**Arganiella Giusti & Pezzoli, 1980 (Caenogastropoda: Truncatelloidea: Hydrobiidae): a widespread genus or several narrow-range endemic genera?**

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Abstract. Most valvatiform genera of the gastropod family Hydrobiidae are narrow-range taxa. One exception is the genus *Arganiella*, which is comprised of three congeners: the type species *A. pescei* from the Apennine Peninsula, *A. wolfi* from the Iberian Peninsula and *A. tabanensis* from the Balkans. The genus assignment of the latter two species was based on morphological similarities with *A. pescei* in the shell, operculum, radula and genitalia. Given that the morphology of hydrobiids is sometimes susceptible to convergence, this study re-evaluates the taxonomic status of species of *Arganiella* by analysing mitochondrial (mtCOI) and nuclear (18S rRNA) sequences of topotypes or near topotypes to infer their phylogenetic position. Our phylogenetic analyses depicted *Arganiella* as a non-monophyletic group within Hydrobiidae, and sequence divergence among the three species ranged from 14.5 to 16.7% for mtCOI and 2.0 to 3.8% for 18S. We also re-examined the extent of morphological variation among species of *Arganiella* and found a few differences among them and other valvatiform genera. Consequently, we propose two new genera for *A. wolfi* and *A. tabanensis*. Our results conflict with the classification of valvatiform hydrobiid species solely based on traditional phenotypical methods and suggest further taxonomic evaluation within a molecular framework.

Keywords. Springsnails, valvatiform hydrobiids, phylogeny, morphology, Mediterranean region.
Introduction

The gastropod genus *Arganiella* Giusti & Pezzoli, 1980 (family Hydrobiidae Stimpson, 1865) comprises three narrow-range endemic species, each living on a different Mediterranean peninsula. These tiny springsnails (1.0–1.5 mm) have a depressed trochiform to valvatiform shell and simplified anatomical structures. The type species, *A. pescei* Giusti & Pezzoli, 1980, was described from several wells and springs located in the central-eastern Apennine Peninsula (Giusti & Pezzoli 1980). A proposed representative of the genus *Boetersiella* Arconada & Ramos, 2001, *B. wolfi* Boeters & Glöer, 2007, which is endemic to the southwestern Iberian Peninsula, was transferred to *Arganiella* by Arconada & Ramos (2007a, 2007b). Based on morphological and geographical evidence, these authors also synonymized the species *A. tartesica* Arconada & Ramos, 2007 with *A. wolfi* (Arconada & Ramos 2007b). The third species, *A. tabanensis* Boeters, Glöer & Pešić, 2014, was recently erected for a hydrobiid population living in a small rheocrene spring in Montenegro, in the Balkan Peninsula (Boeters et al. 2014).

The assignment of *A. wolfi* and *A. tabanensis* to *Arganiella* was based on morphological similarity with the type species in shell, penis and female distal genital features (Arconada & Ramos 2007a, 2007b; Boeters et al. 2014). However, such characters are susceptible to convergent evolution in valvatiform hydrobiid genera (Bodon et al. 2001) and, therefore, the systematics of the known species of *Arganiella* needs to be re-evaluated through molecular analyses. To date, DNA sequence data are available only for *A. wolfi*, and phylogenetic relationships inferred from a multilocus dataset of valvatiform hydrobiid genera from the Iberian Peninsula resolved *A. wolfi* as the sister taxon of the Iberian genus *Iberhoratia* Arconada & Ramos, 2007 (Delicado et al. 2019). Sequence data from the other two species of *Arganiella* are needed to confidently assess the taxonomic composition of this wide-ranging genus.

As might be expected in taxa with poor dispersal capabilities, we hypothesize that phylogenetic analyses would depict species of *Arganiella* as three unrelated, narrow-range lineages rather than as a widely distributed monophyletic group. This result would challenge the genus assignment of these valvatiform hydrobiid genera (Bodon et al. 2001) and, therefore, the systematics of the known species of *Arganiella* needs to be re-evaluated through molecular analyses. To date, DNA sequence data are available only for *A. wolfi*, and phylogenetic relationships inferred from a multilocus dataset of valvatiform hydrobiid genera from the Iberian Peninsula resolved *A. wolfi* as the sister taxon of the Iberian genus *Iberhoratia* Arconada & Ramos, 2007 (Delicado et al. 2019). Sequence data from the other two species of *Arganiella* are needed to confidently assess the taxonomic composition of this wide-ranging genus.

Material and methods

We assessed the taxonomic status of the three recognized species of *Arganiella* using DNA sequence and morphological information from these and other valvatiform genera occurring in the Apennine, Balkan and Iberian peninsulas. One individual of *A. pescei* was used for the DNA assessment. This specimen was collected from Susanna Springs, in the region of the type locality (i.e., central-eastern Apennine Peninsula), and deposited in the collection of the University of Giessen Systematics and Biodiversity (UGSB) (Diehl et al. 2018) in Germany (UGSB 10365). Following an exhaustive morphological examination, Bodon et al. (2001) had assigned the hydrobiid populations living in these springs to *A. pescei*. Morphological characters from *A. pescei* were scored using the original description by Giusti & Pezzoli (1980) and the re-description by Bodon et al. (2001). For the Iberian *A. wolfi*, we used the morphological description by Arconada & Ramos (2007a) and Boeters & Glöer (2007), and the sequences of a topotype used by Delicado et al. (2019) for the molecular analyses. The shell morphology and anatomy of the Balkan *A. tabanensis* are as illustrated by Boeters et al. (2014). Additional data on
radular and opercular features of this species, as well as partial sequences of the studied DNA markers, were also collected for the present study. We collected ca 50 topotypes from the *A. tabanensis* type locality (Taban Spring, Montenegro; 42.52795° N, 19.21921° E) in 2015 and preserved them in 80% ethanol in the field. Shells and opercula were photographed using a Keyence VHX 2000 3D Digital Microscope. Six adults were dissected, and their buccal mass extracted, under an Olympus SZX12 stereo microscope. Radulae were extracted, cleaned and prepared as described by Delicado et al. (2016) for imaging on a field emission scanning electron microscope (FESEM) DSM982 Gemini (Carl Zeiss GmbH, Germany). The collected specimens were then deposited in the UGSB collection (UGSB 18847).

We isolated total DNA from one individual per species (for *A. pescei* and *A. tabanensis*) following the CTAB protocol performed by Wilke et al. (2006). Fragments of the mitochondrial cytochrome *c* oxidase subunit I (COI) and nuclear ribosomal 18S rRNA (18S) were amplified and sequenced using the primer pairs LCO1490 (Folmer et al. 1994) and COR722b (Davis et al. 1998) for COI and the universal primers for metazoan 18S (Holland et al. 1991). Amplification conditions for both gene fragments were as previously described by Delicado et al. (2012). The annealing temperature used was 52°C. Samples were sequenced in an ABI 3730 XL sequencer (Life Technologies, Carlsbad, CA, USA) using a BigDye Terminator Kit ver. 3.1 (Life Technologies). The new sequences were deposited in GenBank (Table 1).

We assessed the taxonomy of the three species of *Arganiella* by analysing the COI and 18S sequences of these species along with those of other European valvatiform and (closely related) non-valvatiform genera available from GenBank (Table 1). Forward and reverse sequences were aligned and edited in Sequecher ver. 5.4.6 (Gene Codes, Ann Arbor, MI). MEGA ver. 7.0.14 (Kumar et al. 2016) was used to assemble the gene-partition datasets and to calculate genetic distances (uncorrected *p*-distances). The COI dataset was manually aligned also using MEGA. The rRNA 18S fragment was aligned using MAFFT ver. 7.402 (Katoh & Standley 2013), with default settings for gap penalties. According to the corrected Akaike’s information criterion (AICc; Akaike 1974; Sugita 1978; Hurvich & Tsai 1989), jModelTest ver. 2.1.7 (Darriba et al. 2012) selected TrN (Tamura & Nei 1993) + I (invariable sites) + G (rate variation among sites) and TrNef (Tamura-Nei model with equal base frequencies; Tamura & Nei 1993) + I + G models of nucleotide evolution for the COI and 18S datasets, respectively. We used DAMBE ver. 7 (Xia 2018) and the proportion of invariant sites (*P*inv = 0.47) obtained in jModelTest to conduct the saturation test (Xia et al. 2003; Xia & Lemey 2009) on the COI partition. The observed saturation (*I*ss = 0.39) was significantly lower than the critical values (*I*ss.c = 0.71; *p* < 0.001), suggesting little saturation in our COI dataset.

Phylogenetic analyses based on maximum likelihood (ML) methods were conducted using the RAxML BlackBox web-server [https://raxml-ng.vital-it.ch/#/; Kozlov et al. 2019] with 10 random starting trees and the optimal substitution models for each gene partition selected in jModelTest. Bayesian inference (BI) analyses were run with mixed substitution models in MrBayes ver. 3.2.6 (Ronquist et al. 2012) for 5 million generations with a sampling frequency of 1000. After verifying convergence of the BI analysis (standard deviation of split frequencies < 0.01), the first 10% of generations were discarded as burn-in. Branch robustness was evaluated by rapid bootstrapping (BS) (Stamatakis et al. 2008) with an automatic cut-off for ML and by Bayesian posterior probability (BPP) for BI. Inferred topologies and branch supports were visualized in FigTree ver. 1.4.3 (Rambaut 2010).

For the morphological comparisons of species of *Arganiella* with other valvatiform hydrobiid genera from the Iberian, Apennine and Balkan peninsulas (Table 2), we scored morphological character states, including features of the shell, radula, operculum and anatomy, according to the information provided in the following studies: Arconada & Ramos (2001) for *Boettersiella* and *Chondrobasis* Arconada & Ramos, 2001; Girardi & Boeters (2012) for *Corbellaria* Girardi & Boeters, 2012; Delicado et al. (2019) for *Deganta* Arconada & Ramos in Delicado et al., 2019; Arconada et al. (2007) for *Iberhoratia* Arconada & Ramos, 2007; Arconada & Ramos (2006) for *Josefus* Arconada & Ramos,
Table 1 (continued on next page). Taxon name, locality name, GenBank numbers and original references for sequences analysed in this study.

| Taxon                  | Locality                                                                 | GenBank # I8S | GenBank # COI | Original reference                   |
|------------------------|---------------------------------------------------------------------------|---------------|---------------|--------------------------------------|
| Agrafia wiktori        | Agrafa Mountains, Sikiá, Evrytania, Greece (39.36861, 21.63319)           | JF906758      | JF906765      | Szarowska & Falniowski (2011)        |
| Alzoniella finalina    | Spring at the Porra River, Molino, Liguria, Savona, Italy (44.219, 8.255) | AF367686      | AF367650      | Wilke et al. (2001)                  |
| Arganiella pescei      | Santa Susanna Springs, Rivodutri, Latium, Italy (42.49513, 12.84604)     | MW561453      | MW553909      | Present study                        |
| Arganiella tabanensis  | Taban Spring, Montenegro (42.52795, 19.21921)                             | MW561454      | MW553910      | Present study                        |
| Arganiella wolfi       | Virgen de los Ángeles hermitage, Sierra de Aracena, Huelva, Spain (37.8785, -6.66881) | MH348095      | MH350192      | Delicado et al. (2019)               |
| Avenionia brevis       | Spring of the fountain of St.-Victor-La Coste, Gard, France (44.057, 4.636) | AF367670      | AF367638      | Wilke et al. (2001)                  |
| Belgrandia thermalis   | Thermal channel near S. Giuliano, Tuscany, Pisa, S. Giuliano Terme, Italy (43.751, 10.440) | AF367684      | AF367648      | Wilke et al. (2001)                  |
| Belgrandiella kusceri  | Rakovski potok [Crab stream], near Rakovski Skocjan, Rakėk, Slovenia (45.77750, 14.18556) | JX970574      | KT218520      | Wilke et al. (2013); Falniowski & Beran (2015) |
| Boetersiella sturmi    | La Mata Spring, Mata Bejid, Jaén, Spain (37.69503, -3.50703)              | MH348097      | MH350200      | Delicado et al. (2019)               |
| Bullaregia tunisiensis | A spring in Djebba, Province Béja, Tunisia (36.47125, 9.09972)           | MN575790      | KX821683      | Khalloufi et al. (2017, 2020)        |
| Chondrobasis levantina | Caroche Spring, Teresa de Cofrentes, Valencia, Spain (39.09608, -0.9156) | MH348098      | MH350203      | Delicado et al. (2019)               |
| Corbellaria celtiberica| Manubles River, Soria, Spain (41.60997, -1.95586)                         | MH348099      | MH350207      | Delicado et al. (2019)               |
| Daphniola graeca       | Daphne Spring, about 30 km north of Larissa, Greece (39.89111, 22.60722) | EF070624      | EU047764      | Szarowska (2006); Falniowski et al. (2007) |
| Deganta azarum         | La Fontona Spring, Borondes, Asturias, Spain (43.31890, -6.01494)         | MH348100      | MH350208      | Delicado et al. (2019)               |
| Fissuria boui          | Spring near La Prouveresse, Alpes Maritimes, France (43.64279, 6.88735)  | AF367960      | AF367654      | Wilke et al. (2001)                  |
| Graecoarganiella       | Small spring south of Eptalofos, Parnassus mountains, Greece (38.59278, 22.5039) | JN202341      | JN202348      | Falniowski & Szarowska (2011a)       |
| parnassiana            |                                                                              |               |               |                                      |
| Graziana alpestris     | Spring at the Porra River, Liguria, Savona, Molino, Italy (44.219, 8.255) | AF367673      | AF367641      | Wilke et al. (2001)                  |
| Grossuana delphica     | Kastalia Spring at Delphi, Greece (38.48306, 22.50528)                    | EF061917      | EF061922      | Szarowska et al. (2007)              |
| Hauffenia tellinii     | Isonzo River near Sagrado Spring, Friuli-Venetia Giulia, Gorizia, Italy (45.8743, 13.4856) | AF367672      | AF367640      | Wilke et al. (2001)                  |
| Horatia klecakiana     | Studenci Spring, N of Kućiće, in the valley of the Cetina River, Croatia (43.44881, 16.80708) | KJ159127      | KJ159128      | Szarowska & Falniowski (2014)        |
| Iberhoratia morenoi    | Prado del Rey Spring, Cádiz, Spain (36.66042, -5.45028)                   | MH348101      | MH350210      | Delicado et al. (2019)               |
2006 and Milesiana Arconada & Ramos, 2006; Quiñonero-Salgado & Rolán (2017) for shells of Navalis Quiñonero-Salgado & Rolán, 2017; Boeters et al. (2019) for Salaeniella Boeters, Quiñonero-Salgado & Ruiz-Cobo, 2019; Arconada & Ramos (2002) for Spathogyna Arconada & Ramos, 2002; Ramos et al. (2000) for Tarraconia Ramos & Arconada, 2000; Falniowski & Szarowska (2011a) for Graecoarganiella Falniowski & Szarowska, 2011; Radea et al. (2013) for Isimerope Radea & Parmakelis, 2013; Radea et al. (2016) for Myrtoessa Radea, 2016 and Bodon et al. (2001) for the remaining genera. Character and character states were coded following the terminology of Hershler & Ponder (1998).

**Results**

**Molecular analyses**

The data matrix constructed of COI (658 bp) and 18S (492 bp) sequences yielded an alignment with a total length of 1150 bp. Our ML and BI analyses generated similar tree topologies and branch supports (Fig. 1). In both inferences, Arganiella did not form a monophyletic group; instead, both A. wolfi and A. tabanensis were distantly related to the type species A. pescei. Also in both analyses, A. wolfi resolved as the sister taxon to the genus Iberhoratia from the Iberian Peninsula (BS = 100%, BPP = 1.00), and A. tabanensis, to the genus Kerkia Radoman, 1978 from the Balkan Peninsula and adjacent islands (BS = 91%, BPP = 0.95). The phylogenetic position of A. pescei was not well resolved in either of the phylogenetic analyses. Sequence divergence among the three species of Arganiella ranged from 14.5 to 16.7% for COI and 2.0 to 3.8% for 18S. Divergence among genera ranged from 9.1 to 22.5% for COI and 0 to 4.5% for 18S.

Overall, our results show that DNA sequence divergence values among the three species of Arganiella fall within the range of inter-generic variation and that both A. wolfi and A. tabanensis not only have a sister group relationship with a genus from their respective Mediterranean peninsulas (Fig. 1) but also
clear morphological differences with these sister genera, indicating the two species are representatives of distinct genera (Table 2). This evidence, altogether, demonstrates the need to revise the genus assignment of the non-type species *A. wolfi* and *A. tabanensis*.

**Taxonomy**

Phylum Mollusca Cuvier, 1795  
Class Gastropoda Cuvier, 1795  
Superorder Caenogastropoda Cox, 1960  
Superfamily Truncatelloidea Gray, 1840  
Family Hydrobiidae Stimpson, 1865

Genus *Aretiana* Delicado & Ramos gen. nov.  
urn:lsid:zoobank.org:act:7B207B6D-47E8-4211-8139-0CF98DA353D7

Type species  
*Boetersiella wolfi* Boeters & Glöer, 2007.

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**Fig. 1.** Maximum likelihood tree based on the combined (COI, 18S) dataset. Statistical support of the nodes is indicated when Bayesian posterior probabilities $\geq 0.95$ and bootstrap supports $\geq 75\%$ (black dots). On the right, the morphology of shell, penis and distal female genitalia is presented for: a) *Arganiella wolfi* Boeters & Glöer, b) *A. pescei* Giusti & Pezzoli, 1980 and c) *A. tabanensis* Boeters, Glöer & Pešić, 2014. Reproductive organs were re-drawn from Arconada & Ramos (2007a), Bodon *et al.* (2001) and Boeters *et al.* (2014) for the three species, respectively. Scale bar below topology: substitutions per site.
Revised diagnosis
Shell trochiform; whorls 3.5–4.0; aperture complete, rounded; outer lip narrow, straight in lateral profile; umbilicus wide. Operculum corneous, yellowish, thin, pliable, oval to rounded, paucispiral with a central nucleus, without peg. Two pairs of basal cusps on each central radular tooth. Ctenidium well developed, with approximately 14 gill filaments. Osphradium positioned opposite to approximately the middle of the ctenidium. Stomach without gastric caecum; rectum forms a gentle U-shape in the mantle cavity. Bursa copulatrix medium-sized, pyriform, pedunculated and protruding beyond the posterior edge of the albumen gland; bursal duct shorter than bursal length; unpigmented renal oviduct that makes a complete loop over the pallial gland; one elongated or pyriform seminal receptacle arising from the renal oviduct, just above the insertion point with the bursal duct. Prostate gland bean-shaped, about twice as long as wide. Penis small and simple, gradually tapering. Nervous system unpigmented.

Etymology
The genus is named after Villa Aretiana, the Roman name of the town Aracena, which gives its name to the mountain range where the genus was found (i.e., Sierra de Aracena); gender feminine.

Remarks
Aretiana Delicado & Ramos gen. nov. can be distinguished from Arganiella as the former has a taller shell, a more oval operculum, pigmentation on the body and eyespots, fewer gill filaments, a narrower penis, a larger and pyriform bursa copulatrix located beyond the posterior edge of the albumen gland and fewer cusps on the lateral radular teeth (for comparison, see Giusti & Pezzoli 1980; Boeters & Glöer 2007; Arconada & Ramos 2007a). The new genus differs from the closely related genus Iberhoratia by its taller shell with a narrower umbilicus, the absence of lobes on the inner edge of the penis and of a proximal seminal receptacle (SR2) and presence of two pairs of basal cusps on each central radular tooth (see Arconada et al. 2007). Mean COI divergence for Aretiana Delicado & Ramos gen. nov. was 15.9% with Arganiella and 11.2% with Iberhoratia.

Genus Docleiana Delicado & Pešić gen. nov.
urn:lsid:zoobank.org:act:04639DB3-91D2-46B7-BFAB-FFC1EA3C27BD

Type species
Arganiella tabanensis Boeters, Glöer & Pešić, 2014.

Revised diagnosis
Shell valvatiform; whorls ca 3; aperture complete, from rounded to ellipsoidal; outer lip narrow, straight in lateral profile; umbilicus wide. Operculum corneous, yellowish, thin, pliable, rounded, paucispiral with a central nucleus, without peg. Two pairs of basal cusps on each central radular tooth. Ctenidium well developed, with 10–11 gill filaments. Animal unpigmented. Osphradium positioned opposite approximately to the middle of the ctenidium. Stomach without gastric caecum; rectum forms a gentle V-shape in the mantle cavity. Bursa copulatrix small, globular, pedunculated and positioned beyond the posterior edge of the albumen gland; bursal duct longer than bursal length; unpigmented renal oviduct; one pyriform seminal receptacle arising at the insertion point with the bursal duct loop. Penis small and simple, gradually tapering.

Etymology
The new genus is named after Doclea, the name of the Roman city located on the site of modern Podgorica on whose municipal territory a new genus was found; gender feminine.
Remarks

Docleiana Delicado & Pešić gen. nov. can be distinguished from Arganiella as the former has a more ellipsoidal shell aperture, a narrower base of the penis, a smaller bursa copulatrix that is positioned beyond the posterior edge of the albumen gland, a larger seminal receptacle and more cusps on the lateral radular teeth (Fig. 2; for comparison, see Giusti & Pezzoli 1980; Boeters et al. 2014). The new genus differs from the closely related genus Kerkia by its smaller shell, absence of lobes on the inner edge of the penis, a smaller bursa copulatrix, a more globular seminal receptacle and V-shaped rectum (see Radoman 1978; Bodon et al. 2001). Mean COI sequence divergence for Docleiana Delicado & Pešić gen. nov. was 14.5% with Arganiella and 12.3% with Kerkia.

Discussion

Previous taxonomic studies of valvatiform hydrobiids have relied on morphological similarities to classify species, a practice that can be difficult for this group because of their small size and simplified morphologies. Considering this, more recent systematic revisions have incorporated molecular
methods to re-evaluate the taxonomy of valvatiform hydrobiid species and have indeed revealed genus
misidentifications. Some examples are the species Hauffenia plana (Bole, 1961), later transferred to the
genus Bracenica Radoman, 1973 by Hofman et al. (2020); Horatia hadei Gittenberger, 1982 transferred to
Daphniola Radoman, 1973 by Falniowski & Szarowska (2011b); and Neohoratia azarum Boeters &
Rolán, 1988, later recognized as Islamia azarum (Boeters & Rolán, 1988) by Arconada & Ramos
(2006) and currently combined as Deganta azarum by Arconada & Ramos in Delicado et al. (2019).
In the case of Arganiella, A. wolfi and A. tabanensis, two endemic species from the Iberian and Balkan
peninsulas, respectively, were classified within this genus on the basis of shell and genital similarities
with the Apennine species A. pescei (Arconada & Ramos 2007a, 2007b; Boeters &
Rolán, 1988), which is only known by empty shells, and Arganiella Delicado & Ramos gen. nov. is
-distances 9.1–22.5% for COI). Moreover,
Arganiella Delicado & Ramos gen. nov. is
-in shell size, the shape of the
Arganiella Delicado & Pešić gen. nov. as an independent lineage sister
to the Balkan genus Kerkia, this relationship may change as more valvatiform species are sequenced
and analysed. Until then, morphological characters support the placement of A. tabanensis into a new
genus. The Balkan genera Hadziella Kuščer, 1932 and Dabriana Radoman, 1974, which have not yet

However, incorrect systematic conclusions can be drawn from molecular phylogenies when the
specimens have previously been misidentified (e.g., Radomaniola/Horatia in Szarowska & Falniowski,
2014). Detailed morphological examinations of the studied material are therefore needed for a more
reliable systematic interpretation. Our comparative morphological study (Table 2) indicated close
similarities among the, until now, considered species of Arganiella, especially in those characters related
to the penis, ctenidium and radula and, to a lesser degree, the shell, pigmentation and distal genitalia
of females. Some character states found in species of Arganiella, such as a simple penis without lobes,
the presence of a single distal seminal receptacle and two cusps at both sides of the basis of the central
radular tooth, are rarely present in other valvatiform genera (Bodon et al. 2001; Radea et al. 2016) and
can, thus, lead to genus misidentification. However, differences in other characters can be observed
among the three species. The most dissimilar species is A. wolfi: it has a larger and more trochiform
shell, body pigmentation and a larger bursa copulatrix (Arconada & Ramos 2007a; Boeters & Glöer
2007). Arganiella tabanensis is more similar to A. pescei than A. wolfi, especially in shell shape and
body pigmentation (Boeters et al. 2014). However, it differs from A. pescei in shell size, the shape of the
bursa copulatrix and the bend type of the rectum. On the basis of this DNA and morphologic evidence,
we assign the species A. wolfi and A. tabanensis to two distinct genera.

We base the establishment of the two new genera, Aretiana Delicado & Ramos gen. nov. for Arganiella
wolfi and Docleiana Delicado & Pešić gen. nov. for Arganiella tabanensis, on the high level of DNA
sequence divergence and phylogenetic position of the two species (Fig. 1), as well as on their unique
combination of morphological characters (Table 2). Given the restricted geographic distribution of
most valvatiform hydrobiids (Bodon et al. 2001), we focus our taxonomic decision on comparisons
with geographically close groups. Most genera of Iberian valvatiform hydrobiids were included in our
phylogenetic study, except Navalis, which is only known by empty shells, and Salaeniella, for which
the females are still unknown and no well-preserved specimens are available for molecular studies.
Regardless, these genera are morphologically very distinct from Aretiana Delicado & Ramos gen. nov.
and have distant distribution ranges within the Iberian Peninsula (Quiñonero-Salgado & Rolán 2017;
Boeters et al. 2019). According to our comparative morphological study, the most similar Iberian genus to
Aretiana Delicado & Ramos gen. nov. is Boetersiella. These two genera are highly divergent genetically
and display clear differences in shell and female genital features, validating them as different genera.
DNA sequence data of valvatiform genera living in the Balkan springs are still scarce. Although our
phylogenetic analyses recovered Docleiana Delicado & Pešić gen. nov. as an independent lineage sister
to the Balkan genus Kerkia, this relationship may change as more valvatiform species are sequenced
and analysed. Until then, morphological characters support the placement of A. tabanensis into a new
genus. The Balkan genera Hadziella Kuščer, 1932 and Dabriana Radoman, 1974, which have not yet
Table 2 (continued on next page). Morphological characters recorded from the valvatiform hydrobiid genera distributed over the Iberian, Apennine and Balkan peninsulas. This data set is an extension of table 2 included in Radea et al. (2016).

| Taxa                      | SL  | SW  | SK  | UW  | AS  | OP  | BaC | BC  | SR(s) | SRP | P   | PL  | PP  | PS  | Ct  | Eyes | Rectum |
|---------------------------|-----|-----|-----|-----|-----|-----|-----|-----|-------|-----|-----|-----|-----|-----|-----|-------|---------|
| **Iberian Peninsula**     |     |     |     |     |     |     |     |     |       |     |     |     |     |     |     |       |         |
| *Arctiana* gen. nov.      | 1.7 | 1.7 | 0   | 2   | 0   | 0   | 2   | 3   | 1     | 0   | 0   | 0   | 0   | 0   | 1   | 1     | S       |
| Boetersiella              | 1.6 | 1.5 | 0   | 1   | 1   | 0   | 1   | 2   | 1     | 0   | 0   | 0   | 0   | 0   | 0   | 1     | U       |
| Chondrobasis              | 1.3 | 1.2 | 0   | 1   | 1   | 0   | 1   | 3   | 1     | 0   | 1   | 1   | 0   | 0   | 0   | 1     | U       |
| Corbellaria               | 0.5 | 1.3 | 0   | 2   | 0   | 0   | 1   | 3   | 2     | 2   | 1   | 2   | 0   | 0   | 0   | 0     | S       |
| Deganta                   | 1.7 | 1.6 | 0   | 1   | 0   | 0   | 2   | 3   | 1     | 0   | 2   | 1   | 2   | 0   | 0   | 0     | U       |
| Iberhoratia               | 1.5 | 1.8 | 0   | 2   | 0   | 0   | 1   | 1   | 2     | 2   | 1   | 2   | 0   | 0   | 1   | 1     | U or S  |
| Josefus                   | 1.5 | 1.6 | 0   | 1   | 1   | 0   | 2   | 0   | 2     | 1   | 1   | 4   | 0   | 0   | 0   | 0     | U       |
| Milesiana                 | 1.1 | 1.5 | 0   | 2   | 0   | 0   | 2   | 0   | 2     | 1   | 1   | 2   | 0   | 0   | 1   | 1     | U       |
| Navalis                   | 0.8 | 1.0 | 1   | 2   | 0   | –   | –   | –   | –     | –   | –   | –   | –   | –   | –   | –     | –       |
| Salaeniella               | 1.2 | 1.6 | 0   | 2   | 1   | 0   | –   | –   | –     | –   | –   | –   | –   | –   | –   | –     | –       |
| Spathogyna                | 1.0 | 1.4 | 0   | 2   | 1   | 0   | 1   | 1   | 2     | 2   | 1   | 2   | 0   | 0   | 1   | 1     | V       |
| Tarraconia                | 1.8 | 1.8 | 0   | 2   | 0   | 0   | 1   | 3   | 0     | 0   | 1   | 2   | 0   | 0   | 1   | 1     | U       |
| **Apennine Peninsula**    |     |     |     |     |     |     |     |     |       |     |     |     |     |     |     |       |         |
| *Arganiella*              | 1.0 | 2.0 | 0   | 2   | 0   | 0   | 2   | 1   | 1     | 0   | 0   | 0   | 0   | 0   | 1   | 0     | S       |
| Fissuria*                 | 1.6 | 1.8 | 0   | 2   | 0   | 0   | 1   | 1   | 2     | 2   | 3   | 1   | 3   | 1   | 0   | 1     | 0       |
| Hauffenia*                | 1.2 | 2.3 | 0   | 2   | 0   | 1   | 1   | 3   | 1     | 0   | 0   | 0   | 0   | 1   | 0   | 1, 0   | Z       |
| Pezzolia                  | 1.0 | 1.4 | 0   | 2   | 1   | 0   | 0   | 3   | 2     | 2   | 0   | 0   | 0   | 0   | 0   | 0     | S       |
| **Balkan Peninsula**      |     |     |     |     |     |     |     |     |       |     |     |     |     |     |     |       |         |
| *Docleiana* gen. nov.     | 1.1 | 1.5 | 0   | 2   | 0   | 0   | 2   | 2   | 1     | 0   | 0   | 0   | 0   | 0   | 1   | 0     | Z       |
| Bracenica                 | 1.0 | 2.0 | 0   | 2   | 0   | 1   | 1   | 3   | 2     | 2   | 1   | 2   | 0   | 0   | –   | 0     | –       |
| Dabriana                  | 2.2 | 2.5 | 0   | 2   | 0   | –   | 0   | 3   | 1     | 0   | 0   | 0   | 0   | 0   | 0   | 0     | –       |
| Dalmatella                | 1.6 | 2.2 | 1   | 2   | 0   | –   | –   | –   | –     | –   | –   | –   | –   | –   | –   | –     | –       |
| Daphniola                 | 1.3 | 1.2 | 0   | 1   | 1   | 0   | 1   | 1   | 2     | 2   | 1   | 2   | 0   | 0   | 1   | 1     | –       |
| Gocea                     | 0.9 | 1.6 | 0   | 2   | 1   | 1   | 1   | 3   | 1     | 0   | 0   | 0   | 0   | 0   | 0   | 0     | Z       |
| Graecoarganiella          | 1.2 | 1.5 | 0   | 2   | 1   | 0   | 1   | 3   | 2     | 2   | 1   | 1   | 1   | 0   | 0   | 1     | –       |
**Table 2** (continued).

| Taxa          | SL | SW | SK | UW | AS | OP | BaC | BC  | SR(s) | SRP | P | PL | PP | PS | Ct | Eyes | Rectum |
|---------------|----|----|----|----|----|----|-----|-----|-------|-----|---|----|----|----|----|-------|---------|
| *Hadziella*   | 0.6| 1.8| 0  | 2  | 1  | 1  | 1   | 1   | 1     | 0   | 0 | 0  | 0  | 0  | 0  | 0     | Z       |
| *Horatia*     | 2.1| 2.1| 0  | 1  | 1  | 0  | 1   | 1   | 2     | 2   | 1 | 2  | 0  | 0  | 1  | 1     | 0       |
| *Isimerope*   | 1.6| 2.0| 0  | 2  | 1  | 0  | 1   | 3   | 0     | 0   | 1 | 3  | 1  | 0  | 0  | 1     | U       |
| *Islamia*     | 2.0| 1.8| 0  | 0  | 0  | 0  | 1   | 0   | 2     | 2   | 1 | 4  | 0  | 0  | 1  | 1     | U       |
| *Karevia*     | 1.1| 1.7| 1  | 2  | 2  | 0  | 1   | 3   | 2     | 2   | 1 | 3  | 0  | 0  | –  | 1     | –       |
| *Kerzia*      | 1.4| 2.3| 0  | 2  | 0  | 1  | 2   | 1   | 1     | 0   | 1 | 3  | 0  | 0  | 1  | 0     | S       |
| *Lynhidia*    | 1.2| 1.1| 0  | 0  | 2  | 0  | 1   | 1   | 1     | 0   | 1 | 4  | 0  | 0  | –  | 1     | –       |
| *Myrrhoa*     | 1.1| 1.4| 0  | 2  | 0  | 0  | 1   | 1   | 2     | 2   | 0 | 0  | 2  | 0  | 1  | 1     | V       |
| *Ohrigocea*   | 1.0| 1.5| 1  | 2  | 1  | 0  | 1   | 1   | 2     | 2   | 1 | 3  | 0  | 0  | –  | 1     | –       |
| *Prespolio*   | 1.0| 1.3| 0  | 2  | 0  | 0  | 1   | 1   | 2     | 2   | 1 | 3  | 0  | 0  | –  | 1     | –       |
| *Pseudo*      | 1.3| 1.4| 0  | 1  | 0  | 1  | 1   | 2   | 2     | 2   | 1 | 3  | 0  | 0  | 1  | 1     | 0       |
| *Pseudosl*    | 1.1| 1.3| 0  | 2  | 1  | 0  | 1   | 1   | 2     | 2   | 1 | 3  | 0  | 0  | –  | 1     | –       |
| *Strugia*     | 1.6| 1.7| 0  | 2  | 0  | 0  | 1   | 1   | 1     | 0   | 1 | 3  | 0  | 0  | –  | 1     | –       |
| *Zuoria*      | 1.5| 1.2| 0  | 0  | 0  | 0  | 1   | 1   | 1     | 0   | 1 | 4  | 0  | 0  | –  | 0     | –       |

* Genera distributed outside of the categorized regions.

Continuous (in mm) and discrete (or categorical) characters: AS = aperture shape: rounded (0), oval (1), square (2); BaC = pairs of basal cusps on the central radular tooth; BC = bursa copulatrix: absent (0), ovoid (1), globular (2), pyriform (3); Ct = ctenidium: absent (0), present (1); eyes: absent (0), present (1); OP = operculum peg: absent (0), present (1); P = penis: simple without lobes (0), with one lobe (1), with two lobes (2), with more than two lobes (3); PL = penial lobe(s): absent (0), basal lobe (1), medial lobe (2), lobe at ½ of penis length (3), apical lobe (4); PP = penial papilla: absent (0), present (1); PS = penial stylet: absent (0), present (1); Rectum: without bend (0), S-like (S), U-like (U), V-like (V), Z-like (Z); SK = shell keels or other ornaments: absent (0), present (1); SL = shell length (maximum recorded value for the type species); SR = seminal receptacle: absent (0), one seminal receptacle (1), two seminal receptacles (2); SRP = seminal receptacle position: absent (0), close to each other (1), distantly positioned from each other (2); SW = shell width (maximum recorded value for the type species); UW = umbilicus width: very narrow (0), narrow (1), wide (2); – = no data. Continuous variables provided in mm.
been sequenced, resemble *Docleiana* Delicado & Pešić gen. nov. in penis morphology but differ in shell size and radular and female genital features; they also lack a ctenidium (Table 2).

The systematic findings suggesting erroneous assignment of the three geographically disjunct species to the same genus due to morphological similarities conflict with the use of traditional taxonomy to classify valvatiform hydrobiid taxa and highlight the need to integrate morphological and molecular data for more robust taxonomic assessments.

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