Evolution, mitochondrial DNA phylogeny and systematic position of the Macaronesian endemic Hydrotarsus Falkenström (Coleoptera: Dytiscidae)

IGNACIO RIBERA 1, DAVID T. BILTON 2, MICHAEL BALKE 1 and LARS HENDRICH 3

1Department of Entomology, The Natural History Museum, London, U.K., 2School of Biological Sciences, University of Plymouth, Drake Circus, Plymouth, U.K. and 3Mörchinger Strasse 115A, 14169 Berlin, Germany

Abstract. Hydrotarsus Falkenström (Coleoptera, Dytiscidae) is the only genus of aquatic beetle considered to be endemic to the Atlantic islands of Macaronesia. Its three known species (H. lundbladi Falkenström from Madeira, and H. compunctus (Wollaston) and H. pilosus Guignot from the Canary Islands) are revised, and their phylogenetic position studied based on sequences of the 16S rRNA and Cytochrome Oxidase I genes of mitochondrial DNA. Our results clearly indicate that the species of Hydrotarsus fall deep within the genus Hydroporus Clairville, in a clade formed by (in addition to Hydrotarsus) species currently included in the H. fuscipennis, marginatus, nigrita and tessellatus groups, and in consequence a new synonymy is proposed: Hydrotarsus Falkenström = Hydroporus Clairville syn.n. The origin of the species of ‘Hydrotarsus’, based on molecular clock estimations, is late Miocene, relatively recent in the history of the Atlantic islands. They are therefore not palaeoendemics but highly derived, recently evolved elements in the Macaronesian fauna. The estimated ages of divergence among species are much more recent than the emergence of any of the islands on which they are found, suggesting rare long-range dispersal as the mechanism driving the diversification of the lineage. Morphological modifications of the species of ‘Hydrotarsus’ are discussed, as well as those of other dytiscids from hygropetric habitats.

Introduction

The islands of the central North Atlantic, particularly those belonging to the Macaronesian subregion (Madeira, Salvages, Canary Islands, Cape Verde), are well known for their endemic radiations of invertebrates (Kunkel, 1976; Juan et al., 2000). Within the Coleoptera, intra-island and intra-archipelago radiations are characteristic of the fauna of Madeira and the Canary Islands (e.g. Wollaston, 1865; Machado, 1992; Machado & Oromi, 2000), many of these radiations being endemic to single islands or archipelagos at the generic level. However, amongst the aquatic Coleoptera only a single genus of Dytiscidae is currently recognized as being endemic to Macaronesia: Hydrotarsus Falkenström, which currently includes three species, two of them endemic to the western Canary Islands (H. compunctus (Wollaston) and H. pilosus Guignot), and H. lundbladi Falkenström endemic to the main island of Madeira (Machado, 1987; Alarie & Bilton, 2001) (Fig. 1).

All three species of Hydrotarsus are specialist madicolous (see Balke et al., 1997), being restricted as both larvae and adults to hygropetric habitats (running water films or small springs over exposed bedrock), which probably explains why they have been rarely collected by entomologists. In fact, the true habitat of the species was only discovered as recently as 1986 during a survey of Madeiran water beetles (Balke & Hendrich, 1989). The scarcity of Hydrotarsus species in collections has hampered adequate descriptions of the taxa, and made identification of specimens difficult in the past (Balke et al., 1990). In addition, the phylogenetic
placement of this group of species has been highly problematic. The genus belongs to the tribe Hydroporini Sharp, which currently includes 36 genera worldwide (Nilsson, 2001). Along with Necterosoma MacLeay and Sternopriscus Sharp, species of Hydrotarsus are characterized by pentamerous pro- and mesotarsi (compared to pseudotetramerous in all other Hydroporinae, see Fig. 7). Although this character state has led to their basal placement within the Hydroporinae (Francisco, 1979; Machado, 1987) or even the erection of their own subfamily (Hyporinae, Falkenström, 1938), no other characters suggest a close relationship between these three genera.

Madicolous species of Dytiscidae are apparently subject to severe morphological constraint associated with the invasion of wet rock habitats (e.g. Balke et al., 1997), and many features of Hydrotarsus, such as reduced swimming hairs, and indeed the tarsal structure may be associated with the evolution of madicoly. Recent work on the larva of H. compunctus (Alarie & Bilton, 2001) points to a close relationship between Hydrotarsus and the large Holarctic genus Hydroporus Clairville, rather than a basal placement within the subfamily, a finding backed up by preliminary results using ribosomal 18S rRNA sequence data (Ribera et al., 2002b). Here we present a mitochondrial DNA (mtDNA)-based phylogeny of Hydrotarsus and other species of Hydroporinae, which demonstrates that, rather than representing an isolated basal lineage, species currently assigned to Hydrotarsus actually fall deep within the genus Hydroporus. We therefore synonymize Hydrotarsus with Hydroporus and provide a redescription of its species, as well as discussing the biogeographical history and ecology of the ‘Hydrotarsus’ lineage.

**Materials and methods**

**Morphological analysis**

Morphometric measures were taken with an ocular micrometer attached to an Olympus (Hamburg, Germany) dissecting microscope at 40×. Beetle structures were examined with a Zeiss (Jena, Germany) Stemi SV6, at 12–80× (fluorescent bulb for diffuse light). Scanning electron micrographs were taken from gold–palladium-coated preparations with a Hitachi S2500 (Tokyo, Japan). Photographs of the habitus were taken with a Synoptics Automontage digital imaging system attached to a Zeiss Stemi SV6 at the Natural History Museum, London. Acronyms of the collections in which the material is deposited are as follows: BMNH, The Natural History Museum, London; MNHN, Muséum National d’Histoire Naturelle, Paris; NMW, Naturhistorisches Museum, Wien; CBH, coll. M. Balke & L. Hendrich (Berlin); CDB, coll. D. T. Bilton (Plymouth); CFP, coll. F. Pederzani (Ravenna); CGI, coll. G. Israelson (Uppsala); CGW, coll. G. Wewalka (Wien); CHF, coll. Hans Fery (Berlin); CNS, coll. N. Sanfilippo (Genoa).

**Taxon sampling and DNA sequencing**

The three recognized species of Hydrotarsus were included in the analysis: H. pilosus from Tenerife, H. compunctus from La Gomera and H. lundbladi from Madeira (Machado, 1987; Balke & Hendrich, 1989; Balke et al., 1990) (Table 1; Fig. 1). Preliminary analysis with a representation of different species groups of Hydroporus, together with other genera of Hydroporini, showed that Hydrotarsus belonged within the H. fuscipennis group (sensu Nilsson &
Table 1. Material studied, with species codes, species group (following Nilsson, 2001), geographical origin, collector, and GenBank accession numbers.

<table>
<thead>
<tr>
<th>Species</th>
<th>Species gr</th>
<th>Country</th>
<th>Collector</th>
<th>16S</th>
<th>COI</th>
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<td>26</td>
<td>H. landbladi Falkenström</td>
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<td>–</td>
<td>–</td>
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<td>27</td>
<td>H. pilosus Guignot</td>
<td>–</td>
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<td>D. Bilton</td>
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</tr>
</tbody>
</table>

Holmen, 1995; Nilsson, 2001). A more comprehensive sampling of this group, and of morphologically similar groups, was thus conducted (Table 1). Most of the remaining Palaearctic species groups of Hydroorus are represented by single species.

Outgroups include all genera of the Hydroorus group as defined in Ribera et al. (2002b) (formerly included in the single genus Hydroorus, see, e.g. Alarie, 1991), with the exception only of the recently described Hydrocolus Roughley & Larson (Larson et al., 2000); Suphrodytes Gozis, Neoporus Guignot, Heterosternata Strand and Sanfilippodytes Franciscolo. Other included Hydroporini were the Deroneces group of genera (sensu Nilsson & Angus, 1992) (Nebiroporus Régimbart, Deroneces Sharp, Stictotarsus Zimmermann, Scarodytes Gozis and Oreodytes Seidlitz) and the Stictotarsus group of genera sensu Ribera et al. (2002b) (Stictotarsus Brinck). The tree was rooted in Laccornis Gozis (tribe Laccornini), which certainly lies outside Hydroporini and probably in a basal position within Hydroporinae (Wolfe, 1985; Miller, 2001; Ribera et al., 2002b).

Soft tissue from single specimens collected in absolute ethanol was digested and DNA isolated using a phenol–chloroform extraction (as described in Vogler et al., 1993) or using the DNeasy kit (QUIAGEN, CRAWLE, UK). Sequences of 16S rRNA were amplified in a single fragment of c. 500 bp, using primers 16Sa (5’-ATGTTTGTGTTAACAGGCG)
for the 5’ end of the gene, and 16Sb (5’-CCGGTCTGAACCTGAGATCATG) for the 3’ end. A single fragment of c. 800 bp of Cytochrome Oxidase I (COI) (from the middle of the region E3 to the COOH end, Lunt et al., 1996) was amplified using the primers ‘Jerry’ (5’-CAACATTATTTTGATTITTTTG) for the 5’ end of the gene, and ‘Pat’ (5’-TCCAATGCACTATCGCCATAT) for the 3’ end (Simon et al., 1994). All sequences generated in this study were deposited in GenBank (Acc. Nos. AF518252–AF518312, Table 1).

The following cycling conditions were used: 1–2 min at 95°C; 30 s at 94°C, 30 s at 47–50°C (depending on the melting temperatures of the primer pair used), and 1–2 min at 72°C (repeated for 35–40 cycles); 10 min at 72°C. Amplification products were purified using a GeneClean II kit (Bio 101, Inc., Nottingham, U.K.). Automated DNA sequencing reagents were supplied by Perkin Elmer Applied BioSystems Ltd (Foster City, USA). (ABI PRISM Big Dye Terminator Cycle Sequencing Ready Reaction Kit). Sequencing reactions were purified by ethanol precipitation and were electrophoresed on an ABI3700 sequencer. Sequencing errors/ambiguities were edited using the Sequencher 3.1 software package (Gene Codes Corporation, Ann Arbor, USA).

Phylogenetic analysis

Sequences for COI were not length variable, and 16S rRNA sequences differed in length only minimally, affecting mostly outgroup taxa (see Results). Alignment was therefore performed manually, by attempting to maximize sequence similarities. Phylogenetic analysis was performed with PAUP4.0b8 (Swofford, 1999), using parsimony procedures for tree reconstruction [a heuristic search with 1000 Tree–Bisection–Reconnection (TBR) replicates]. Constraint trees for determining Bremer Support values (Bremer, 1994) and partitioned Bremer Support values (Baker & DeSalle, 1997) were generated with TREEROT (Sorenson, 1996). The significance of the incongruence length difference (ILD) (Farris et al., 1994) was assessed with the Partition Homogeneity Test as implemented in PAUP (using a heuristic search with 100 random-addition replicates). Non-parametric bootstrap support values (Felsenstein, 1985) were found in PAUP using 100 iterations of 100 TBR replicas each.

The COI nucleotide sequence was translated into an amino acid sequence using MACCLADE 4.0 (Maddison & Maddison, 2000), and analysed in PAUP. Owing to the low resolution attained, third codon positions were included in all analyses, and all characters were equally weighted.

Rate of diversification

The optimal evolutionary model was estimated using MODEST 3.04 (Posada & Crandall, 1998). To estimate node ages we fitted maximum likelihood (ML) branch lengths in the preferred tree assuming a molecular clock using the optimal model as estimated with MODEST, and compared the likelihood to that obtained assuming no clock (Felsenstein, 1981). When the two likelihood values are not significantly different (as measured with the likelihood ratio, which follows a chi-squared distribution, Felsenstein, 1981) it can be considered that the rate of evolution of the sequence is clock-like, and an ultrametric tree can be enforced.

To calibrate the branch lengths we used the standard 2% divergence per million years (Myr) for arthropod mitochondrial DNA (Brown et al., 1979; Brower, 1994), corresponding to a base rate (per branch) of 0.01 substitutions per site Myr\(^{-1}\). In Ribera et al. (2001) it is shown that for species of Dytiscidae (genus Agabus Leach) the 2% Myr\(^{-1}\) estimation for the combined COI plus 16S rRNA genes is equivalent to the much slower estimation of Gómez-Zurita et al. (2000) in a group of leaf beetles for the gene 16S rRNA alone (0.76% divergence Myr\(^{-1}\)).

Results

Phylogenetic analysis

Amplification of the COI gene was successful in all specimens, and a final interior continuous fragment of 769 bp was used for analyses. Uncorrected genetic distances among the taxa ranged from 1% to 17% (14% within the ingroup) (Table 2). The heuristic search resulted in two equally parsimonious trees (Table 3), with Hydrotarsus sister to

<table>
<thead>
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<tr>
<td>COI</td>
<td>H. tessellatus/H. basinotatus</td>
<td>N. dimidiatum/L. oblongus</td>
<td>H. Lundbladi/H. niger</td>
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<tr>
<td>0.01</td>
<td>0.17</td>
<td>0.14</td>
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<tr>
<td>16S rRNA</td>
<td>H. tessellatus/H. basinotatus</td>
<td>H. melanarius/S. epleuricus</td>
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<td>0.08</td>
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<td>0.16</td>
<td>0.14</td>
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<td>H. Lundbladi/S. halensis</td>
<td>H. Lundbladi/H. niger</td>
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**Table 3.** Tree statistics. CI, consistency index; RI, retention index; Inf. cha., number of parsimony informative characters.

<table>
<thead>
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<th>CI</th>
<th>RI</th>
<th>No. cha</th>
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<td>0.30</td>
<td>0.43</td>
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<td>388</td>
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</table>

*H. acutangulus* and *H. fuscipennis* (see Table 1 for the authors of the species included in the analyses), in a node including all the sampled species of the *H. fuscipennis* (excluding *H. limbatus*, nigrita, tessellatus and marginatus groups. This clade was sister to an unresolved group formed by the sampled species of the *H. longulus* and *mennoniun* groups. *Suphrodytes* was included within *Hydroporus*. Outgroup relationships were similar to that of the combined tree (see Fig. 2).

Parsimony analysis of the protein sequence resulted in a largely unresolved tree. The *Hydroporus* group of genera (including *Hydrotarsus*) formed a monophyletic lineage with 69% bootstrap support. Within it, only three monophyletic groups were resolved: Heterosternuta + Neoporus (99% bootstrap support), *H. gyllenhalii* + *H. vagpectus* (bootstrap support lower than 50%) and *H. erytrocephalus + H. niger + H. obscurus + H. rufifrons* (58% bootstrap support) (all of them included in the combined tree, Fig. 2).

16S rRNA sequence varied in length between 506 and 512 bp, although within *Hydroporus*, *Sanfilippodytes* and *Suphrodytes* differences were restricted to a single base pair (a deletion either in position 14 or 45). Uncorrected genetic distances among the taxa ranged from 0% for some species pairs with identical sequence to 16% (8% within the ingroup) (Table 2). A parsimony search using gaps as a fifth character resulted in 80 equally parsimonious trees (Table 3). The two species of *Hydrotratus* for which 16S rRNA could be obtained (*H. pilosus* and *H. compactus*, Table 1) had an identical sequence, which was included in an unresolved monophyletic group including *H. basinotatus, H. tessellatus, H. anulis, H. discreus, H. nigrita* and *H. pubescens*. This clade, together with the rest of the sampled species of the *H. fuscipennis, nigrita, tessellatus and marginatus* groups, formed an unresolved monophyletic group. *Suphrodytes* plus *Hydroporus* was also monophyletic although largely unresolved. The *Hydroporus* group of genera was monophyletic, and outgroup relationships were very similar to those of the combined analysis (see Fig. 2).

The combined analysis, using gap as a fifth character and both genes equally weighted, resulted in a single tree (hit 452 out of 1000 replicates) (Table 3 and Fig. 2). The incongruence among genes was not significant, as measured with the Partition Homogeneity Test (*P* = 0.08). Coding gaps as missing resulted also in a single tree of 2316 steps and CI = 0.29, with identical topology to that shown in Fig. 2.

To assess the possible effect of alternative alignments of the outgroup sequence, a search was conducted using only the species of *Hydroporus* plus *Suphrodytes* (which were unambiguously aligned, see above). The single tree found (1362 steps, CI = 0.37) had identical topology to that including outgroups (when rooted in *Suphrodytes*).

In the combined analysis, *Hydrotarsus* was included also in a monophyletic clade formed by the sampled species of the *H. fuscipennis, nigrita, tessellatus and marginatus* groups, hereafter referred to as the extended *H. fuscipennis* group (i.e. the less inclusive well-supported node including the species of *Hydrotarsus*), with high bootstrap support (86%) (Fig. 2). Within this group, relationships, although fully resolved, had low node support values.

The sister to the extended *H. fuscipennis* group was a clade formed by the sampled species of the *H. mennoniun* and *longulus* groups (with the exception of *H. obsoletus*), with the remaining species of *Hydroporus* basal. Other clades within *Hydroporus* with bootstrap values above 50% are *H. gyllenhalii*, *H. vagpectus* and *H. scalesianus* (representing the *tritis, striola* and *angustatus* groups, respectively); and *H. erytrocephalus, H. rufifrons, H. niger* (representing the *erytrocephalus, rufifrons* and *niger* groups, respectively), plus *H. obsoletus* (Figs 2, 3, Table 1). *Suphrodytes* was included within *Hydroporus* as sister to the later clade, although with low node support. The clade formed by *Hydroporus* plus *Suphrodytes* was, however, highly supported (89% bootstrap), as well as the *Hydroporus* group of genera (83% bootstrap, Fig. 2).

**Rate variation and molecular clock estimates**

Estimates of rate variation were restricted to the extended *H. fuscipennis* group (including *Hydrotarsus*). The best ML model (as estimated with modeltest, both using the Akaife Information Criteron or Hierarchical Likelihood Ratio Tests) was a complex GTR + I + G, with estimated base frequencies, among-site rate variation and a Gamma distribution shape parameter of 0.654. The likelihood ratio of the estimate enforcing and not enforcing a molecular clock was not significant (−lnML clock 4413.1; −lnML clock 4421.9; −2×(lnC − lnNC) = 17.50, 13 d.f., *P* = 0.18). The exclusion of *H. lundbladi* (with no 16S rRNA sequence) did not change the results (−lnML no clock 4282.9; −lnML clock 4292.2; −2×(lnC − lnNC) = 18.65, 12 d.f., *P* = 0.10).

The estimated ultrametric tree using the optimal ML model and enforcing a molecular clock (Fig. 4) showed maximum divergences rates within the extended *H. fuscipennis* group of approximately 16% (0.08 substitutions/site/branch) (*H. marginatus* vs. remaining species). The species of *Hydrotarsus* differ by c. 12.5% from their sister clade, although within them maximum differences were 5% (*H. lundbladi*). *Hydrotarsus pilosus* and *H. compactus* differ only by c. 2% (and only for the COI sequence, see above) (Fig. 4).

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Fig. 2. Phylogenetic hypothesis for the relationships of 'Hydrotarsus' based on a combined parsimony analysis of the 16S rRNA and COI genes. Numbers below the branches are partitioned Bremer support values (COI/16S); numbers above the branches are bootstrap proportions (only shown if >50%). See Table 1 for details of species.
Fig. 3. Phylogram of the single most parsimonious tree obtained in the combined parsimony analysis of the 16S rRNA and COI genes, with the species groups and the main clades. See Table 1 for details of species.

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**Taxonomy**

*Hydroporus* Clairville, 1806  
*Hydatoporus* Gistel, 1856  
*Hydrocoptus* Motschulsky, 1853  
*Hydroporidius* Guignot, 1949  
*Hydroporinus* Guignot, 1945  
*Schizoporus* Ádám, 1996  
*Sternoporus* Falkenström, 1930  
*Hydrotarsus* Falkenström, 1938, syn.n.

*Hydroporus compunctus* lineage

The three species of the former genus *Hydrotarsus* share a very similar external morphology: stout body (Fig. 5), antenna compact with roundish flagellomeres. Fore and especially middle tarsi clearly five segmented: 4th protarsomere not mostly concealed by lateral lobes of 3rd tarsomere (pseudotetramerous condition, see Nilsson & Holmen, 1995: 56) (Fig. 7); 4th mesotarsomere fully exposed. Head dark brown, with contrasting orange areas in anterior, median and frontal parts; pronotum black with paler lateral and anterior margins; elytron dark brown; appendages orange; ventral side dark brown. Metacoxae slightly produced backwards medially, concave on each side (Fig. 7) (this character state is present also in *Heterosternuta*, Zimmermann, 1931; Larson et al., 2000). Fringes of swimming hairs on legs rudimentary or absent, consisting of a dorsal row of few long hairs (Fig. 6E). Legs without swimming hairs on the ventral side (Fig. 6C,D,F,G) (only *H. compunctus* males possess a few stout hairs ventrally, Fig. 6A,B). Median lobe of the aedeagus very similar in all species (Fig. 7), and similar to other species of the group (see, for example, Wewalka, 1992; Nilsson & Holmen, 1995; Larson et al., 2000).

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Fig. 4. Ultrametric tree for the extended *Hydroporus fuscipennis* group of species, obtained with maximum likelihood (optimal model) enforcing a molecular clock, with the topology of the parsimony tree including outgroups (see Figs 2, 3). Numbers on branches are their estimated length. The absolute timescale corresponds to an estimated rate of 0.01 substitutions per site per Myr per branch, equivalent to the standard divergence rate of 2% Myr⁻¹. See Table 1 for details of species.
Key to the species of the *Hydroporus compunctus* lineage

1. Elytra and pronotum dull due to presence of strong microreticulation; angle in lateral outline between base of pronotum and elytron small (Fig. 5); males with first metatarsal segment strongly expanded, two first metatarsomers with long, strong ventral setae (Fig. 6D,E); metacoxa densely covered with stout setae (Fig. 7). Madeira............... *H. lundbladi* (Falkenström)
   – Elytra and pronotum shinier, with faint microreticulation; lateral outline of body continuous, without recognizable angle between base of pronotum and elytron (Fig. 5); first metatarsal segment in male not expanded, with fewer stout ventral setae (Fig. 6A–C,F,G); metacoxa less densely setose (Fig. 7). Canary Islands....................................................2
2. Body shape broadly oval (Fig. 5A); puncturation on pronotum coarser and denser; male two first metatarsal segments with several long, stout ventral setae, but without longer, stout hairs (Fig. 6F). Gran Canaria, Tenerife...........................*H. pilosus* (Guignot)
   – Body shape elongate oval (Fig. 5B); puncturation on pronotum slightly finer and sparser; male two first metatarsal segments with fewer stout setae and longer stout hairs (Fig. 6A,B). La Gomera, La Palma, Tenerife?.........................*H. compunctus* Wollaston

*Hydroporus compunctus* Wollaston


? = *Hydroporus pubescens* (Gyllenhal, 1808); Bedel 1925: 369.


Type locality. Tenerife, ‘? in the Barranco at Ycod el Alto’.

Type material. Holotype female in BMNH (Wollaston Canarian Island Collection, ‘Supplementary Material’ drawer).


Diagnosis. See Table 4 for measurements. Body form shortly oval; lateral outline not interrupted between pronotum and elytron (Fig. 5B). Pronotum widest at base, gently narrowing towards head. Head with distinct microreticulation, which is, however, only moderately deeply impressed. Pronotum and elytron both with faint microreticulation. First male metatarsal segment not enlarged, with only few stout ventral setae and with few longer, thick hairs (Fig. 6A,B); first female metatarsal segment only with few stout setae (Fig. 6C). Median lobe of aedeagus as in Fig. 7A.

Distribution. Canary Islands: La Gomera and La Palma. The type locality (Ycod el Alto, Tenerife) may be mislabelled.

Ecology. At Siete Fuentes, the beetles were found on a wet, almost vertical, 4–5-m-high rock surface in the main course of an otherwise dry streambed. There was a slight trickle of water, and the beetles were collected from underneath slices of stone, moss and washed out of small crevices in the rock. At Garajonay adults and larvae were collected from a vertical madicolous surface beside the road. Specimens were found crawling rapidly in the water film, and hiding in crevices and under dead leaves. Adults were observed feeding on moribund Oligochaeta.

Table 4. Morphometric measurements of the species of the *H. compunctus* lineage. TL-h, total length minus head (~96% of total length); TW, maximum width; Lp, length of pronotum medially; Wp, maximum width of pronotum. All measurements are in millimetres.

<table>
<thead>
<tr>
<th>Species</th>
<th>Locality</th>
<th>N</th>
<th>TL-h (mean)</th>
<th>TW</th>
<th>Lp</th>
<th>Wp</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>H. compunctus</em></td>
<td>La Palma</td>
<td>10</td>
<td>2.60–3.00 (2.80)</td>
<td>1.60–1.80</td>
<td>0.60–0.65</td>
<td>1.30–1.52</td>
</tr>
<tr>
<td><em>H. pilosus</em></td>
<td>Gran Canaria</td>
<td>15</td>
<td>2.80–3.00 (3.00)</td>
<td>1.85–2.00</td>
<td>0.55–0.62</td>
<td>1.42–1.60</td>
</tr>
<tr>
<td><em>H. pilosus</em></td>
<td>Madeira</td>
<td>10</td>
<td>2.75–2.90 (2.80)</td>
<td>1.63–1.80</td>
<td>0.62–0.67</td>
<td>1.47–1.55</td>
</tr>
<tr>
<td><em>H. lundbladi</em></td>
<td>Madeira</td>
<td>10</td>
<td>2.60–3.00 (2.80)</td>
<td>1.60–1.80</td>
<td>0.60–0.65</td>
<td>1.30–1.52</td>
</tr>
</tbody>
</table>
Remarks. Wollaston (1865) noted: ‘The only specimen which I have seen of this Hydroporus was taken by the Messrs. Crotch, during the summer of 1864, in Tenerife - I believe, in the Barranco at Ycod el Alto’. There are thus some doubts on the true origin of the holotype.

**Hydroporus lundbladi (Falkenström), comb.n.**


Type locality. Madeira: Ribeira do Inferno.

Type material. Two syntypes (one male, one female) ‘Ribeira do Inferno [the female under moss near waterfall], 10.viii.1935’ in Naturhistoriska Riksmuseet Stockholm, not studied.

Additional material examined. 1 ex., Ribeira Joao Delgado, 19.vi.1935, 1450 m, d’Orchymont (MNHN); 94 exx, Pico de Ferreiro, 1500 m, 25–27.vii.1986, Balke & Hendrich leg. (NMW, CBH, CHF, CGW, CFP, CNS). 15 exx, Pico de Ferreiro, 1500 m, iii.1995; 8.iii.2001, D. T. Bilton leg. (CDB, 1 ex. 8.iii.2001 used for a non-destructive DNA extraction, BMNH ref. IR664).

Diagnosis. See Table 4 for measurements. Body form shortly oval, with a rather stout appearance, body appendages stout; angle between base of pronotum and base of elytron small but visible (Fig. 5C). Pronotum widest shortly anterior to base, gently narrowing towards head and slightly curved towards posterior angle. Head with distinct microreticulation moderately to deeply impressed. Pronotum and elytron with distinct microreticulation. First male metatarsal segment strongly enlarged, with numerous long, stout ventral setae; without swimming hairs (Fig. 6D,E); second segment distinctly enlarged; third and fourth only slightly enlarged. This is the only known species of Dytiscidae in which males have expanded metatarsomeres. Median lobe of aedeagus as in Fig. 7B.

Distribution. Madeira.

Ecology. At Pico de Ferreiro, M.B. and L.H. found specimens on a vertical rock surface, at the bottom of which a trickle of water had formed from a spring below the otherwise dry summit area. Specimens were found under hepatics, such as Marchantia polymorpha (L.), stones and slices of rock, which could be lifted with a chisel (Fig. 8). The same locality was visited by D.T.B. at wetter times of year, when specimens could be found crawling actively on vertical madicolous surfaces devoid of bryophyte cover. It is probable that the species retreats into crevices etc. in response to the seasonal drying of its wet rock habitat.

Remark. We do not consider the designation of a lectotype, as the description of this species is unambiguous.

**Hydroporus pilosus (Guignot), comb.n.**


**Fig. 8.** Habitat of *Hydroporus lundbladi* in Pico de Ferreiro, Madeira. A, general view of the rock wall and the streamlet; B, detail of the microhabitats.
**Type locality.** Gran Canaria: Teror.

**Type material.** 1 female syntype, Gran Canaria, Teror, 650 m, 11.v.1935, d’Orchymont leg. (MNHN, coll. F. Guignot). The remaining male syntype, which should be housed in the Institut Royal des Sciences Naturelles de Belgique, Bruxelles, is probably lost (cf. Machado, 1987: 31).

**Additional material examined.** Circa 650 exx, Gran Canaria, Pico de las Nieves area, 1200–1700 m, 5–7.vii.1989, Balke, Hendrich & Fery leg. (NMW, CBH, CHF, CGW, CFP, CNS); 12 exx, Tenerife, Anaga Massif, Roque Chino, xii.1997, D. T. Bilton leg. (CDB, 1 ex. used for DNA extraction, BMNH ref. IR37).

**Diagnosis.** See Table 4 for measurements. Body form shortly, broadly oval; lateral outline without noticeable angle between pronotum and elytron (Fig. 5A). Pronotum widest at base, gently narrowing towards head. Head with distinct microreticulation, only moderately deeply impressed. Pronotum and elytron with faint microreticulation; on elytron generally very hardly visible and often rudimentary. With well-developed metasternal wings (five specimens dissected). First male metatarsal segment not enlarged, with numerous stout ventral setae (Fig. 6F), female with fewer stout setae (Fig. 6G). Median lobe of aedeagus as in Fig. 7C.

**Distribution.** Canary Islands: Gran Canaria, Tenerife.

**Ecology.** On Gran Canaria specimens were found on wet, more or less vertical rock surfaces and overhanging rocks in an otherwise dry stream bed on a steep, isolated slope, hiding under stones, grass and moss. On Tenerife beetles were collected from water films on vertical rock surfaces beside roads and tracks in laurisylva.

**Remark.** We do not consider the designation of a lectotype, as the description of this species is unambiguous.

**Discussion**

**Phylogenetic position of ‘Hydrotarsus’ and the extended fuscipennis group**

Our results firmly support the monophyly of the three species of the former genus ‘Hydrotarsus’ and its inclusion within Hydroporus. Several very robust nodes support this conclusion: the Hydroporus group of genera (Hydroporus, Saphrophylax, Sanfilippodytes, Heterosternuta and Neoporus, with 83% of bootstrap support), the genera Hydroporus plus Saphrophylax (89% of bootstrap support) and the clade we have designated as an extended H. fuscipennis group (see below) (86% bootstrap support). The genus Hydrotarsus was considered previously to be a basal offshoot of the subfamily Hydroporinae, mostly on the basis of its penta-

merous protarsi (Falkenström, 1938; Franciscolo, 1979). However, in Alarie & Bilton (2001) its close relationship with Hydroporus and Sanfilippodytes was noted, as well as the possibility that the divergent, and supposedly plesiomorphic features of the species of ‘Hydrotarsus’ could in fact be derived characters related to their madicolous habits.

Our sequence data strongly support the monophyly of the clade we introduce as the extended H. fuscipennis group (we retain the name ‘fuscipennis’ as this group is the most speciose of those combined here, Nilsson, 2001). We prefer this option to maintaining the individuality of former groups by re-distributing selected species because relationships within the extended H. fuscipennis group are not robust (see below), and are very likely to vary with the inclusion of new taxa.

The *Hydrotarsus fuscipennis* group (planus group *sensu* Zimmermann, 1931) so far contains 29 species, and has a Holarctic distribution (Nilsson, 2001). The group was revised partially by Wewalka (1992) (some Palaeartic species close to *H. planus*) and Larson *et al.* (2000) (Nearctic species). It was defined primarily on the basis of the absence of microreticulation on the elytral disc, which presents a smooth and shining surface between punctures (Nilsson & Holmen, 1995). Under our phylogenetic hypothesis, however, the ancestral condition of the extended *H. fuscipennis* group is with microreticulation (present in the most basal species, *H. marginatus*, as well as in their sister lineage, the *H. melanarius, longulus* and *memnonius* groups). As re-constructed with our data, the evolution of the elytral reticulation is ambiguous, with either four independent origins of a smooth surface (in *H. pubescens, H. discreitus, H. analis* and the five species of the *H. planus* clade) using the ‘delay changes’ (DELTRAN) option in MACCLADE 4.0; or a single origin (at the base of the group, excluding *H. marginatus*) with three reversals to the plesiomorphic, reticulate condition (the ‘Hydrotarsus’ lineage, *H. nigrita*, and *H. tessellatus* plus *H. basinotatus*), using the ‘accelerate changes’ option (ACTTRAN). Regardless, this character is shown to be highly labile and can no longer be considered a reliable synapomorphy of the group.

The species included in our extended *H. fuscipennis* group all have a broad lateral beading of the pronotum, elytron with the lateral margin weakly ascending to the humeral angle, and the metacoxal processes with its posterior margin conjointly truncate (although the species of ‘Hydrotarsus’ have a produced metacoxal process, see Zimmermann, 1931). On the basis of these characters they would, however, be part of a larger clade, including the species of the *memnonius* and *longulus* groups (the latter also with a produced metacoxal process) (Nilsson & Holmen, 1995). At present we cannot recognize any defining morphological synapomorphy for the species of an extended *H. fuscipennis* group. The utility of defining species groups based solely on molecular characters could of course be questioned, although in our opinion the information content of a monophyletic group, even when not well defined morphologically, is always higher than that of species assemblages based on homoplastic characters. Species groups within *Hydroporus*
were introduced originally by Zimmermann (1931), who dealt with the Palaearctic fauna. Since their inception, these groups have been modified and redefined, largely to incorporate a Holarctic perspective (Nilsson & Holmen, 1995; Larson et al., 2000; Nilsson, 2001), and many remain rather ill-defined. As demonstrated here, whilst they may serve as convenient ways to link morphologically similar species, some of these species groups may not reflect phylogeny, which is a strong argument for refraining from affording them formal taxonomic status (Nilsson & Holmen, 1995).

Within the extended H. fuscipennis group the position of ‘Hydrotarsus’ is less well established. In the combined analysis it is sister to a clade of species mostly with a western Mediterranean distribution, although with low bootstrap and Bremer support values. This uncertainty is not likely to result from incomplete sampling, as all the species of the extended H. fuscipennis group known to occur in Morocco (the continental area closest to Madeira and the Canary Islands, and the likely area of origin of the H. compunctus lineage) were included in the analysis (Wewalka, 1992; Nilsson, 2001).

In terms of the remaining species of the genus Hydroporus, our results support the conclusions of Nilsson (1989) in considering as synonyms the former subgenera Sternoporus and Hydropropidius (represented in our study by the species H. longulus and H. melanarius, respectively). These two groups are found to be closely related to the sampled species of the H. memnonius group (with 100% of bootstrap support), as suggested by different authors (see, for example, Nilsson, 1989; Fery, 1999). The species H. obsoletus was included formerly in the group ferrugineus, which was subsequently merged with the H. memnonius group by Nilsson (2001). Our results do not support the relationship between this species and those of the H. memnonius group.

We found the genus Suphrodytes nested within Hydroporus, although with low node support. The genus was included formerly in Hydroporus, but excluded from it by Angus (1985) based on a series of internal and external morphological characters. A more comprehensive sampling of the basal lineages of Hydroporus is necessary to clarify its systematic position.

The monophyly of the Hydroporus group of genera sensu Ribera et al. (2002b) (with the inclusion of Sanfilippodytes) is strongly supported. Suphrodytes, Heterosternta and Neoporus formerly were considered subgenera within Hydroporus (e.g. Matta & Wolfe, 1981; Wolfe, 1984; Angus, 1985), as well as most of the species later transferred to Sanfilippodytes, which were known previously as the ‘Hydroporus vilis group’ (Rochette, 1983). In a phylogenetic analysis of the larvae, Alarie (1991) provides several synapomorphies for the group, although subsequent morphological phylogenetic work found them paraphyletic with respect to the Deronectes group of genera (Alarie & Delgado, 1999; Alarie et al., 1999). We found the Hydroporus and Deronectes groups of genera, respectively, monophyletic, in agreement with Alarie (1991) and traditional taxonomy.

**Biogeography and ecology**

Species of the *Hydroporus compunctus* lineage are restricted to humid areas of the Canary Islands and Madeira, typically occurring in more or less permanent seepages on vertical rock faces in areas above 800 m (Balke & Hendrich, 1989; Balke et al., 1990; Alarie & Bilton, 2001), primarily, although not exclusively, in areas of monteverde (laurisylva and fayal-brezoal). Such regions are subject to frequent precipitation in the form of rain and cloud banks, delivered by the north-easterly trade winds (Kunkel, 1976). *Hydroporus lundbladii* is restricted to the main island of Madeira, where it occupies areas on the central ridge. Within the Canary Islands, both *H. pilosus* and *H. compunctus* occur on two islands each. Such a lack of single-island endemism is typical of the Canarian Dytiscidae and other groups of aquatic insects (Bilton et al., 2001; Kelly et al., 2001), and contrasts considerably with most terrestrial coleopteran radiations. *Hydroporus pilosus* was described from Gran Canaria, and subsequently found on Tenerife. The precise distributional limits of *H. compunctus* are more problematic. The species was described from specimens apparently collected on Tenerife (Wollaston, 1865), but, following the discovery of this species on La Palma, Balke et al. (1990) considered this record to have resulted from mislabelling, known to have occurred with other species in Wollaston’s material. The discovery of this species on La Gomera (Alarie & Bilton, 2001), however, demonstrates that this species is not endemic to the island of La Palma, and increases the possibility that the species does, or did, occur on Tenerife. As one of the youngest islands in the archipelago, La Palma has few unique endemic invertebrate taxa (e.g. Machado, 1992).

According to the standard rate of c. 2% divergence per Myr of insect mitochondrial DNA (Brower, 1994), the species of the *H. compunctus* lineage originated c. 6 Myr ago (late Miocene). *Hydroporus lundbladii* has been separated from the rest for c. 2.3 Myr, and the separation between *H. compunctus* and *H. pilosus* dates to c. 1 Myr. Based on these divergence time estimates, rather than being relictual, ancestral Hydroporinae (Falkenström, 1938; Franciscolo, 1979), this group is highly derived within the wider genus *Hydroporus*, and represents a relatively recent invasion of Macaronesia. Such a finding is in keeping with other molecular studies of Canarian Coleoptera, almost all of which suggest a recent origin of extant endemic species, rather than relictual status (e.g. Juan et al., 1995; Emerson et al., 1999, 2000a, b; Rees et al., 2001; Ribera et al., 2003; see Juan et al., 2000, for a review). The unique character states of ‘Hydrotarsus’ are likely to be associated with the switch to obligate madicoly (see below).

The radiation of the species of the *H. compunctus* lineage within Macaronesia cannot be clearly related to the emergence times of islands, based on mtDNA sequence divergence. Speciation within the clade clearly post-dates the appearance of individual islands, which range from 20 Myr (Madeira) to 2 Myr (La Palma) (Juan et al., 2000). Interpretation of the colonization history of the group within and...
The hygropetric syndrome in Dytiscidae

‘Hygropetric’ (Thienemann, 1905) or ‘petrimadicolous’ (Vaillant, 1956) habitats are those in which a thin layer of water (usually of only a few millimetres) runs over bare rock surfaces. These habitats are frequent in mountain rock outcrops and on coastal cliffs, and usually are fringed by dense layers of mosses, ferns and a diverse vascular vegetation. The aquatic beetle faunas of hygropetric habitats in most parts of the world have been largely overlooked, and some of their most characteristic species were considered until recently extremely scarce oddities of mysterious provenance (e.g. Agabus aubei Perris in Corsica, Balke et al., 1997; or the species of ‘Hydrotarsus’, Machado, 1987).

Most hygropetric species of Coleoptera show deviant morphological features, meaning that many were described originally as separate genera (e.g. Metronectes aubei, now Agabus aubei, Balke et al., 1997; or ‘Hydrotarsus’), or even higher taxa (e.g. Hydrotrupes palpalis Sharp, considered to represent a monogeneric subfamily by Larson et al., 2000, based on the phylogenetic analysis of Beutel, 1994; or the recently described hygropetric family Aspidytidae, Ribera et al., 2002a). In some of the cases in which it has been possible to obtain molecular data, the phylogenetic analyses have demonstrated that characters considered to be plesiomorphic were instead secondarily derived, and the species usually more recent than expected (A. aubei, Ribera et al., 2001; the former Hydrotrarsus, this paper). Even in the case of Aspidytes Ribera et al., in which combined analysis of three genes and a morphological matrix demonstrate an ancient origin, the lack of swimming abilities, and the primitive appearance of the legs (without any apparent modifications for aquatic life) are considered to be secondarily derived (Ribera et al., 2002a). The study of the phylogenetic position of other hygropetric genera with a deviating morphological specializations to the aquatic life. The fact that the derived morphology of the hygropetric species resembles in many cases the supposed plesiomorphic condition of the family Dytiscidae (or the group of aquatic families of Adephaga) suggest the involvement of relatively simple changes in regulatory pathways that could, however, result in major morphological modifications. This could also be the case for the aberrant morphology of the meta-
tarsi of H. lundbladi (which, unique among dytiscids, have the same structure as the pro- and mesotarsi). It is clear that the detailed study of hygropetric habitats and their associated fauna is of maximum interest both for the understanding of the phylogeny of Dytiscidae and the evolution of their morphological adaptations to the aquatic life.

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References


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conserved primers for phylogenetic studies. Insect Molecular Biology, 5, 153–165.


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