1843 (see Barbero et al. 2003 for
1228 further details), and its taxonomic status is not disputed at present. Balthasar (1959:464)
1229 described *Digitonthophagus* as a subgenus of *Onthophagus*, with *D. bonasus* (Fabricius,
1230 1775) as type species of the taxon, furnishing later (Balthasar 1963) the list of the 20
1231 species originally included in the taxon. The author remarked that the majority of the
1232 *Digitonthophagus* species had an Oriental distribution, and only two were located in the
1233 Eastern Palearctic region. Furthermore, according to Balthasar's observations (1959), it
1234 was also very likely that some Afrotropical species would have to be included in this
1235 taxon. Subsequently, Zunino (1981) raised *Digitonthophagus* to a generic level,
1236 including only two out of the 20 species: the Afrotropical *Digitonthophagus gazella*
1237 (Fabricius, 1787) having now a worldwide distribution, and the Oriental *D. bonasus*
1238 (Fabricius, 1775). The remaining Balthasar's *Digitonthophagus* species were later
1239 assigned to five different subgenera within *Onthophagus* (Ochi 2003a, 2003b), that
1240 were subsequently included in the *Serrophorus* complex (Tarasov and Kabakov 2010;
1241 Tarasov and Solodovnikov 2011).

1242

1243

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1269 Onthophagini. *Fauna of Saudi Arabia*, 3, 408–416.
- 1270
- 1271


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1272 #NEXUS
1273
1274 [ File saved by NDE version 0.5.0 ]
1275
1276 BEGIN TAXA;
1277     DIMENSIONS NTAX=18;
1278     TAXLABELS
1279         'S. seniculus'
1280         'D. gazella'
1281         'D. bonasus'
1282         'P. ardea'
1283         'P. rufosignatus'
1284         'P. wittei'
1285         'K. signatus'
1286         'K. quadraticiceps'
1287         'K. caffrarius'
1288         'O. nuchicornis'
1289         'O. coenobita'
1290         'O. illyricus'
1291         'O. medius'
1292         'E. flavimargo'
1293         'O. nigriventris'
1294         'O. ovatus'
1295         'O. bituberculatus'
1296         'O. depressus'
1297     ;
1298 ENDBLOCK;
1299
1300 BEGIN CHARACTERS;
1301     DIMENSIONS NCHAR=35;
1302     FORMAT DATATYPE=STANDARD MISSING=? GAP=- SYMBOLS="01234";
1303     CHARLABELS
1304         [1] 'Epipharynx, the fore half till the proplegmatium'
1305         [2] 'Epipharynx, the fore margin'
1306         [3] 'Epipharynx, corypha'
1307         [4] 'Epipharynx, anterior epitorma'
1308         [5] 'Epipharynx, anterior epitorma'
1309         [6] 'Epipharynx, the fore triangular sclerotized area of
1310 haptomerum'
1311         [7] 'Epipharynx, proplegmatium placed'
1312         [8] 'Epipharynx, chaetopariae'
1313         [9] 'Epipharynx, apex of the crepis left turned and'
1314         [10] 'Epipharynx, laeotorma and dexiotorma distal part'
1315         [11] 'Epipharynx, pternotormae'
1316         [12] 'Mentum, fore margin'
1317         [13] 'Mentum, the basal margin'
1318         [14] 'Head, clypeus fore margin'
1319         [15] 'Head, genae'
1320         [16] 'Pronotum, on the whole'
1321         [17] 'Pronotum hind margin'
1322         [18] 'Pronotum, fore angles'
1323         [19] 'Legs, fore tibia'
1324         [20] 'Elytra, 7th stria'
1325         [21] 'Elytra, 8th stria'
1326         [22] 'Male genitalia, phallobase/parameres ratio'
1327         [23] 'Male genitalia, paramers'
1328         [24] 'Male genitalia, lamella copulatrix'
1329         [25] 'Male genitalia, lamella copulatrix present and'
1330         [26] 'Male genitalia, endophallus carrying apically'

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1331      [27] 'Male genitalia, raspula'
1332      [28] 'Male genitalia, FLP sclerite'
1333      [29] 'Male genitalia, SA+A sclerites'
1334      [30] 'Male genitalia, BSC sclerite'
1335      [31] 'Female genitalia, infundibular wall'
1336      [32] 'Female genitalia, vagina'
1337      [33] 'Female genitalia, infundibular tube'
1338      [34] 'Female genitalia, receptaculum seminis'
1339      [35] 'Female genitalia, receptaculum seminis at base'
1340      ;
1341      STATELABELS
1342      1
1343          'subtrapezoidal'
1344          'subrectangular'
1345          'sickle-shaped',
1346      2
1347          'rectilinear'
1348          'only slightly sinuate in the middle'
1349          'deeply and narrowly notched'
1350          'largely notched'
1351          'convex',
1352      3
1353          'absent, only a row of few and rade large setae is
1354 present along the anterior epitorma'
1355          'present, constituted by a thick tuft of short setae'
1356          'present, constituted by short and thin setae',
1357      4
1358          'never reaching the fore margin'
1359          'reaching the fore margin',
1360      5
1361          'thick'
1362          'narrow'
1363          'very narrow',
1364      6
1365          'large and lowered'
1366          'narrow and lowered'
1367          'narrow and elongate',
1368      7
1369          'in the anterior third of the epipharynx'
1370          'in the half of epipharynx surface'
1371          'in the posterior third of the epipharynx',
1372      8
1373          'subrectilinear'
1374          'angulate'
1375          'arched',
1376      9
1377          'sharp'
1378          'blunt',
1379      10
1380          'medium length, with the insertion to mandibles area
1381 drop-like'
1382          'very short and rmarkedly rounded at level of
1383 insertion to mandibles'
1384          'very thin and often elongate, with the insertion to
1385 mandibles very narrow'
1386          'short and spatulate',
1387      11
1388          'short and thick'
1389          'longer and narrower'

```

1390		'greatly reduced',
1391	12	
1392		'a large and rounded incisure'
1393		'deeply and triangular notched'
1394		'a large and triangular incisure',
1395	13	
1396		'triangular notched'
1397		'sinuate'
1398		'rectilinear',
1399	14	
1400		'not-incised '
1401		'only feebly sinuate'
1402		'deeply V-notched',
1403	15	
1404		'not especially developed'
1405		'protruding',
1406	16	
1407		'ovalar-transversal'
1408		'rounded',
1409	17	
1410		'angulate'
1411		'rounded'
1412		'straight',
1413	18	
1414		'more or less developed, blunt and always
1415		rectilinear, facing forward'
1416		'well-developed, sharp, and outward turned',
1417	19	
1418		'markedly dimorphic in the two sexes, being narrow
1419		and inward arched in male'
1420		'almost identical in both sexes, but showing
1421		differences in the tooth shape'
1422		'showing no sexual dimorphism',
1423	20	
1424		'sinuate'
1425		'rectilinear',
1426	21	
1427		'absent'
1428		'present but incomplete, and distally fused to 7th
1429		one',
1430	22	
1431		'reaching almost 2:1'
1432		'about 1:1',
1433	23	
1434		'quadrangular simmetrical, carrying laminar
1435		expansions ventrally, the apices rounded, with a digitiform expansion
1436		subapically'
1437		'simple, symmetrical, without laminar expansion on
1438		ventral side, the apices sharp but short, largely triangular-shaped'
1439		'arched, the apices large and sharp, carrying a
1440		laminar, rounded expansion subapically'
1441		'triangular-shaped ventrally, pointed at apex, and
1442		flat apically',
1443	24	
1444		'present'
1445		'absent',
1446	25	
1447		'constituted by a globose expansion and a
1448		rectilinear part, comma shaped'

1449 'well-developed, globose, C-shaped'
 1450 'cupoliform, globose, well-sclerotized',
 1451 26
 1452 'many small teeth diffused sometimes almost
 1453 inapparent'
 1454 'diffused scales, and two well-defined, ventral areas
 1455 with more tickened scales; reduced scales, but a large transversal
 1456 bent with well-developed scales',
 1457 27
 1458 'absent'
 1459 'present',
 1460 28
 1461 'laminar, double, with projections encircling SA+A'
 1462 'band-shaped, encircling the SA+A'
 1463 'large, well-developed, with evident expansions
 1464 apically and basally'
 1465 'linked to SA+A'
 1466 'well sclerotized, and pluridigitate',
 1467 29
 1468 'rod-shaped, usually separate'
 1469 'C-shaped, connected',
 1470 30
 1471 'absent'
 1472 'present, well-developed'
 1473 'present, very reduced',
 1474 31
 1475 'carrying a large longitudinal sclerotization
 1476 subrectangular or mushroom-shaped'
 1477 'supported by a thin, "arched" sclerotization'
 1478 'with a sinuate asymmetrical and folded
 1479 sclerotization'
 1480 'complex sclerotization, with two pillar-shaped
 1481 lateral sclerotizations and a key-hole central sclerotization',
 1482 32
 1483 'transversal '
 1484 'elongate and wrinkled'
 1485 'rounded'
 1486 'extremely narrow and elongate',
 1487 33
 1488 'very sclerotized, orthogonal to infundibular wall'
 1489 'non sclerotized, lowered at floccular level'
 1490 'sigmoidal, placed below the well-developed
 1491 sclerotization of the infundibular wall',
 1492 34
 1493 'tapering to apex, elongate, often the apex sharp'
 1494 'elongate, subequal on the whole lenght, the apex
 1495 slightly narrowed, but never sharp'
 1496 'short and tough, rounded at apex',
 1497 35
 1498 'cone-shaped, carrying a sclerotized portion on
 1499 infundibular tube'
 1500 'gently rounded'
 1501 'truncated',
 1502 ;
 1503 MATRIX
 1504 'S. seniculus' 0000000000 0000000000 0000000000
 1505 00000
 1506 'D. gazella' 0101100200 0001010000 0001-00211
 1507 02202

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1508          'D. bonasus'          0101100200 0001010000 0001-00211
1509 02202
1510          'P. ardea'           0201100202 0000102001 1001-00211
1511 02201
1512          'P. rufosignatus'     1201000202 0000102011 1001-00211
1513 02201
1514          'P. wittei'           1201000202 0000102011 1001-00211
1515 02201
1516          'K. signatus'         1110121111 2112010010 0021-00211
1517 02202
1518          'K. quadraticeps'     2110000111 2100010111 0021-00211
1519 02202
1520          'K. cafferarius'      2110000111 2100010111 0021-00211
1521 ??????
1522          'O. nuchicornis'       1021101210 0001000010 0010200100
1523 11010
1524          'O. coenobita'        1121101200 0101001010 0010200100
1525 11012
1526          'O. illyricus'        1201101100 1100001010 0110010400
1527 10021
1528          'O. medius'           1021101200 0101001010 0010200100
1529 10010
1530          'E. flavimargo'       1411212113 2120011010 0010111000
1531 33010
1532          'O. nigriventris'     1001001200 1101010020 0110010400
1533 10021
1534          'O. ovatus'           1121121210 0011001010 0010211100
1535 11022
1536          'O. bituberculatus'   0311211000 0202001111 0031-00302
1537 22102
1538          'O. depressus'        0111211000 0212001111 0031-00302
1539 22102
1540          ;
1541 ENDBLOCK;
1542
1543 BEGIN ASSUMPTIONS;
1544     OPTIONS DEFTYPE=UNORD POLYTCOUNT=MINSTEPS;
1545 ENDBLOCK;
1546
1547 BEGIN NOTES;
1548     [Taxon comments]
1549
1550     [Character comments]
1551
1552     [Character state comments]
1553
1554     [Attribute comments]
1555
1556     [Taxon pictures]
1557
1558     [Character pictures]
1559
1560     [Character state pictures]
1561
1562     [Attribute pictures]
1563 ENDBLOCK;
1564
1565

```

1566 **Table 3.** Pairwise distance matrix (overall average value = 0.416), in which estimates of evolutionary divergence between sequences were
 1567 conducted using the Kimura 2-parameter model. See table 1 for the acronyms.

	SEN	GAZ_1	GAZ_2	ARD	RUF_1	RUF_2	RUF_3	SIG_1	SIG_2	NUC	COE	ILL_1	ILL_2	MED	FLA_1	FLA_2	NIG	OVA	INT_1	INT_2	DEP	
SEN																						
GAZ_1	0,150																					
GAZ_2	0,208	0,081																				
ARD	0,130	0,119	0,186																			
RUF_1	0,128	0,132	0,201	0,125																		
RUF_2	0,123	0,130	0,196	0,125	0,017																	
RUF_3	0,123	0,134	0,201	0,130	0,017	0,011																
SIG_1	0,126	0,124	0,175	0,130	0,160	0,148	0,153															
SIG_2	0,128	0,129	0,175	0,142	0,157	0,150	0,155	0,013														
NUC	0,677	0,761	0,814	0,721	0,722	0,700	0,700	0,733	0,699													
COE	0,697	0,752	0,828	0,764	0,741	0,724	0,719	0,735	0,711	0,100												
ILL_1	0,734	0,768	0,844	0,726	0,745	0,712	0,706	0,765	0,746	0,110	0,130											
ILL_2	0,734	0,768	0,844	0,726	0,745	0,712	0,706	0,765	0,746	0,110	0,130	0,000										
MED	0,702	0,795	0,864	0,753	0,759	0,736	0,731	0,764	0,728	0,081	0,117	0,143	0,143									
FLA_1	0,146	0,171	0,221	0,176	0,162	0,153	0,153	0,152	0,155	0,697	0,715	0,732	0,732	0,739								
FLA_2	0,141	0,160	0,226	0,164	0,141	0,141	0,137	0,162	0,164	0,677	0,696	0,706	0,706	0,728	0,026							
NIG	0,781	0,869	0,923	0,795	0,804	0,791	0,785	0,849	0,820	0,145	0,164	0,173	0,173	0,178	0,798	0,769						
OVA	0,677	0,747	0,839	0,718	0,705	0,684	0,674	0,715	0,703	0,091	0,106	0,119	0,119	0,121	0,701	0,676	0,177					
INT_1	0,126	0,169	0,216	0,132	0,148	0,137	0,137	0,127	0,126	0,601	0,657	0,657	0,657	0,653	0,126	0,128	0,732	0,620				
INT_2	0,126	0,164	0,211	0,130	0,146	0,134	0,134	0,127	0,126	0,601	0,653	0,653	0,653	0,653	0,124	0,130	0,743	0,620	0,007			
DEP	0,152	0,163	0,205	0,161	0,156	0,150	0,143	0,173	0,175	0,713	0,751	0,741	0,741	0,754	0,157	0,150	0,807	0,713	0,143	0,136		

1569

1570 **Table 4.** Results of the first CVA in which the major groups classification was
 1571 confirmed, since 100.0% of original grouped cases were correctly classified, and after
 1572 the cross validation the 98.8% of cross-validated grouped cases were correctly
 1573 classified.

1574

Classification Results							
code group		Predicted Group Membership					Total
		1	2	3	4		
Original	Count	1	62	0	0	0	62
		2	0	2	0	0	2
		3	0	0	2	0	2
		4	0	0	0	18	18
	%	1	100,0	0,0	0,0	0,0	100,0
		2	0,0	100,0	0,0	0,0	100,0
		3	0,0	0,0	100,0	0,0	100,0
		4	0,0	0,0	0,0	100,0	100,0
Cross-validated	Count	1	62	0	0	0	62
		2	0	2	0	0	2
		3	0	0	2	0	2
		4	1	0	0	17	18
	%	1	100,0	0,0	0,0	0,0	100,0
		2	0,0	100,0	0,0	0,0	100,0
		3	0,0	0,0	100,0	0,0	100,0
		4	5,6	0,0	0,0	94,4	100,0

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1576

1577

1578

1579 **Table 5.** Results of the second CVA in which the genera classification within the
 1580 *Phalops* complex was confirmed, since 100.0% of original grouped cases were correctly
 1581 classified, and after the cross validation the 98.4% of cross-validated grouped cases
 1582 were correctly classified.

Classification Results						
code genus		Predicted Group Membership			Total	
		1	2	3		
Original	Count	1	37	0	0	37
		2	0	12	0	12
		3	0	0	13	13
	%	1	100,0	0,0	0,0	100,0
		2	0,0	100,0	0,0	100,0
		3	0,0	0,0	100,0	100,0
Cross-validated	Count	1	37	0	0	37
		2	0	12	0	12
		3	0	1	12	13
	%	1	100,0	0,0	0,0	100,0
		2	0,0	100,0	0,0	100,0
		3	0,0	7,7	92,3	100,0

1583

1584