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A NEW SPECIES OF *PSEUDAMNICOLA* PAULUCCI, 1878 FROM KITHIRA ISLAND, GREECE

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ABSTRACT: A new species of *Pseudamnicola, P. magdalenae*, is described from the spring at Karavas on Kithira Island, Greece. Cytochrome oxidase subunit I (*COI*) sequences of mtDNA indicate its species distinctness. The shell, female reproductive organs and penis are described. The shell resembles that of the geographically close *P. macrostoma* (Küster, 1853) from the Greek mainland, but the penis is more triangular and the bursa copulatrix is bigger in *P. magdalenae*. The simplified anatomy coupled with the wide variation overlapping between the congeneric species is emphasised.

KEY WORDS: molecular distinctness, mtDNA, cytochrome oxidase subunit I (*COI*), morphological distinctness, shell, penis, female reproductive organs

INTRODUCTION

The genus *Pseudamnicola* Paulucci, 1878 contains species which are known from the British Isles, the Netherlands and Spain, through France, Italy and the southern Balkans, to Romania (although the latter not confirmed anatomically), Turkey, Cyprus and some other circum-Mediterranean countries (e. g. DELICADO & RAMOS 2012, DELICADO et al. 2014), but the systematics of this group is poorly understood (SZAROWSKA et al. 2015).

In Greece, six nominal species of *Pseudamnicola* were distinguished (SCHÜTT 1980, SZAROWSKA et al. 2006), four of them inhabiting the islands [*P. brachia* (Westerlund, 1886), *P. pieperi* Schütt, 1980, *P. chia* (Martens, 1889) and *P. negropontina* (Clessin, 1878)], and two known from continental Greece [(*P. macrostoma* (Küster, 1853) and *P. exilis* (Frauenfeld, 1863)]. However, all of them were distinguished based solely on shell characters. Recently, RADEA et al. (2016) described two more species from Rhode island (*P. ianthe* Radea et Parmakelis, 2016 and *P. ilione* Radea et Parmakelis, 2016), considering molecular data as well. The taxonomy of the genus is poorly understood, the shells are very variable and the anatomy is simple and variable even within a population. Thus the morphology alone may not resolve the systematics of this group and molecular data are required.

In the Truncatelloidea, morphological characters alone are sometimes insufficient even for proper determination of a genus (e.g. SZAROWSKA & FALNIOWSKI 2008, FALNIOWSKI et al. 2012) and all the characters are highly variable within species, the variation ranges overlapping between the species (FALNIOWSKI 1987). For the Greek *Pseudamnicola* molecular data – mostly mtDNA cytochrome oxidase subunit I (*COI*) – are already available for several taxa (SZAROWSKA et al. 2006, 2015, SZAROWSKA & FALNIOWSKI 2011, RADEA et al. 2013, 2016). Molecular data suggest that the number of species of the Greek *Pseudamnicola* described so far is underestimated – at least 16 lineages of presumably species rank have been found in Greece (SZAROWSKA et al. 2015). One of the most distinct lineages inhabits the spring at Karavas on Kithira Island.

MATERIAL AND METHODS

About fifty specimens of *Pseudamnicola* were collected from the spring at Karavas, northern part of Kithira Island; 36°20'50.4"N, 22°56'57.8"E, 66 m a.s.l. In this big spring, in the form of a medium-sized pond, *Pseudamnicola* co-occurred with *Melanopsis*. Snails were collected by hand or with a sieve, and fixed with 80% ethanol. The shells and the soft body parts were photographed with a CANON EOS 50D digital camera, under a NIKON SMZ18 microscope with dark field and phase contrast. Five males and five females were dissected; photographs of the soft parts were used for drawing the features of internal anatomy. Character state terminology follows that defined by HERSHLER & PONDER (1998).

SYSTEMATIC PART

Family: Hydrobiidae Troschel, 1857

Subfamily: Hydrobiinae Troschel, 1857

Genus: *Pseudamnicola* Paulucci, 1878

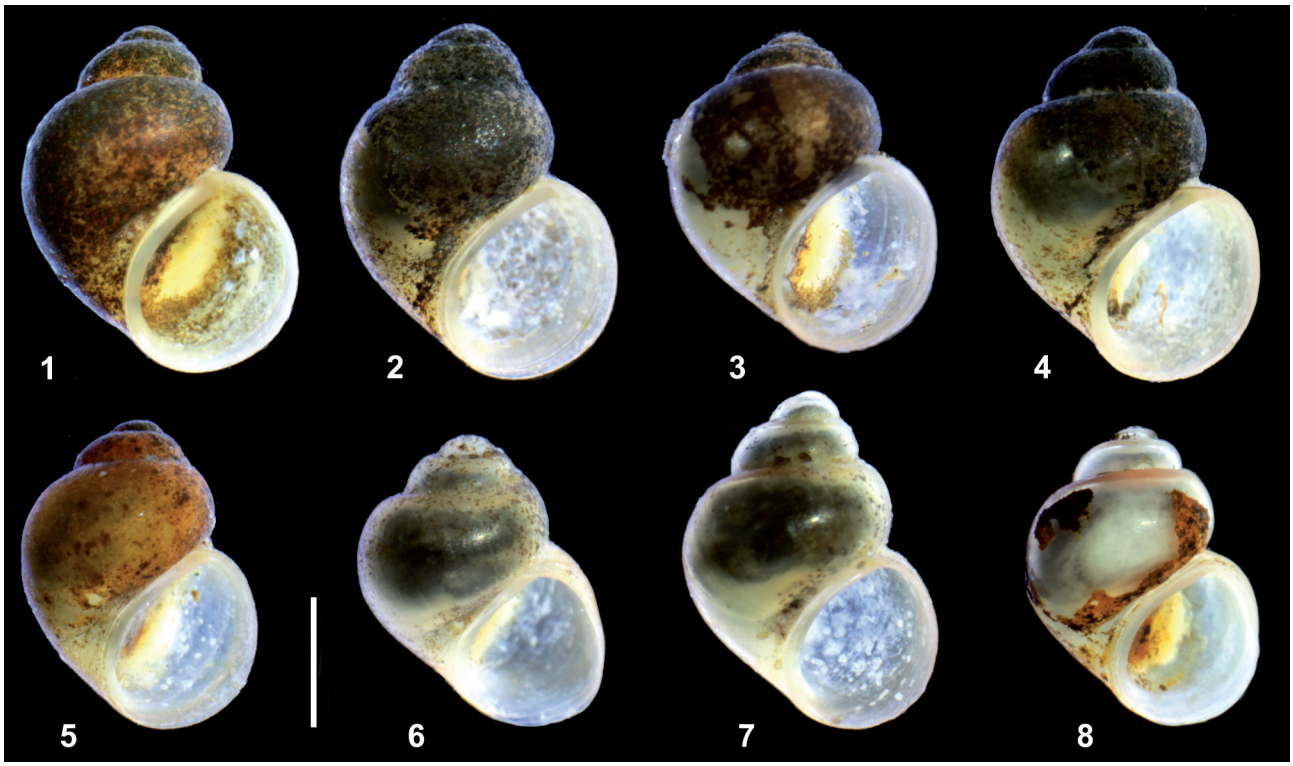
Pseudamnicola magdalenae sp. n.

Types. Ethanol-fixed specimens, huge spring forming a pond at Karavas, N. part of Kithira Island; Greece, 36°20'50"N, 22°56'58"E, 66 m a.s.l.; September 2009, holotype ZMUJ-M.1923, 20 paratypes: ZMUJ-M.1924-ZMUJ-M.1943. Eight sequences of mitochondrial cytochrome oxidase subunit I (*COI*) gene, GenBank numbers: JF921906-JF921913.

Diagnosis. Shell trochiform with relatively high spire and simple outer lip, soft parts pigmented black, female reproductive organs with big bursa copulatrix and long loop of oviduct, penis simple, big, broad-

Table 1. Measurements of the holotype and n=10 paratypes

	shell height (mm)	spire height (mm)	body whorl width (mm)	aperture height (mm)	aperture width (mm)	number of whorls
holotype	3.04	0.56	1.93	1.70	1.42	5.2
mean	2.65	0.42	1.69	1.56	1.30	4.75
SD	0.2073	0.0839	0.1123	0.0998	0.0941	0.3375
minimum	2.32	0.25	1.52	1.36	1.16	4.2
maximum	3.04	0.56	1.93	1.70	1.42	5.2



Figs 1–8. Shells of *Pseudamnicola magdalenae*: 1 – holotype, 2–8 – paratypes. Bar equals 1 mm

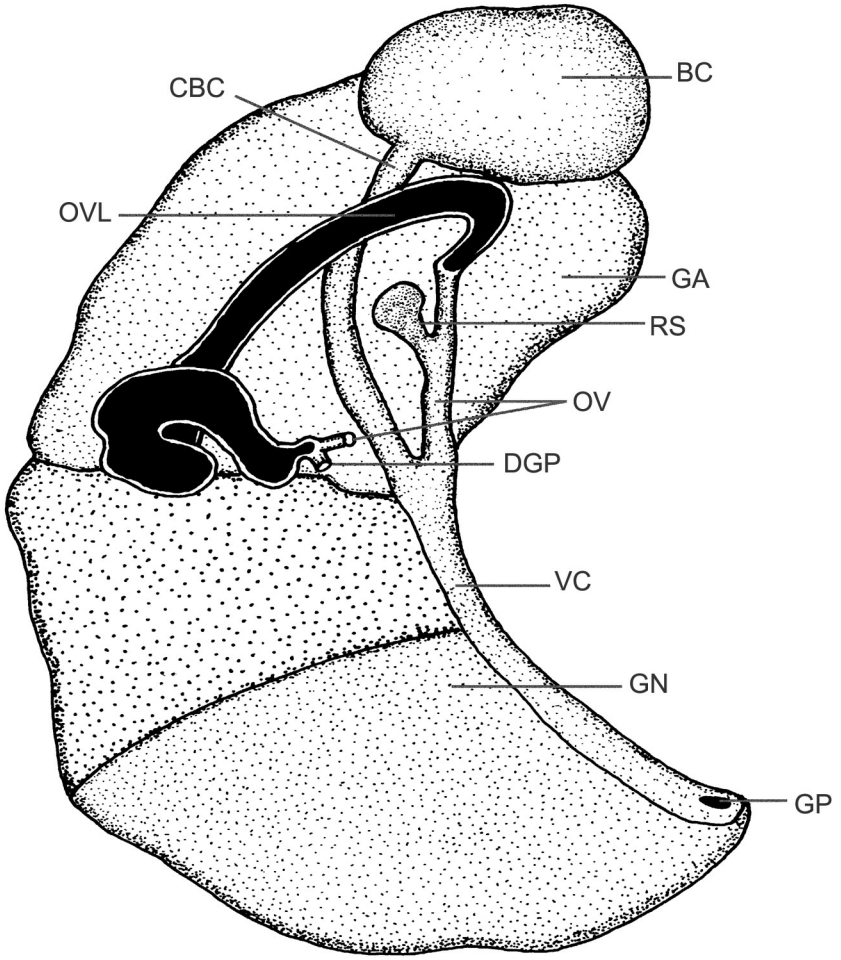


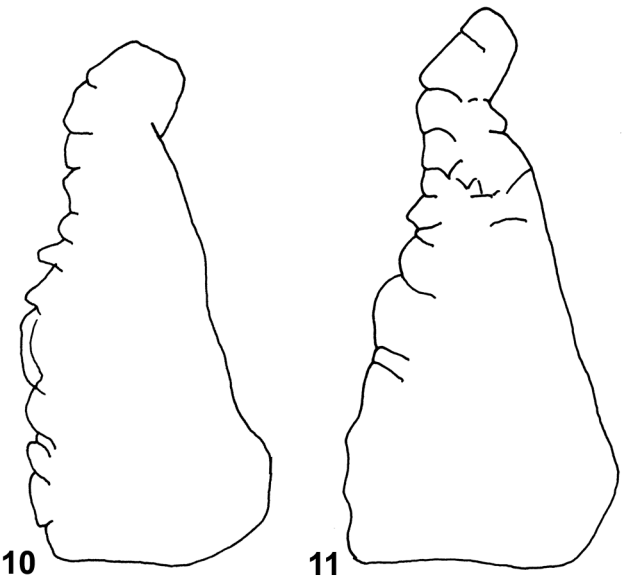
Fig. 9. Renal and pallial section of the female reproductive system of *Pseudamnicola magdalenae*; abbreviations: BC – bursa copulatrix, CBC – duct of bursa copulatrix, DGP – gono-pericardial duct, GA – albumen gland, GN – capsule gland, GP – gonoporus, OV – oviduct, OVL – coil (spiral) of (renal) oviduct, RS – seminal receptacle, VC – ventral channel

ly triangular without any outgrowth. Easily distinguished from *P. macrostoma*, geographically close and with a similarly-shaped shell, by the more triangular penis and the bigger bursa copulatrix. Shell spire in *P. magdalenae* lower than in *P. brachia* and higher than in *P. exilis*.

Description. Shell (Figs 1–8) trochiform, up to 3.04 mm high, having 4.2–5.2 whorls, spire height approximately 16% of shell height and 24.5% of body whorl width. Teleoconch whorls moderately convex, evenly rounded, growing rapidly in diameter. Aperture circular, outer lip simple, parietal lip complete, umbilicus very broad or absent. Teleoconch with delicate growth lines, periostracum pinkish or white. Shell parameters of the holotype and ten paratypes are given in Table 1. Operculum smooth on its inner and outer surface, yellowish.

Soft parts densely pigmented black. Female reproductive organs (Fig. 9) with big, spherical bursa with long duct, one small and bulky receptaculum seminis in the position of rs_1 and long loop of oviduct pigmented black.

Penis simple, big, broadly triangular without any outgrowth (Figs 10–11).



Figs 10–11. Penes of *Pseudamnicola magdalenae*

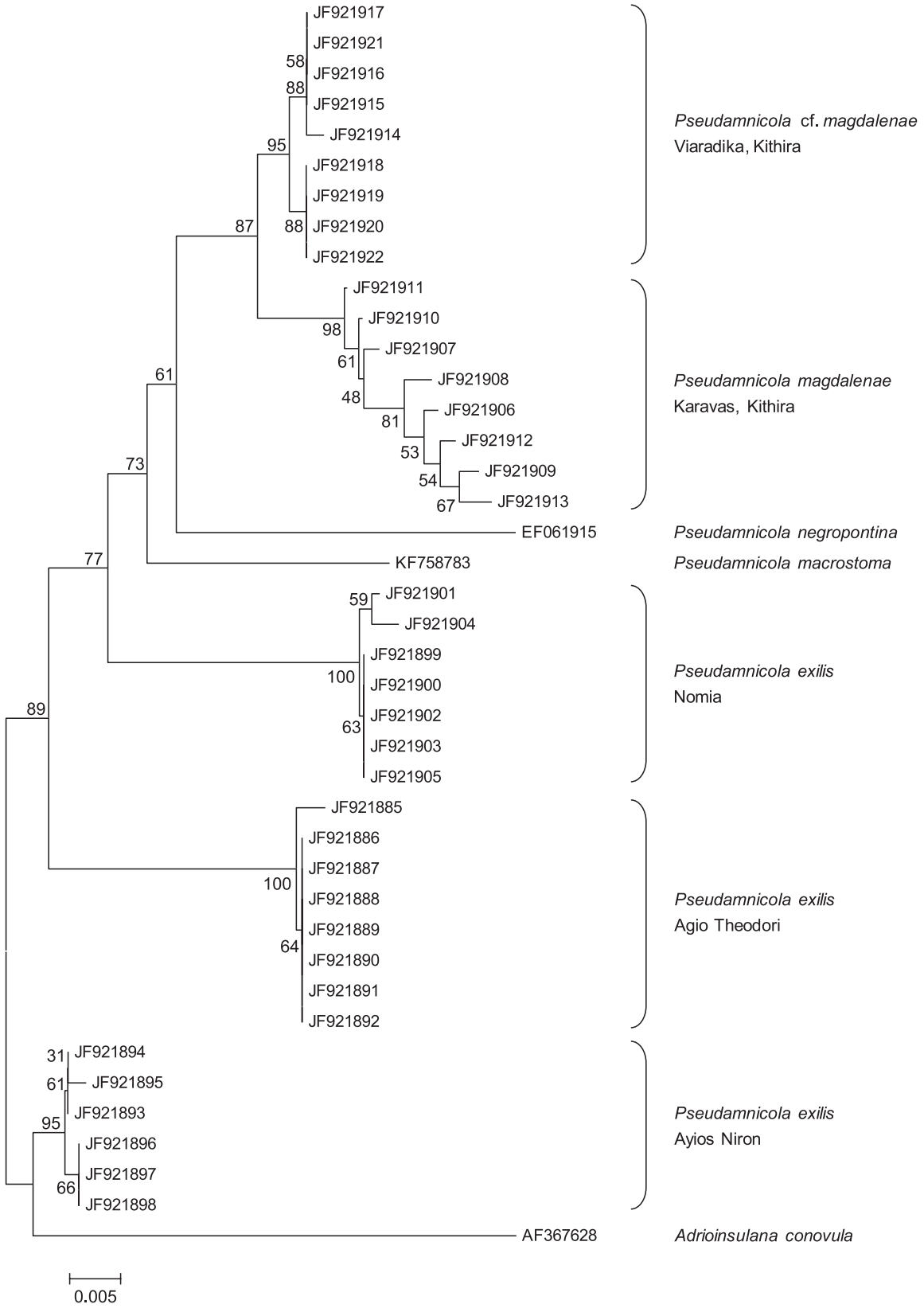


Fig. 12. Neighbour-Joining tree based on the 602-nt-long fragment of *COI* sequences from GenBank: JF921885-JF921922 for five populations of *Pseudamnicola exilis* (SZAROWSKA & FALNIOWSKI 2011); AF367628 for *Adrioinsulana conovula* (WILKE et al. 2001); EF061915 for *P. negropontina* (SZAROWSKA et al. 2006) and KF758783 for *P. macrostoma* (RADEA et al. 2013). The sequences were analysed using the Neighbour-Joining method (SAITOU & NEI 1987) implemented in MEGA6 (TAMURA et al. 2013) using the Kimura two-parameter model (K2P) for pairwise distance calculations (KIMURA 1980). NJ tree branches were supported by bootstrap analysis with 1,000 replicates (FELSENSTEIN 1985).

Etymology. Named in memory of Dr Magdalena Szarowska, a malacologist, my wife and best friend, who loved Greece.

Distribution and habitat. Known from the type locality only. However, considering the relatively small genetic distance (p = 0.02), the population inhabiting the spring at Viaradika, central Kithira Island, may belong to this species.

DISCUSSION

As already stated in the Introduction, external morphological features within the Truncatelloidea usually lack the states which are unique or characteristic at least; almost always the range of variation is wide and overlaps between the species or even genera (e.g. FALNIOWSKI 1987, SZAROWSKA & FALNIOWSKI 2008, 2011, FALNIOWSKI et al. 2012, SZAROWSKA et al. 2015). Thus, considering only morphology, *P. magdalenae* cannot always be distinguished from the other Greek *Pseudamnicola*. However, p-distance values (0.021–0.063) between *P. magdalenae* and the other presumed species of the Greek *Pseudamnicola* (SZAROWSKA et al. 2015) are within the range of values recorded for congeneric species of Truncatelloidea. The long-lasting isolation of Kithira from the mainland or from any other island most probably resulted in species distinctness of *P. magdalenae*, clearly visible in the maximum likelihood tree (Fig. 12, see also: SZAROWSKA et al. 2015: fig. 1). The *Pseudamnicola* population in the spring at Viaradika in central Kythira may also belong to *P. magdalenae*. It is noteworthy that at Viaradika a relatively small spring was inhabited by dozens of thousands of specimens, but only three *COI* haplotypes were found there. On the other hand, the spring at Karavas formed a large pond, inhabited by very few *Pseudamnicola*, but as many as eight *COI* haplotypes were found there. This was the highest number of haplotypes found at any locality in the Greek *Pseudamnicola* (SZAROWSKA et al. 2015).

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