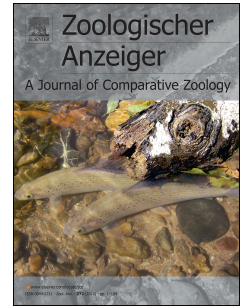


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The Genus *Camelobaetidius* Demoulin (Ephemeroptera: Baetidae) in America: Phylogenetic and Biogeographic analyses

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1 **The Genus *Camelobaetidius* Demoulin (Ephemeroptera: Baetidae) in**
2 **America: Phylogenic and Biogeographic analyses**

3
4 Phylogenic and Biogeographic analyses of *Camelobaetidius*

5
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31 **Abstract**

32 The genus *Camelobaetidius* is the second most diverse genus of the family Baetidae in
33 America. Also, two other genera were described as closely related to *Camelobaetidius*:
34 *Corinnella*, and *Tapajobaetis*. A peculiar spatulate tarsal claw is the character shared
35 among them. We used TNT (Tree analysis using New Technology) and GEM
36 (Geographically explicit Event Model) to recover the phylogenetic and biogeographic history
37 of this genus. A matrix of 42 taxa and 77 morphological characters was constructed, of
38 which 68 were treated as discrete and nonadditive (unordered), and eight as continuous. All
39 available records of *Camelobaetidius* plus *Corinnella*, *Tapajobaetis*, and the species's
40 outgroup were included in the biogeographic analysis. *Corinnella* and *Tapajobaetis* were
41 obtained into *Camelobaetidius*. The founder events were the most frequent in this genus,
42 followed by vicariance, sympatry and point sympatry. The long-distance dispersal ability in
43 mayfly seems to be the rule instead of the exception in the history of this clade.

44
45 **Keyword.** Mayfly, Neotropical region, Cladistics, Biogeography, Founder Event, Long
46 Distance Dispersal.

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59 1.1. Introduction

60 The genus *Camelobaetidius* Demoulin is the second most diverse genus of the family
61 Baetidae (Ephemeroptera) in America. Demoulin described it in 1966 and, since then, the
62 description of new species has not stopped. Although it is distributed between Canada in
63 the North (Lehmkuhl, 1976) to Argentina in the South (Traver and Edmunds, 1968; Nieto,
64 2003), the vast majority of the species are distributed in the Neotropical region.

65 *Camelobaetidius* has 45 species described, of which 26 were described from nymphs, five
66 from imagoes, 13 from nymphs and male imago and one from nymph and female imago
67 (Jacobus and McCafferty, 2005; Salles and Serrão, 2005; Domínguez et al., 2006; Boldrini
68 et al., 2012a, b; Boldrini et al., 2013; Boldrini and Pes, 2014; Sibajaya-Araya and Esquivel,
69 2018; Salinas-Jimenez et al., 2019). *Dactylobaetis* described by Traver and Edmunds in
70 1968, was proposed as a sister group of *Camelobaetidius* (Traver and Edmunds, 1968).
71 However, it was later synonymized with *Camelobaetidius* in 1990 by McCafferty and
72 Waltz.

73 *Camelobaetidius* is common in lotic habitats, above rocks in areas of moderate to fast
74 current. The nymphs have a particular powerful grasping claw (Berner and Pescador,
75 1988). Boldrini et al. (2012) proposed the spatulate claw was used for adhering to substrate
76 in areas of the strong current, rather than as a tool for removing food particles as was
77 suggested by Baptista et al. (2006). There are a few species with gut content analyzed.

78 *Camelobaetidius mexicanus* (Traver and Edmunds) was defined as scraper (Wagner, 1996)
79 consuming diatoms and filamentous algae. Also, the genus was reported as detritivore with
80 a high percentage of FPOM (Fine Particulate Organic Matter) and low content of algae in
81 its gut (Granados-Martínez et al., 2016). Related to the life cycle, *Camelobaetidius* was
82 reported as multivoltine, species such as *C. penai* (Traver and Edmunds) exhibited a
83 bivoltine life cycle (Corigliano et al., 2008), while *C. variabilis* had five generations per
84 year (Perry and Kennedy, 2016).

85 Boldrini and Salles (2009) proposed that the species of *Camelobaetidius* could be divided
86 into three morphological groups: the first one with the terminal filament reduced and with a
87 projection on the inner margin of the fore femora; the second with the terminal filament
88 reduced and without a projection on the inner margin of the fore femora and the last one
89 with the terminal filament almost as long as cerci. Also, two other genera were described as

90 closely related to *Camelobaetidius*, *Corinnella* Thomas and Dominique described from
91 French Guiana (Nieto, 2010) and later reported from Brazil (Boldrini et al., 2013), and
92 *Tapajobaetis* Boldrini and Salles described from Brazil (Boldrini et al., 2017).
93 The knowledge of biogeographic history of the genus is scarce, almost null. McCafferty et
94 al. (1992) sustained that *Camelobaetidius* may have had a recent Neotropical center of
95 dispersal because “it follows the pattern of having an arid-favored distribution in the
96 Nearctic and being warm-water sublimited”.

97 In this paper, we proposed cladistic and biogeographic analyzes of *Camelobaetidius* in
98 order to improve the knowledge of this diverse and widely distributed genus throughout
99 America. The main object was to test the monophyly of *Camelobaetidius* plus *Corinnella*
100 and *Tapajobaetis*. Additionally, we reconstructed the biogeographic pattern of the genus,
101 evaluating which events influenced the distribution and speciation of this clade. Also, we
102 evaluated the three morphological groups proposed by Boldrini and Salles (2009) and we
103 analyzed the evolution of different characters.

105 2. Material and Methods

106 Intensive fieldwork was carried out collecting different rivers along with South America in
107 order to collect fresh specimens of the different species. The nymphs were collected using
108 traditional methods, with a D-frame net (Domínguez & Fernandez, 2009). All the material
109 was conserved in 96% alcohol. For morphology, mature nymphs was selected and dissected
110 where possible. The material examined is housed in the following institutions: Instituto de
111 Biodiversidad Neotropical (IBN), Tucuman, Argentina; Universidade Federal de Roraima
112 (UFRR), Universidade Federal de Viçosa (UFVB), and Instituto Nacional de Pesquisas da
113 Amazônia (INPA), Brazil. Additional sources included systemactic studies performed on
114 material from other collections: Florida A & M University (FAMU), Tallahassee; Colorado
115 State University (CSU); and Texas A&M University (TA&M), USA.

116 2.1. Taxa

117 A matrix of 42 taxa and 77 morphological characters was constructed (Appendix S1). 37
118 species were included in the ingroup: two of which belong to *Corinnella*: *C. eximia* and *C.*
119 *thomasi*, one to *Tapajobaetis*: *T. labiosus* and 34 to *Camelobaetidius*, representing 75% of
120 the total species. We selected the species better described, with the specimens available,

121 and we omitted species only described from adults. Five species were included as out-
122 group: *Centroptilum luteolum*, *Harpagobaetis gulosus*, *Nanomis galera*, *Americabaetis*
123 *longetron*, *Spiritiops silvudus*, all of them belong to Baetidae. We chose genera belonging
124 to different clade in the family Baetidae, following Cruz et al. 2020, from genus very closer
125 to *Camelobaetidius* such as *N. galera*, to lesser close such as *C. luteolum*. The tree was
126 rooted with *C. luteolum*. Although the result did not change with other taxa as the
127 outgroups.

128

129 **2.2. Characters**

130 All characters in this study were derived from the external morphology of nymphs. Sixty
131 characters were binary, 17 were multistate, 68 characters were treated as discrete and
132 nonadditive (unordered), and 8 as continuous (Appendix S2 and S3).

133

134 **2.3. Phylogenetic Analysis**

135 The matrix was analysed using TNT (Tree analysis using New Technology, Goloboff et al.,
136 2003a), under extended implied weights (Goloboff, 2014) (TNT default) following the
137 script published by Mirande et al. (2013). The analyses provide 21 values of K and their
138 respective topologies. For each K-value, under extended implied weighting, extra searches
139 were carried out using parsimony ratchet (Nixon, 1999), sectorial searches, tree fusing, and
140 tree drifting (Goloboff, 1999) until the best fit was obtained three times, all using TNT
141 default. Trees obtained in each K-value, were reduced to a strict consensus (if more than
142 one); the result of these searches were three different trees. Mirande (2009, 2010) and
143 Mirande et al. (2011, 2013) proposed a measure of stability as a criterion to choose the
144 most stable topologies under different parameters. The distortion coefficient (“tcomp”)
145 (Farris, 1989) was used as a measure of stability and later a strict consensus of the most
146 stable solutions was calculated.

147 The strict consensus trees obtained in each K-value (21) were saved in TNT memory
148 (RAM) and distortion coefficient was found using the following scripts.

149

(i)

```
macro=;  
loop 0 20
```

```
loop 0 20
      tcomp= #1 #2
stop;
stop;
proc/;
```

150 The best average of “tcomp” was selected to compose the set of topologies (Table 1).
151 Group support was obtained by applying three estimators, absolute and relative Bremer
152 supports (Bremer, 1988, 1994) and frequency differences (Goloboff et al., 2003b). Bremer
153 supports were calculated for the most-parsimonious trees and 15 000 suboptimal trees up to
154 ten steps longer. Suboptimal trees were distributed in 12 stages: 0.1, 0.2, 0.3, 0.4, 0.5, 0.6,
155 0.7, 1, 2, 3, 4, 5, 6, 7 and 10 units of fit, saving 1000 trees at each stage. Frequency
156 differences were calculated with 500 replicates of jackknifing (Farris et al., 1996) by
157 symmetrical resampling of the original matrix (each character presents the same probability
158 of being positively or negatively reweighted, Goloboff et al., 2003b). Each replicate of
159 jackknifing was calculated by ten random addition sequences plus tree bisection–
160 reconnection, saving ten trees per replicate. After obtaining the shortest trees, the value of
161 homoplasy for each character was calculated (adjusted homoplasy). Characters with low
162 homoplasy present values near 0.00, whereas characters with high homoplasy have values
163 near 1.00 (some are mentioned in the results).

164

165 **2.3. Biogeographic Analysis**

166 All available records of the species of *Camelobaetidius* included in the matrix, plus
167 *Corinnella*, *Tapajobaetis*, and the species’s outgroup were mapped with the free software
168 QGIS, Geographic Information System (<http://qgis.org>). Species distribution have been
169 plotted and overlapped with layer of political subdivisions of the world and the ecoregions
170 proposed by Olson et al., 2001.

171 To infer the particular biogeographic history of *Camelobaetidius*, a biogeographic analysis
172 was carried out using Geographically explicit Event Model (GEM, Arias, 2017). This
173 phylogenetic biogeographic method does not assume predefined areas or a hierarchical
174 relationship between them. The primary purpose of this method is to discover cladogenetic
175 events such as vicariance, sympatry, point sympatry and founder event among sister
176 groups. Distributions were represented as absence/presence data in a raster grid with pixels

177 of $1^\circ \times 1^\circ$ degrees, with a filling of 2. The cost of the four cladogenetic events was set to
178 one. To penalize large ancestral ranges a $Z=50$ was used. The search was made with the
179 flipping algorithm applying 1000 flip replicates.

180

181 **3. Results**

182 **3.1. Phylogenetic Analysis**

183 An unique tree was obtained in a range of value of K, from 1.71 to 3.63, and with a length
184 of 243.27 steps (fit = 47.756). The tree ranging from these values of K were also the most
185 stable according to the measure of tcomp. This tree was proposed as the final hypothesis.
186 The shortest tree obtained with *C. luteolum* as the outgroup recovered the genus *Corinnella*
187 and *Tapajobaetis* into *Camelobaetidius* (Fig. 1). Five synapomorphies (Appendix S4)
188 sustained this result (Node A): length of tibia/ length of femur, 0.944-0.956 (character 6);
189 frontoclypeal suture is Y shaped (character 8); left mandible with the subtriangular process
190 elevated (character 26); articulation of the tibia absent (character 64); and tarsal claws
191 spatulated (character 67). The analysis recovered three main groups of *Camelobaetidius*
192 (Nodes B, C, D). Node B included 16 species and it is sustained by three synapomorphies
193 (Appendix S4): relationship in the left mandible between length of mola and length
194 between triangular process and incisor, 1.069 (character 1); number of denticles in the
195 tarsal claws, 0.009 (character 7) and first denticle of the tarsal claw distinctly larger than
196 the other (character 68). In this clade, five groups were recovered with good support (Node
197 E, F, G, H, I). Node B confirmed the synonymy of *Dactylobaetis* under *Camelobaetidius*,
198 because this clade recovered the type species of both nominal genus *C. warreni* (Traver and
199 Edmunds) and *C. leentvari* Demoulin, respectively. Node F, sustained by four
200 synapomorphies, grouped all the species with the terminal filament reduced, about the size
201 of the 10th abdominal segment (character 76). However, Node G is nested to this group.
202 Node G recovered the species with a tubercle on the inner margin of the femur (character
203 61).
204 Node C recovered seven species: the two species of *Corinnella*, the single species of
205 *Tapajobaetis*, and four species of *Camelobaetidius*, and six synapomorphies supported it:
206 relationship in the left mandible between length of mola and length between triangular
207 process and incisor, 0.700-0.762 (character 1); relationship in the maxillary palp between

208 length of palp and length of galea-lacinia, 1.418-1.638 (character 2); shape of incisors of
 209 left mandible not same level (character 19); teeth at apex of the crown long and slender
 210 (character 36); outer margin of maxilla convex (character 37); development of base of
 211 glossa of the labium not overlapping (character 41). The two species of *Corinnella*, were
 212 sustained by ten synapomorphies including the following: maxillary palp bifid (character
 213 33), galea of maxilla robust (character 38), setae on surface of labial palp many and long
 214 (character 49), row of setae subparallel of the patelo-tibial suture present (character 65).
 215 *Tapajobaetis* plus *C.ortizi*, *C.carolinae*, *C.cruzi* and *C. matilei* are supported by two
 216 synapomorphies: incisive of maxilla with setae on the apex (character 34) and outer margin
 217 of paraglossa curved (character 54).
 218 Node D grouped 14 species is sustained by 4 synapomorphies including the following:
 219 number of setae on outer margin of femora, 0.050 (character 4), relationship between length
 220 of femur and width of 1/3 basal of femora, 3.262-3.422 (character 5), row of setae between
 221 molar and incisors of left mandible absent (character 24), and segment III of labial palp
 222 rounded (character 50).

223

224 **3.2. Systematic**

225 *Camelobaetidius* Demoulin, 1996: 9; McCafferty and Waltz, 1990: 777; Lugo-Ortiz and
 226 McCafferty, 1999, 258; Dominique et al., 2000: 40; McCafferty and Randolph, 2000: 259;
 227 Dominique et al., 2001: 18; Dominique and Thomas, 2001: 27; Nieto, 2002: 57; Nieto,
 228 2003: 233; Salles and Serrão, 2005: 267; Jacobus and McCafferty, 2005: 153; Domínguez
 229 et al., 2006: 118; Boldrini et al., 2012b: 2033.

230 (Type-Species: *Camelobaetidius leentvaari* Demoulin, original description)

231 *Dactylobaetis* McCafferty and Waltz, 1990: 777.

232 *Corinnella* (Thomas and Dominique, 2006) SYN. NOV.

233 *Tapajobaetis* Boldrini and Salles, 2017. SYN. NOV.

234 *Diagnosis.* 1) labrum with dorsolateral arc of setae; 2) incisors of mandibles fused; 3)
 235 maxillary palp segment II bifid or simple; 4) segment II of labial palp with or without
 236 distomedial projection; 5) thoracic gill present or absent at base of forecoxa; 6) hind wing
 237 pads usually present (absent in few species); 7) tarsal claws spatulate; 8) tarsal claws with

238 6–140 denticles; 9) paraproct with pointed or rounded marginal spines; 10) abdominal gills
 239 present on segments I–VII; 11) terminal filament as long as cerci or reduced.

240

241 Included species: **Species described based on nymphs:** *C. baumgardneri* Boldrini, 2014;
 242 *C. edmundsi* Dominique, Thomas and Mathuriau, 2002; *C. eximia* (Thomas and
 243 Dominique, 2006), **new combination**; *C. guaycara* Sibaja-Araya and Esquivel, 2018; *C.*
 244 *hamadae* Salles and Serrão, 2005; *C. ipaye* Nieto, 2003; *C. juparana* Boldrini and Salles,
 245 2012; *C. kondratieffi* Lugo-Ortiz and McCafferty, 1995; *C. kickapoo* McCafferty, 2000; *C.*
 246 *labiosus* (Boldrini and Salles, 2017), **new combination**; *C. lassance* Salles and Serrão,
 247 2005; *C. leentvaari* Demoulin, 1966; *C. lourii* (Salles, Victoriano, Boldrini and Cabette,
 248 2017), **new combination**; *C. maidu* Jacobus and McCafferty, 2005; *C. maranhensis* Salles
 249 and Serrão, 2005; *C. mathuriae* Dominique and Thomas, 2001; *C. metae* Salinas-Jimenez
 250 and Boldrini, 2019; *C. musseri* (Traver and Edmunds, 1968); *C. patricki* Dominique and
 251 Thomas, 2001; *C. rufiventris* Boldrini and Salles, 2009; *C. sallesi* Boldrini, 2014; *C.*
 252 *serapis* (Traver and Edmunds, 1968); *C. shepardi* Randolph and McCafferty, 2001; *C.*
 253 *spinus* Boldrini and Salles, 2012; *C. tepequensis* Boldrini, 2014; *C. tuberosus* Lugo-ortiz
 254 and McCafferty, 1999; *C. variabilis* Wiersema, 1998; . **Species described based on**
 255 **nymphs and adults:** *C. anubis* (Traver and Edmunds, 1968); *C. billi* Thomas and
 256 Dominique, 2001; *C. cayumba* (Traver and Edmunds, 1968); *C. francischettii* Salles,
 257 Andrade and Da-Silva, 2005; *C. huarpe* Nieto, 2003; *C. janae* Dominique and Thomas,
 258 2001; *C. mexicanus* (Traver and Edmunds, 1968); *C. penai* (Traver and Edmunds, 1968);
 259 *C. phaedruss* (Traver and Edmunds, 1968); *C. suapi* Nieto, 2002; *C. warreni* (Traver and
 260 Edmunds, 1968); *C. waltzi* McCafferty, 1994; *C. yacutinga* Nieto, 2003; **Species described**
 261 **based on adults:** *C. alcyoneus* (Traver, 1943); *C. aneto* (Traver, 1971); *C. arriaga* (Traver
 262 and Edmunds, 1968); *C. dryops* (Needham and Murphy, 1924); *C. tantillus* (Needham and
 263 Murphy, 1924); *C. thomasi* (Boldrini and Salles, 2013), **new combination**.

264

265 3.3. Biogeography

266 The search with GEM found one reconstruction with a cost of 226.820. The events
 267 recovered were: eight vicariance events, eight sympatry events, four point sympatry, and 21
 268 founder events (Fig. 2). All ancestral areas assignments are available in Figs. S5. The

269 founder events occurred more frequently in the deep clades meanwhile vicariance in the
270 apical branches. Sympatry and point sympatry appeared both at the base of the tree and at
271 the terminals. Also, with a few exceptions, the nodes with the best supports in the
272 phylogeny analysis were recovered as vicariants in the biogeography analysis. For the
273 origin of the clade “*Camelobaetidius*” (Node 3), the reconstruction found a sympatric
274 event. The first event in the genus was the founder event from Southeastern Brazil to
275 northeastern South America (Node 5, Fig. S5 6). Descendants of Node 6 arose in a
276 sympatry event in northeastern South America (Fig. S5 7). Related to Node 7 all its
277 terminals were restricted to the Guiana Highlands and Amazon ecoregions, and both the
278 founder events as well as point sympatry were the most frequent events recovered. In Node
279 13 a sympatry event occurred between the northeastern and central South America (Fig. S5
280 14). From this area occurred different founder events to the south of South America (Node
281 15 and 17, Figs. S5 16-18) as well as to the north of South and Central America (Node 18
282 and 19 Figs. S5 19-20). Node 26 was characterized by founder events, from southeastern
283 Brazil to North and Central America (Fig. S5 27). Node 31 presented a vicariant event in
284 the southeastern Brazil, specifically in Atlantic Forest ecoregion (Fig. S5 32), between the
285 north and south. Finally, in the rest of the clade, distinct sympatry events followed by some
286 vicariant events around the north and central of South America were recovered.

287

288 **4. Discussion**

289 **4.1. Phylogenetic Analysis**

290 *Evolution of selected characters*

291 In 1968, when Traver and Edmunds described *Dactylobaetis*, they proposed that the
292 spatulated claws were of 2 types representing two separate phyletic lines: *Camelobaetidius*
293 and *Dactylobaetis*. However, they did not explain the difference between them, both genera
294 were distinguished by other characters instead of the claw. *Corinnella* was established
295 based on the tarsal claws with few denticles but spatulated (Thomas and Dominique, 2006).
296 *Tapajobaetis* was described by having spatulated claws with more than 100 denticles, along
297 with other characteristics related to mouthpart morphology (Boldrini et al., 2017). Here, in
298 our analysis, the character "tarsal claws spatulated" was recovered as a synapomorphy of

299 Node A with the best adjusted homoplasy, including *Corinella*, *Tapajobaetis* and all the
300 species of *Camelobaetidius* considered in the analysis.

301 The labial palp, especially the second segment, presents a high variability within the genus.
302 Although this structure has many forms in the family, most of this variation is found among
303 different genera (and not so often among species of the same genus). However, in
304 *Camelobaetidius*, the inner margin of the second segment of the labial palpi can be slightly
305 protruded (instead of parallel to outer margin, the inner margin is gently curved) to strongly
306 protruded (in these cases a distinct distomedial projection is present) (Nieto, 2003, Salles
307 and Serrão, 2005), with an enormous range of forms. The character 46: projection of the
308 segment II of the labial palp had the best adjusted homoplasy. However, a morphometric
309 analysis could be better to approach this structure.

310 The length of the terminal filament was considered to distinguish *Camelobaetidius* from
311 *Dactylobaetis* (Traver and Edmunds, 1968), the first one had terminal filament short, while
312 *Dactylobaetis* had terminal filament long. In our analysis, both states of this character
313 were not recovered as synapomorphies of these two groups (character 76). The state
314 "terminal filament subequal in length to cerci" was recovered at the base of the tree, being
315 plesiomorphic for the species of *Camelobaetidius*. Then, in the clade containing *C.*
316 *maranhensis*, *C. billi*, *C. rufiventris*, *C. tepequensis*, *C. ipaye*, *C. francishettii*, and *C.*
317 *leentvaari*, this character changed to short or reduced in length and, thus, is considered a
318 synapomorphy of these species, in coincidence with Boldrini and Salles (2009). Among
319 them, a reversal has occurred in *C. tepequensis*, once the terminal filament is subequal in
320 length to cerci in this species.

321 Many authors have analyzed the accessory gills (maxillary, coxal, and labial gills) if these
322 structures were homologous or not and synapomorphies of a specific clade (Zhou, 2010,
323 Staniczek, 2010). They have been considered as accessory gills, although their respiratory
324 function has not been experimentally confirmed (Staniczek, 2010). In *Camelobaetidius* it is
325 most often present at the base of foreleg (character 57), but also can be found at the base of
326 the second segment of the maxillary palp (*C. jane*, *C. thomasi* and *C. lourii*). At the base of
327 foreleg, based on our analysis, it had two independent acquisitions in the tree, at *C. warreni*
328 and at *C. anubis*+*C. yacutinga*. In two related species, *C. hamadae* and *C. juparana*, these
329 accessory gills are bifid and relatively long. Among the other species they are always

330 simple, and the length is highly variable. However, it is still necessary to map this character
331 and the other similar structures in a large phylogeny of the order.

332 **4.2. Biogeography**

333 *Camelobaetidius* presents a wide distribution along continental America, including a wide
334 variety of climates, however at high altitude, above 2000 m.a.s.l. its presence is minimal or
335 null. The Andean mountain seems to be a barrier instead of the route to diversification.

336 Vicariance was obtained generally in nodes with good support. Many authors have
337 discussed the speciation and adaptive evolution related to vicariant or sympatry events
338 (Brooks and McLennan, 1993, Friesen and Anderson, 1997). Some authors sustained that
339 sympatric speciation involves the adaptive process; meanwhile, vicariant speciation
340 involves the nonadaptive process (Losos and Glor, 2003). The adaptive process involves
341 ecological opportunity, such as dispersal into new territory, extinction of competitors or
342 change in behavior, while the nonadaptive process does not require specific adaptations for
343 speciation to occur (Brooks and McLennan, 1993). Our results showed that the vicariance,
344 worked on the speciation process promoting derived characters or synapomorphies.

345 The founder events were the most common in this genus. In these events, a small number of
346 individuals, sometimes parthenogenetic female or male, take part in a rare, long-distance
347 colonization (Funk et al., 2010), and this population is very rapidly isolated from the
348 ancestral population (Matzke, 2014). This event is an essential mode of lineage splitting
349 and of moving taxa around the planet (Heads, 2012; Matzke, 2014). Mayfly dispersal had
350 been thought to be very limited (Brittain, 1982), until new studies confirmed the long-
351 distance dispersal ability in mayfly species on remote islands such as the Azores (Brinck
352 and Scherer, 1961; Raposeiro et al., 2012), transoceanic dispersal between Madagascar and
353 continental Africa (Monaghan et al., 2005; Vuataz et al., 2013) on the Canary Island and
354 Madeira (Rutschmann et al., 2014), on the Macaronesian archipelago (Rutschmann et al.,
355 2017). In this analysis, we recovered founder events inside South America, sometimes
356 including long distances, such as between Cerrado Ecoregion and Colombia (Node 16), or
357 Cerrado ecoregion and San Juan Province in Argentina (Node 17) or even Cerrado and
358 Costa Rica and Guatemala (Node 20).

359 McCafferty et al. (1992) and McCafferty (1998) sustained that *Camelobaetidius* had a
360 recent Neotropical center of dispersal, specifically South America. We obtained the

361 Amazon ecoregion as the area with the majority founder, vicariance, and sympatry events.
362 This ecoregion was proposed as a refuge area based on the idea that climatic oscillation
363 occurring during the Pleistocene promoted allopatric speciation, different patches of the
364 selve remained isolated. (Haffer 1969, 2008). The theory proposed that during dry climatic
365 periods, humid rainforest blocks remained isolated and separated by dry forest or other
366 intermediate vegetation and this fact promoted the speciation (Haffer 1969, 2008). As in
367 other taxa, the interaction of paleogeographical and paleoclimatic forces would seem to be
368 the cause of the complex processes of diversification in South America (Bush, 1994;
369 Riddle, 1996; Rull, 2008).

370 In coincidence with our results, species of the genus *Asthenopodes* (Polymitarcynae,
371 Polymitarcydae) showed a similar pattern of speciation in the Amazon region (Molineri et
372 al., 2015). The authors sustained that “without a molecular dating of these nodes they can
373 only speculate possible explanations related to Pleistocene refugia”. Later, Molineri et al.
374 (2019) recovered for this region several traceable vicariance events (TVEs, Hovenkamp
375 1997) for species of *Campsurus*, *Homothraulius*, *Callibaetis* and *Caenis*. They reinforced
376 the idea of Amorin and Pires (1996) as this region such as an area of endemism, and a
377 transitional zone between tropical and temperate climates.

378 The biogeographic history of *Camelobaetidius* has resulted in a complex history of plenty
379 of dispersal events or founder events. In that sense, the use of a Geographically explicit
380 Event Model seems to be the appropriate methodology to recover these patterns since other
381 methods, such as Diva, Lagrange, or Dec, fail to take founder event into account (Matzke,
382 2014). The fact to use explicit distributions instead of the predefined area helped us to
383 identify these events inside South America.

384

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576 Figure Legends

577 Fig. 1. The shortest tree obtained with a length of 243.27 steps. The numbers above the
578 nodes indicate frequency differences; unnumbered nodes have support less than 25. The
579 numbers below the nodes indicate absolute, and relative Bremer supports.

580

581 Fig. 2. The optimal reconstructions found for the geographical data. The white triangle
582 represents founder event; white squares represent sympatry, the black square represents
583 vicariance, the white circle represents point sympatry. All ancestral node reconstructions
584 see Appendix S5.

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586 Supporting Information

587 S1. Matrix.

588 S2. Characters and character states.

589 S3. Figures of characters.

590 S4. List of synapomorphies common to the shortest tree.

591 S5. Maps with ancestral reconstructions.

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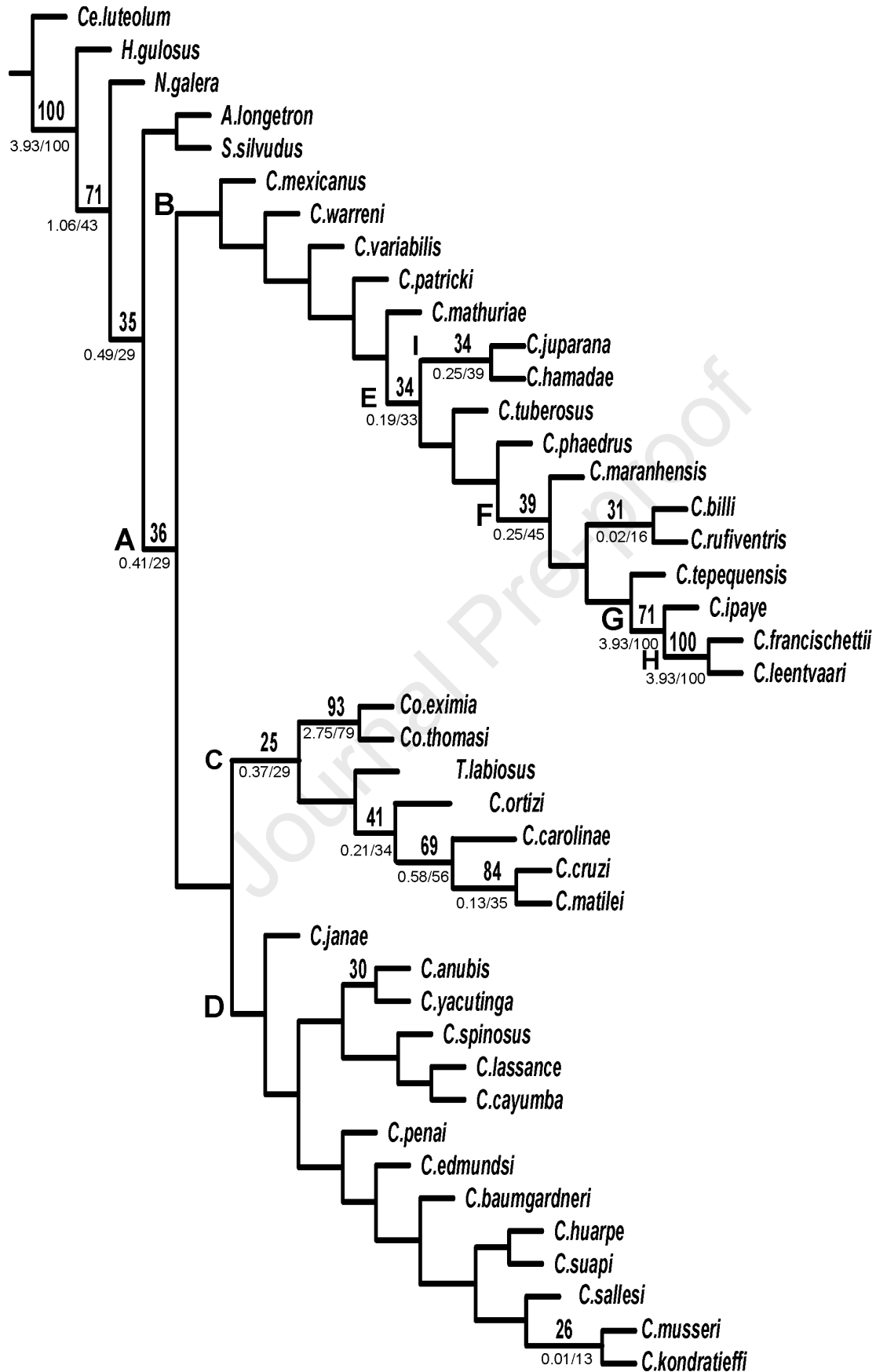
604

605 **Table 1.** Distortion coefficient (tcomp) under different K_values, and number of shortest
 606 trees obtained.

607

Reference	K-value	Trees	Tcomp
		1	
0	1.709284		0.9711982
		1	
1	1.851724		0.9711982
2	2.006551	1	0.9711982
3	2.175452	1	0.9711982
4	2.36044	1	0.9711982
5	2.563926	1	0.9711982
6	2.788832	1	0.9711982
7	3.038727	1	0.9711982
8	3.318022	1	0.9711982
9	3.632229	1	0.9711982
10	3.988329	1	0.9700462
11	4.395302	1	0.9700462
12	4.864885	1	0.9700462
13	5.412733	1	0.9700462
14	6.060189	1	0.9700462
15	6.837136	1	0.9306355
16	7.786738	1	0.9306355
17	8.973741	1	0.9306355
18	10.499887	1	0.9306355
19	12.534749	1	0.9306355
20	15.383556	1	0.9306355

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Declaration of interests

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests:

Journal Pre-proof