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## Monographic revision of the endemic *Helix mazzullii* De Cristofori & Jan, 1832 complex from Sicily and re-introduction of the genus *Ercatella* Monterosato, 1894 (Pulmonata, Stylommatophora, Helicidae)

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### Abstract

*Helix mazzullii* De Cristofori & Jan, 1832 s.l.(Pulmonata, Stylommatophora, Helicidae) is an endemic, rupicolous, saxicolous taxon of northwestern Sicily. Its populations are vulnerable and it is of great ecological significance. However, its taxonomy, phylogeny and biogeography are still uncertain. The present paper reports on a comprehensive analysis of morphological diagnostic characters (shell and genitalia) joined by the molecular study of two mitochondrial (16S rRNA and 12S rRNA) and one nuclear (ITS-2) partial gene sequences, investigated by individual segment analyses or combining the three gene fragments with a concatenate analysis. Our results corroborated the hypothesis that this species is rather a group (i.e., the *mazzullii* group) including three different taxa (*mazzullii*, *cephalaeditana* and *insolida*) recognized as species. Moreover, molecular dating of lineages suggests that this complex might have occurred long before the Messinian salinity crisis. Finally, peculiar morphological and ecological features along with molecular data strongly support the proposal to re-introduce the genus *Ercatella* Monterosato, 1894 for the *H. mazzullii* complex. Synonyms and bibliographic references are reported in the systematic part; collection records are listed in Appendix 1.

**Key words:** Land snails, *mazzullii* group, *Ercatella*, Helicidae, Sicilian endemism, molecular taxonomy, molecular phylogeny, biodiversity, biogeography, Mio-pliocene events

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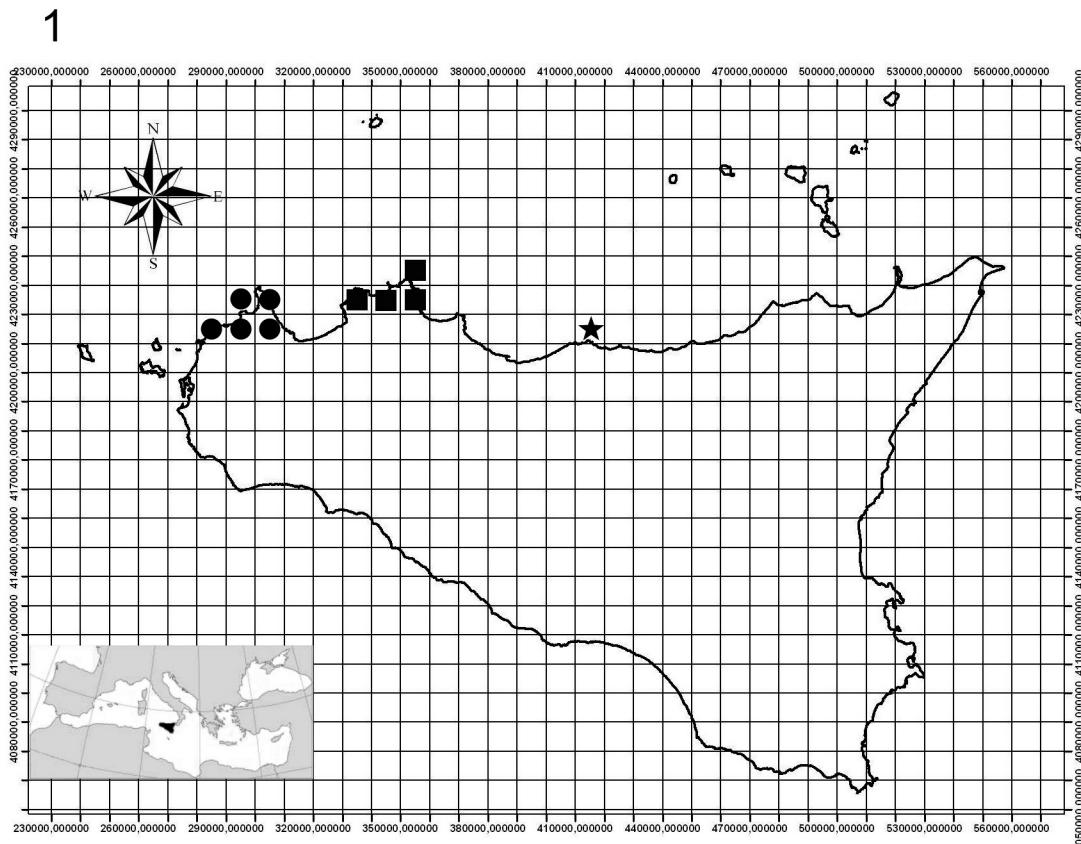
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## Introduction

*Helix mazzullii* De Cristofori & Jan, 1832 is currently considered a single endemic species of northwestern Sicily ranging from Cefalù (Palermo province) to the mountains near Trapani (Alzona 1971; Cesari 1978; Manganelli *et al.* 1995) (Fig. 1). Its restricted distribution range and peculiar biology (it is rupicolous and saxicavous) make it one of the most vulnerable and interesting groups among Mediterranean Helicidae (Manganelli *et al.* 2000).

*Helix mazzullii* shows a high variability in shell morphology which led several authors of the 19th century to establish numerous nominal taxa for these animals.

In the past, *Helix mazzullii* has been included in the genus *Helix* Linné and various other genus group taxa, such as *Helicogena* Féruccac, *Cantareus* Risso, *Cryptomphalus* Moquin-Tandon and *Erctella* Monterosato. Giusti *et al.* (1995) and Manganelli *et al.* (1995), taking into account its genital morphology, elevated *Cantareus* to genus rank (instead of a subgenus of *Helix*), including *C. aspersus*, *C. mazzullii* and *C. apertus*. Other authors maintained *Cantareus* for *apertus* whereas *aspersum* and *mazzullii* were included within the genus *Cornu* Born, 1778 (see Bank *et al.* 2001). This choice, however, raised nomenclatural concerns, since *Cornu* was introduced by Born (1778) as *Cornu copiae*, based on a teratological specimen of *Helix aspersa* O.F. Müller. Because, according to ICZN (1985, Article 1b(2) [equivalent with ICZN 1999 Art. 1.3.2], teratological specimens as such are excluded from the provisions of the code, *Cornu* was rejected as an available genus name by some authors (e.g., Giusti *et al.* 1995). However, Gerber (2000) argued that “because it was not proven that Born used the name *Cornu copiae* in 1778 under the conscience and with the intent to describe a teratological specimen [i.e., “as such”] ICZN (1985) Art. 1b(2) does not apply and *Cornu* thus is an available genus name”. This conclusion was supported by Falkner *et al.* (2001). Currently, although the issue is still contentious, most systematists (e.g., Bank *et al.* 2001; Ketmaier *et al.* 2007; Bank 2009) accept these species as *Cantareus apertus*, *Cornu aspersum* and *Cornu mazzullii*.



**FIGURE 1.** Geographic distribution of *Erctella* species in northwestern Sicily. ● = *E. insolida*; ■ = *E. mazzullii*; ★ = *E. cephaeditana*.

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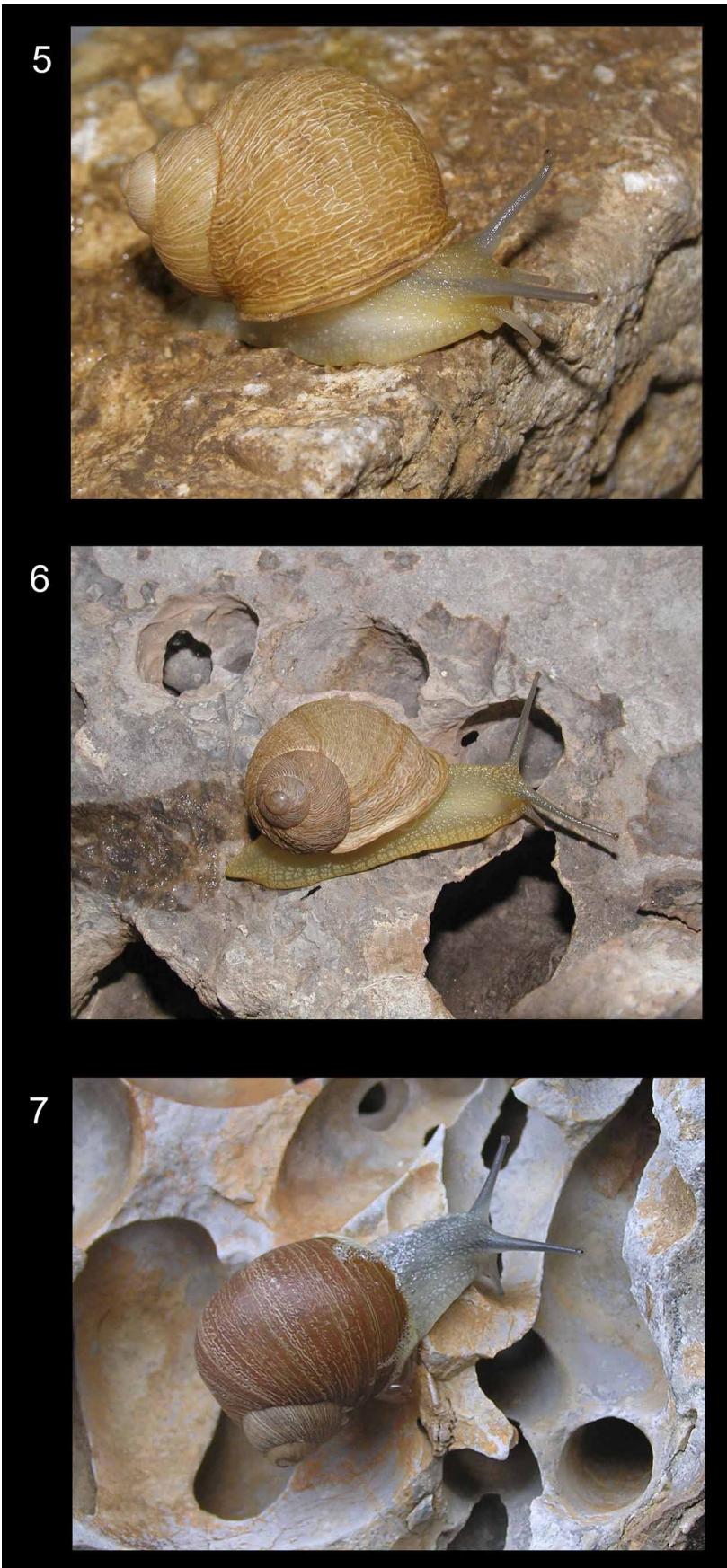
**FIGURE 2.** Landscape of Monte Pellegrino, Palermo.

**FIGURE 3.** Landscape of Cefalù, La Rocca, Palermo.

**FIGURE 4.** Landscape of Monte Cofano, Trapani (by L. Barraco).

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**FIGURE 5.** *Ercitella mazzullii*, Monte Pellegrino.

**FIGURE 6.** *Ercitella cephalaeeditana*, Cefalù.

**FIGURE 7.** *Ercitella insolida*, San Vito lo Capo, Cala Mancina.

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In a previous and preliminary study (Colomba *et al.* 2008), mainly based on anatomical data and molecular analysis of partial mitochondrial 16S rRNA gene sequences, we suggested that *Helix mazzullii* (therein reported as *Cornu mazzullii*) might indeed constitute a complex (i.e., the *mazzullii* group) structured in three discrete taxa clearly defined by distribution, morphology and molecular evidence, corresponding to (i) the populations living in Monte Pellegrino (Palermo) and nearby mountains (taxon *mazzullii* s. str.) (Figs. 2 and 5); (ii) the endemic population of Cefalù, La Rocca (Palermo) (taxon *cephalaeditana*) (Figs. 3 and 6); and (iii) the populations living around Trapani (taxon *insolida*) (Figs. 4 and 7). These findings highlighted a controversy about the taxonomy of the *mazzullii* group with two conflicting hypotheses, the one-species hypothesis versus the three-(sub)species hypothesis.

In order to be able to test the two hypotheses we present a multidisciplinary perspective on distribution, ecology, shell and genital morphology, DNA and fossil record of these nominal taxa. In addition, we 1) better define the still poorly known distribution areas and the eco-biological features of the *mazzullii* group from Sicily; 2) provide detailed morphological descriptions of shells and genitalia; 3) perform a molecular analysis of this group which has never before been investigated from this point of view; and 4) unravel the phylogenetic relationships of its presumed taxa. To this end, sampling was carried out throughout northwestern Sicily, including many localities never reported before as collection sites for the *mazzullii* group. Our data were combined with paleontological records, bibliographic references and records from museum collections (Appendix 1). Observations on the animals' biology were made directly in the field. For each taxon, morphological features were illustrated in detail.

Two mitochondrial DNA markers (16S rDNA and 12S rDNA) which are very informative for high resolution analysis of evolutionary processes and the Internal Transcribed Spacer 2 (ITS-2), a relatively fast-evolving nuclear non-coding element which can be used in phylogenetic reconstructions on the species and genus level, were employed for molecular investigations. Time to the most recent common ancestor (TMRCA) for each lineage and its closest relative was calculated based on 16S rDNA phylogeny. The results and their taxonomic implications are discussed.

## Material and methods

Twenty-six specimens (Table 1) were analysed for both morphological and biometrical key features of shells and animals. Individuals were drowned in water and fixed in 75% ethanol. The reproductive apparatus was extracted by means of scalpel, scissors and forceps. Photographs were taken with a Panasonic Lumix FZ 30 digital camera. Maximum height and maximum diameter of the shell along with some parts of the genitalia were measured (in millimeters) with digital calipers. Illustrations of genitalia were sketched using a camera lucida mounted on a Leica MZ12.5 stereomicroscope, scanned and finished in GIMP (GNU Image Manipulation Program) 2.6. Voucher specimens for the pictures are deposited in the Museo Civico di Storia Naturale di Genova, Italy (catalogue numbers: MSNG55994–MSNG56005).

An additional thirty-one specimens of the *mazzullii* group and six (three per species) *Cornu aspersum* (O.F. Müller, 1774) and *Cantareus apertus* (Born, 1778), chosen as confamilial outgroups, were used for molecular analyses. Voucher specimens (catalogue numbers: CA1–CA3; CAP1–CAP3; CM1–CM5, CMS1–CMS5; COF1–COF4; COL1–COL3; CU4–CU5; MP1–MP5; PEC1–PEC3; SV1–SV4) were stored in the Laboratory of Cytogenetics and Molecular Biology, University of Urbino (Table 2).

Place names are given following the Portale Cartografico Nazionale (PCN, <http://www.pcn.minambiente.it/PCN/>). Each locality and/or collection site is named in the original language (Italian).

**DNA extraction, amplification and sequencing.** Samples were stored separately at -20°C in test tubes. Of each individual, a piece of foot tissue was used for total DNA extraction (by Wizard Genomic DNA Purification Kit, Promega). Fragments of 16S rDNA, 12S rDNA and ITS-2 sequences were amplified using three pairs of primers (Table 3) designed on alignments of several homologous sequences of Helicidae species downloaded from the GenBank database. PCR cycles were as follows: for 16S rDNA (209–223 bp) and ITS-2 (536–541 bp) amplicons, 95°C for 5 min; 95°C for 1 min, 55°C for 1 min, 72°C for 1 min (30 cycles); 72°C for 5 min. For 12S rDNA (192–196 bp) amplicons, 95°C for 5 min; 95°C for 1 min, 50°C for 1 min, 72°C for 1 min (35 cycles); 72°C for 5 min. To remove primers and unincorporated nucleotides, the amplified products were purified with the Wizard SV gel and PCR Clean-up kit (Promega).

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**TABLE 1.** *Ercetella* specimens used for morphological and anatomical analyses. Place-names and coordinates are reported (in Italian) following 1:25,000 scale topographic maps of the Portale Cartografico Nazionale (PCN, <http://www.pcn.mimambiente.it/PCN/>; amsl = above mean sea level).

Taxon	Locality	Collection date	Latitude	Longitude	Elevation	No. of specimens	Collectors
<i>E. mazzullii</i>	Palermo: La Montagnola	31.01.1981	38°11'43" N	13°16'19" E	100 m amsl	2	I. Sparacio
<i>E. mazzullii</i>	Palermo: Cinisi, Monte Pecoraro	16.04.1983	38°09'37" N	13°07'29" E	800 m amsl	4	I. Sparacio
<i>E. insolida</i>	Trapani: Custonaci, Monte Cofano	04.11.1985	38°06'00" N	12°39'55" E	100 m amsl	2	I. Sparacio
<i>E. mazzullii</i>	Palermo: Monte Pellegrino	14.10.1989	38°10'03" N	13°21'02" E	450 m amsl	4	I. Sparacio
<i>E. insolida</i>	Trapani: San Vito lo Capo	01.12.1992	38°10'50" N	12°43'33" E	60 m amsl	4	I. Sparacio
<i>E. mazzullii</i>	Palermo: Monte Pellegrino	21.04.2007	38°10'03" N	13°21'02" E	450 m amsl	2	I. Sparacio
<i>E. cephalaeeditana</i>	Palermo: Cefalù, La Rocca	12.05.2007	38°02'12" N	14°01'44" E	100 m amsl	3	I. Sparacio
<i>E. cephalaeeditana</i>	Palermo: Cefalù, La Rocca	16.01.2008	38°02'20" N	14°01'36" E	200 m amsl	1	S. Giglio
<i>E. mazzullii</i>	Palermo: Carini, Monte Columbrina	18.08.2008	38°09'30" N	13°13'45" E	100 m amsl	1	W. Renda, F. Liberto
<i>E. insolida</i>	Trapani: Custonaci, Monte Cofano	22.02.2009	38°06'11" N	12°40'40" E	250 m amsl	2	F. Liberto, S. Giglio
<i>E. cephalaeeditana</i>	Palermo: Cefalù, La Rocca	26.04.2009	38°02'20" N	14°01'37" E	200 m amsl	1	S. Giglio

**TABLE 2.** *Ercetella* specimens and outgroups (*Cornu aspersum* and *Cantareus apertus*) used for molecular analyses; amsl = above mean sea level.

Sample ID	Taxon	Locality	Collection date	Latitude	Longitude	Elevation	No. of specimens	Collectors
COL1-COL3	<i>E. mazzullii</i>	Palermo: Carini, Monte Columbrina	18.08.2008	38°09'30" N	13°13'45" E	100 m amsl	3	F. Liberto & W. Renda
CMS1-CMS5	<i>E. mazzullii</i>	Palermo: Sferracavallo	04.05.2008	38°11'43" N	13°16'19" E	100 m amsl	5	I. Sparacio & W. Renda
MP1-MP5	<i>E. mazzullii</i>	Palermo: Monte Pellegrino	14.01.2008	38°09'48" N	13°21'25" E	500 m amsl	5	I. Sparacio
PEC1-PEC3	<i>E. mazzullii</i>	Palermo: Cinisi, Monte Pecoraro	18.08.2008	38°09'28" N	13°07'42" E	900 m amsl	3	F. Liberto, W. Renda, I. Sparacio
CM1-CM3	<i>E. cephalaeeditana</i>	Palermo: Cefalù, La Rocca	16.01.2008	38°02'20" N	14°01'35" E	200 m amsl	3	S. Giglio
CM4-CM5	<i>E. cephalaeeditana</i>	Palermo: Cefalù, La Rocca	29.03.2008	38°02'24" N	14°01'38" E	60 m amsl	2	F. Liberto
COF1-COF4	<i>E. insolida</i>	Trapani: Custonaci, Monte Cofano	15.06.2008	38°06'18" N	12°40'31" N	500 m amsl	4	L. Barraco
SV1-SV4	<i>E. insolida</i>	Trapani: San Vito lo Capo, Cala Mancina	13.01.2008	38°10'43" N	12°43'07" E	50 m amsl	4	F. Liberto
CU4-CU5	<i>E. insolida</i>	Trapani: Custonaci, Monte Cofano	13.01.2008	38°06'27" N	12°40'59" E	70 m amsl	2	S. Giglio
CA1-CA3	<i>Cornu aspersum</i>	Palermo: Cefalù, Mazzaforno	18.01.2008	38°01'36" N	13°58'01" E	36 m amsl	3	S. Giglio
CAP1	<i>Cantareus apertus</i>	Palermo: Cefalù, Cocuzzola	17.05.2008	38°01'29" N	13°56'30" E	6 m amsl	1	F. Liberto
CAP2-CAP3	<i>Cantareus apertus</i>	Enna: Assoro, C.da Cernigliere	09.03.2008	37°37'59" N	14°24'27" E	600 m amsl	2	F. Liberto

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**TABLE 3.** Primer list.

Primer name	Sequence	Amplicon size (bp)
MS_16S_F	5'-GACTGTGCAAAGGTAGCATA-3'	211
MS_16S_R	5'-GCTGTTATCCCTAGAGTAAC-3'	
MS_12S_F	5'-AAATATTAGGGGAACCTAC-3'	189
MS_12S_R	5'-TATTTATTACTTTAAGTCC-3'	
ITS-2_F	5'-CATTGAACATCGACATCTTGA-3'	535
ITS-2_R	5'-CTCCGCTTAGTAATATGCTTAA-3'	

Sequencing of the purified PCR products was carried out using automated DNA sequencers at Eurofins MWG Operon (Germany). GenBank accession numbers (GQ402350–GQ402460) for all sequences generated in this study are listed in Table 4.

**Phylogenetic analyses.** Sequences were visualized with BioEdit Sequence Alignment Editor 7 (Hall 1999), aligned with the ClustalW option included in this software and double checked by eye. Standard measures of nucleotide polymorphism [haplotype/genotype diversity (Hd), mean pairwise differences (k), nucleotide diversity within groups ( $\pi = \Pi_i$  and  $\pi_{j_c}$  Pi corrected according to Jukes and Cantor) and nucleotide divergence ( $D_{xy}$ ) between groups] using the full set of all sequences were computed with DnaSP 4 (Rozas *et al.* 2003). Phylogenetic analyses were conducted in MEGA 4 (Tamura *et al.* 2007), BEAST 1.4.8 (Drummond & Rambaut 2007), and PhyML 3.0 (Guindon & Gascuel 2003; available at [http://www.phylogeny.fr/version2\\_cgi/one\\_task.cgi?task\\_type=phyml](http://www.phylogeny.fr/version2_cgi/one_task.cgi?task_type=phyml), see Dereeper *et al.* 2008). To address the phylogenetic relationships among taxa, many different analytical methods were used: Maximum Likelihood (ML), Neighbour Joining (NJ), Maximum Parsimony (MP) and Bayesian Inference (BI). For Maximum Likelihood analyses, best-fit models of molecular evolution were chosen using a hierarchical likelihood ratio test by Findmodel (available at <http://www.hiv.lanl.gov/cgi-bin/findmodel/findmodel.cgi>), a web implementation of Modeltest by Posada & Crandall (1998). Models selected were: GTR+ $\Gamma$  (16S rDNA and concatenation); HKY+ $\Gamma$  (12S rDNA and ITS-2). Neighbour Joining trees were constructed using TN93 (16S rDNA) and Tamura 3-parameter (12S rDNA and ITS-2) model distances; gaps were treated as missing data. MP trees were obtained using the Close-Neighbour-Interchange (CNI) algorithm with search level 3 in which the initial trees were achieved with the random addition of sequences (10 replicates). All positions containing gaps and missing data were eliminated from the dataset (complete deletion option). Characters were assigned equal weights. MP trees were collapsed to obtain a 50% majority rule consensus tree. Support for the internodes was assessed by bootstrap percentages (BP) (1,000 resampling steps for NJ and MP; 100 replicates for ML). For Bayesian analyses four Markov Chain Monte Carlo (MCMC) chains were run for 1,000,000 generations, sampling every 100 generations; from the 10,000 trees found the first 1,000 were discarded as “burn-in”. Finally, a 50% majority rule consensus tree was constructed. We performed NJ, MP, ML and BI analyses on each gene segment; in addition genes were concatenated and analysed by partitioned analyses which allowed each gene to have its own parameters. To test incongruence among genes a partition homogeneity test (Farris *et al.* 1994) was conducted in PAUP\*4.0b10 (Swofford 2002). The test (100 replicates of random addition heuristic search option with tree-bisection reconnection branch swapping) indicated significant heterogeneity among genes ( $p = 0.01$ ). However, since a growing number of studies (Yoder *et al.* 2001) indicate that incongruence tests are not reliable indicators of dataset combinability and no strong supported nodes were in conflict, genes were concatenated into a multigene dataset of about 950 bp. Analyses were conducted in BEAST v1.6.1 by \*BEAST (Heled & Drummond 2010) following the same steps described in the molecular dating section (see below). All phylogenetic trees were rooted using homologous nucleotide sequences of *Cornu aspersum* and *Cantareus apertus* (three specimens each).

**Molecular dating.** Molecular dating was conducted by a Bayesian MCMC approach in the program BEAST (v 1.4.8), where the topology and divergence times can be estimated simultaneously from the data and therefore a starting tree topology is not required, making it particularly appropriate for groups with uncertain phylogenies. BEAST input files were generated with BEAUTi (v 1.4.8). The 16S rDNA dataset was employed and a GTR+ $\Gamma$  was used to describe the substitution model, a Yule model was used to describe speciation. The likelihood ratio statistics didn't allow to reject the molecular clock hypothesis ( $p = 0.9987$ ) and, therefore, a strict molecular clock model was used. BEAST was run for 1,000,000 generations with samples taken every 100 generations. Five inde-

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pendent MCMC runs were conducted and the log and tree files were combined using LogCombiner (v 1.4.8). The results were examined with Tracer (v 1.5) to confirm stationary distribution and adequate effective sample sizes (i.e. ESS>200) that had been obtained for all parameters, indicating that the sampled generations were uncorrelated and the posterior distribution of the parameter was long and accurate. TreeAnnotator (v 1.4.8) was then used to summarize a best supported tree and annotate the tree with the mean age and posterior probabilities of the nodes under investigation. FigTree (v 1.3.1) was used to display the estimated tree with node ages and 95% confidence intervals. Programs BEAST, BEAUTi, LogCombiner, Tracer, TreeAnnotator and FigTree were downloaded from <http://beast.bio.edu.ac.uk>.

## Results

### Molecular data

**DNA sequences and variability.** The mitochondrial 16S rDNA sequence alignment showed nine different haplotypes, defined by 23 polymorphic (segregating) sites ( $S = 23$ ) including a total number of 26 mutations (Eta = 26). Haplotypes (h) revealed to be restricted to unique geographical populations, in particular, one was sampled only in the group from Cefalù, La Rocca; five from Monte Pellegrino and nearby mountains; and three from the sites around Trapani. Haplotype diversity (Hd) was  $0.892 \pm 0.002$ . Nucleotide diversity ( $\pi$ ) was  $0.043 \pm 0.004$  and 0.045 when corrected according to Jukes and Cantor ( $\pi_{JC}$ ). The average number of nucleotide differences (k) was 8.968.

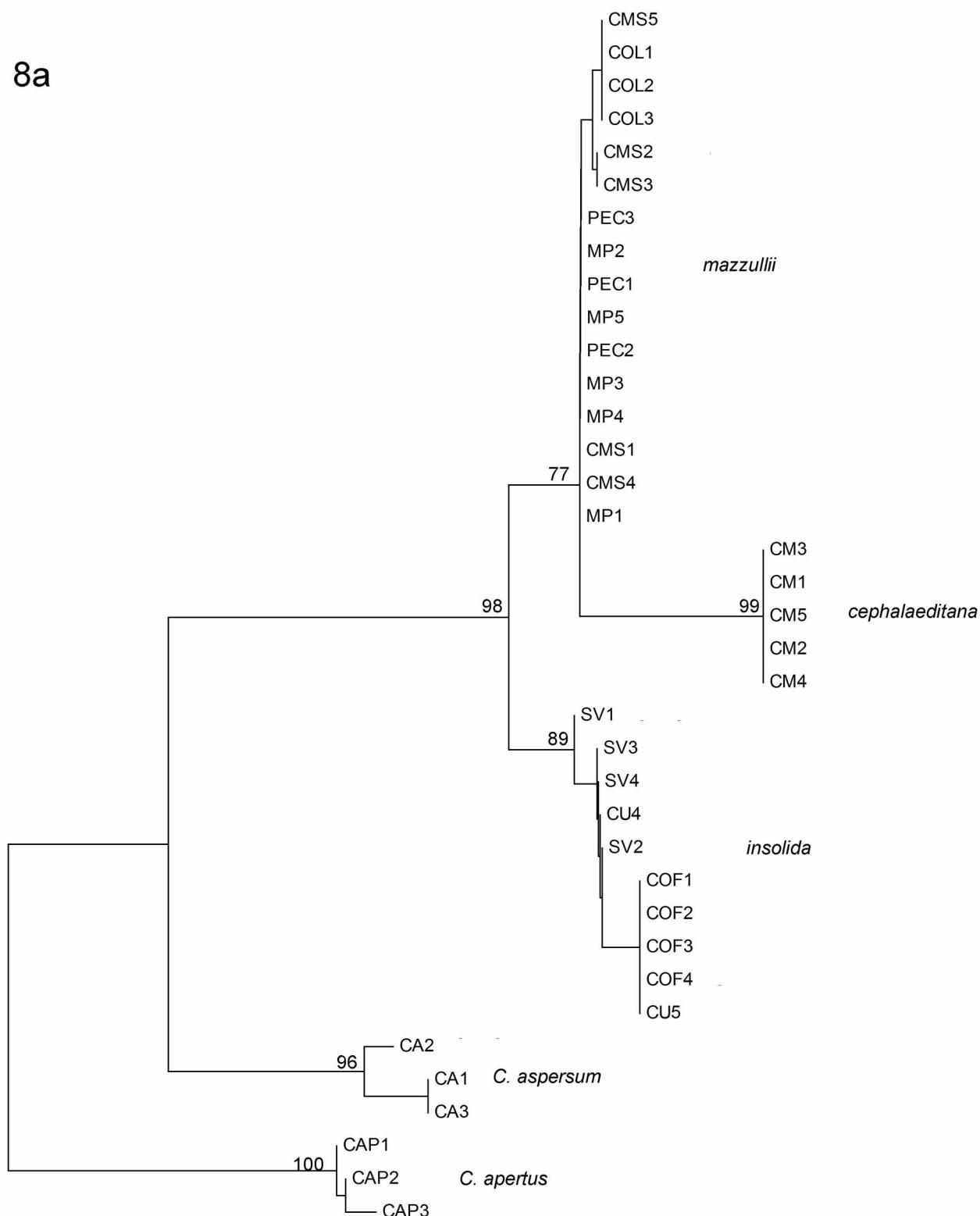
The mitochondrial 12S rDNA sequence alignment showed ten different haplotypes, defined by 37 polymorphic (segregating) sites ( $S = 37$ ) including a total number of 39 mutations (Eta = 39). In this case, haplotypes (h) also revealed to be restricted to unique geographical populations, in particular, two haplotypes were from Cefalù, La Rocca; five from Monte Pellegrino and nearby mountains; and three from the sites around Trapani. Haplotype diversity (Hd) was  $0.914 \pm 0.02$ . Nucleotide diversity ( $\pi$ ) was  $0.082 \pm 0.006$  and 0.091 when corrected according to Jukes and Cantor ( $\pi_{JC}$ ). The average number of nucleotide differences (k) was 15.591.

The ITS-2 sequence alignment showed six different genotypes, defined by 27 polymorphic (segregating) sites ( $S = 27$ ) including a total number of 29 mutations (Eta = 29). Genotypes revealed to be restricted to unique geographical populations, in particular, three were sampled only in the group from Monte Pellegrino and nearby mountains; two from the sites around Trapani; and one from Cefalù. Genotype diversity was  $0.845 \pm 0.026$ . Nucleotide diversity ( $\pi$ ) was  $0.018 \pm 0.002$  and 0.019 when corrected according to Jukes and Cantor ( $\pi_{JC}$ ). The average number of nucleotide differences (k) was 9.856.

**Phylogenetic structure and species delineation.** All (NJ, MP, ML and BI) phylogenetic analyses yielded highly congruent tree topologies that agreed on branching patterns and sister-taxon relationships. Analysis of 16S rDNA, 12S rDNA and multigene combined datasets showed three phylogenetic groups with strong branch support, thus confirming the occurrence of three taxa. The first phylogroup (*mazzullii*) included only the specimens collected on Monte Pellegrino and nearby mountains, the second phylogroup (*cephalaeditana*) the specimens from Cefalù, La Rocca and the third phylogroup (*insolida*) the specimens from Trapani province; *mazzullii* and *cephalaeditana* showed sister-taxon relations. The analysis of the ITS-2 dataset showed phylogenetic trees differing in a few topological details; the BI consensus tree evidenced the occurrence of the three taxa but with a different sister-group relationship; whereas NJ, ML and MP trees didn't distinguish among the three taxa placing all specimens of the *mazzullii* group within the same cluster supported by a strong BP value (86%). Since all reconstructions produced almost identical results not changing any of the interpretations, only the phylogenetic ML and BI consensus trees obtained by the concatenate analysis are displayed as an example in figures 8a, b.

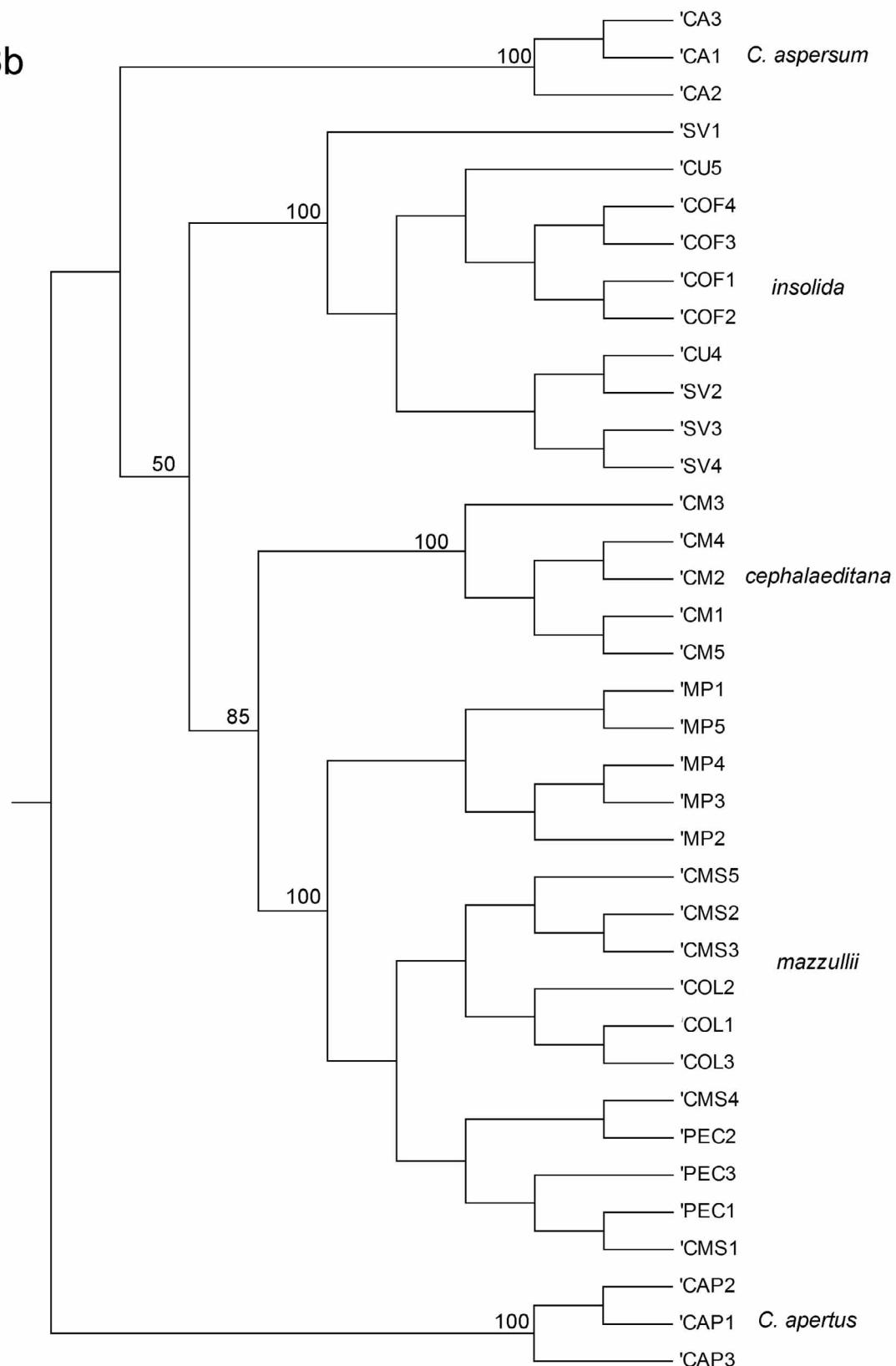
For mtDNA sequences the maximum divergence within each taxon was 1.3% (16S rDNA) and 1.6% (12S rDNA); divergence among groups ranged from 6% to 10% (16S rDNA) and from 7% to 17% (12S rDNA). As far as ITS-2 sequences are concerned, when comparing the taxa two at a time, genetic differences ranged from 0.5% to 2.4% in the *mazzullii* group. Estimates of evolutionary rates of mtDNA and nDNA fragments were 0.48% (both 16S rDNA and 12S rDNA) and 0.50% (ITS-2) per million years.

8a



**FIGURE 8a.** Maximum Likelihood consensus tree inferred from a combined dataset including partial sequences of the mitochondrial 16S rRNA and 12S rRNA and the nuclear ITS-2 genes. Numbers above branches represent bootstrap values.

8b



**FIGURE 8b.** 50% majority rule Bayesian tree inferred from a combined dataset including partial sequences of the mitochondrial 16S rRNA and 12S rRNA and the nuclear ITS-2 genes. Numbers above branches represent Bayesian posterior probabilities.

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**TABLE 4.** Locality codes, place names and GenBank accession numbers of *Ercitella* specimens and outgroups (*Cornu aspersum* and *Cantareus apertus*) used for molecular analyses. N = number of specimens.

Sample ID	taxon	Localities	GenBank 12S rRNA	GenBank 16S rRNA	GenBank ITS-2	N
CA1-CA3	<i>Cornu aspersum</i>	Palermo: Cefalù, Mazzaforno	GQ402350-GQ402352	GQ402387-GQ402389	GQ402424-GQ402426	3
CAP1	<i>Cantareus apertus</i>	Palermo: Cefalù, Coeuzza	GQ402353	GQ402390	GQ402427	1
CAP2-CAP3	<i>Cantareus apertus</i>	Enna: Assoro, C.da Cerniglere	GQ402354-GQ402355	GQ402391-GQ402392	GQ402428-GQ402429	2
CM1-CM5	<i>E. cephalaeeditana</i>	Palermo: Cefalù, La Rocca	GQ402356-GQ402360	GQ402393-GQ402397	GQ402430-GQ402434	5
CMS1-CMS5	<i>E. mazzullii</i>	Palermo: Sferracavallo	GQ402361-GQ402365	GQ402398-GQ402402	GQ402435-GQ402439	5
COF1-COF4	<i>E. insolida</i>	Trapani: Custonaci, Monte Cofano	GQ402366-GQ402369	GQ402403-GQ402406	GQ402440-GQ402443	4
COL1-COL3	<i>E. mazzullii</i>	Palermo: Carini, Monte Columbrina	GQ402370-GQ402372	GQ402407-GQ402409	GQ402444-GQ402446	3
CU4-CU5	<i>E. insolida</i>	Trapani: Custonaci, Monte Cofano	GQ402373-GQ402374	GQ402410-GQ402411	GQ402447-GQ402448	2
MP1-MP5	<i>E. mazzullii</i>	Palermo: Monte Pellegrino	GQ402375-GQ402379	GQ402412-GQ402416	GQ402449-GQ402453	5
PEC1-PEC3	<i>E. mazzullii</i>	Palermo: Cinisi, Monte Pecoraro	GQ402380-GQ402382	GQ402417-GQ402419	GQ402454-GQ402456	3
SV1-SV4	<i>E. insolida</i>	Trapani: San Vito lo Capo, Cala Mancina	GQ402383-GQ402386	GQ402420-GQ402423	GQ402457-GQ402460	4

**TABLE 5.** Sites showing records of the ancient presence of *Ercitella* specimens in Sicily; N.A. = Not Applicable.

Locality	Site	Coordinates	Record	Reference
Militello Rosmarino (Messina)	M. Scurzi	38°03'42" N, 14°39'50" E	Holes in mesozoic limestone	Personal data
Torrenova (Messina)	Contr. a Bicurca	38°04'02" N, 14°40'16" E	Holes in mesozoic limestone	Personal data
Madonie mountains (Palermo), Gratteri	P.zo Giurafelle	37°58'19" N, 14°00'11" E	Holes in mesozoic limestone	Personal data
Madonie mountains (Palermo), Gratteri	C.zo Giampietra	37°58'28" N, 13°59'00" E	Holes in mesozoic limestone	Personal data
Madonie mountains (Palermo), Isnello	C.zo Balatelli	37°55'44" N, 14°00'47" E	Holes in mesozoic limestone	Personal data
Madonie mountains (Palermo), Isnello	Contr. a Aquileia, Stallone	37°56'41" N, 14°02'42" E	Holes in mesozoic limestone	Personal data
Madonie (Palermo), Collesano	R.ca di Uoni	37°57'14" N, 13°53'59" E	Holes in mesozoic limestone	Personal data
Termini Imerese (Palermo)	C. Rainondo	37°58'36" N, 13°38'23" E	Holes in mesozoic limestone	Personal data
Ventimiglia di Sicilia (Palermo)	C.zo Acquapietra	37°55'22" N, 13°34'44" E	Holes in mesozoic limestone	Personal data
Peloritani mountains (Messina): Santa Teresa di Riva	Grotte	N.A.	Fossil	Seguenza, 1902
Favignana island (Trapani)	N.A.	N.A.	Fossil	Fiorentino et al., 2004; Personal data
Levanzo island (Trapani)	N.A.	N.A.	Fossil	De Gregorio, 1894.
Monte S. Calogero, near Termini Imerese (Palermo)	N.A.	N.A..	Shells	Calcara, 1842
Barcellona Pozzo di Gotto (Messina)	N.A.	N.A.	Shells	Benoit, 1857; 1875; 1882

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### Morphological data

Detailed conchological and anatomical descriptions of the individual taxa of the *mazzullii* complex are given in the systematic part. Their shells are illustrated in Figs. 9–20, their genital systems in Figs. 21–23.

The morphological data showed that within the *mazzullii* group the three taxa are different in many features including (i) shell size (decreasing from *insolida* to *mazzullii* to *cephalaeditana*) and, to some extent, shape and sculpture (*insolida* is more globose and smooth, *mazzullii* is regularly wrinkled, whereas *cephalaeditana* is, especially on the last whorls, strongly wrinkled and irregularly reticulated); (ii) diverticulum of bursa copulatrix (BCD) and duct of bursa copulatrix (DBC) length ratio, which is 1:1 for *insolida*, nearly 2:1 for *mazzullii* and 1.5–1.2:1 for *cephalaeditana*; (iii) number of digit-like appendices of the digitiform glands (decreasing from *insolida* to *mazzullii* to *cephalaeditana*); and (iv) number of vaginal longitudinal pleats which varies from 7–8 (*insolida*) to 5–6 (*mazzullii*) to 4–5 (*cephalaeditana*).

Genitalia of *Cornu aspersum*, *Cantareus apertus* and *Eobania vermiculata* (O.F. Müller, 1774), used as external comparisons, are shown in figures 24–26; shells of the three species are illustrated in figures 27–29 (for detailed descriptions, see Giusti *et al.* 1995).

When compared to *Cornu aspersum*, *Cantareus apertus* and *Eobania vermiculata*, genitalia of the three taxa of the *mazzullii* complex notably differ in many morphological characters, although the general architecture of the genitalia is similar (see Giusti *et al.* 1995).

### Geographic distribution

The current confirmed distribution of the *H. mazzullii* complex is shown in Fig. 1. Records from museum and private collections are listed in Appendix 1.

The taxon *mazzullii* inhabits the calcareous mountains around Palermo including Monte Pellegrino, Monte Gallo and the entire mountain chain which proceeds from Monte Cuccio north-west towards Sferracavallo, Capaci, Monte Columbrina, Monte Pecoraro (Carini) up to Monte Palmeto (Terrasini). There is no evidence of these animals at Monte Catalfano and Capo Zafferano which represent the eastern limit of the mountains of Palermo (Lo Brano & Sparacio 2006).

The taxon *cephalaeditana* is endemic of Cefalù, La Rocca, with several small populations occurring either at its base or peak, whereas the record of Rocca di San Nicola (previously known as Feudo Costa) reported by Pirajno (1840) was not confirmed during our field explorations. Finally, records from Barcellona Pozzo di Gotto (Messina province, northeastern Sicily) (Benoit 1857, 1875, 1882) which haven't been verified yet, most likely refer to a taxon related to *cephalaeditana* (see Benoit 1882).

The taxon *insolida* is distributed from the calcareous mountains around Trapani, Monte San Giuliano and Monte Cofano to San Vito lo Capo and the surroundings of Scopello.

In our opinion, the former distribution area of *H. mazzullii* s. l. (= *mazzullii* group) was much wider than the present one, currently limited to northwestern Sicily. This is documented by a high number of collection sites, fossils or peculiar tunnels (holes in mesozoic limestone made by these saxicavous helicids) reported for numerous places of the island, including a few northeastern ones where at present there is no evidence of extant populations of the *mazzullii* complex (Table 5).

### Biology

The taxa within the *H. mazzullii* complex are rupicolous and strictly bound to calcareous cliffs from which they descend only on moist and rainy days to reach the soil. Generally, they move forward vertically on the cliff and stay within peculiar groupings of tunnels (Fig. 30) dug into the limestone (Gaudin 1860; Brehm 1869; Platania 1890; Meunier 1890; De Gregorio 1890, 1916a; Lamy 1930; Kuehnelt 1932; Di Salvo 1932; Rensch 1932a, 1932b, 1937; Manganelli *et al.* 2000).

Bibliographic references, integrated with personal observations, allow us to provide a detailed description of the tunnels. They are mostly cylindrical with a diameter of 3–4 cm and a variable depth ranging from about 8–40 cm. Tunnels are horizontal (perpendicular to the vertical wall) at first, but after a few centimetres the direction turns

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upwards. Tunnels dug on the top of pre-existing holes show a trend which is, from the very beginning, upwards. Tunnels may be distant from or, more frequently, next to each other. In the last case, their progressive enlargement may cause the dissolving or even collapse of dividing walls, resulting in broad cavities the outside margin of which is circular or ellipsoidal (with a diameter from 10 to 30 cm, up to a maximum of more than 60 cm) and whose interior walls are pitted due to the remnants of each tunnel (Fig. 30b).

The snails' rock-boring ability results from the secretion of a  $\text{H}_2\text{CO}_3$ -rich mucus used to dissolve the calcium carbonate of the limestone; residues are then removed by the radula and, according to Sacchi (1955a), also by the rough shell surface. Mucus evaporation may cause re-deposition of calcium carbonate, giving origin to peculiar botroidal concretions close to the tunnel opening (Fig. 30d).

Specimens also show homing behaviour. At night or on moist and rainy days the snails leave the tunnels and crawl on calcareous cliffs or soil moving for several metres, while they return to the place they came from when it gets drier. Sometimes they also aggregate in fissures in the rock. In field experiments in which adult animals were marked and tracked in their natural environment, it was documented that these helicids rest in, leave and go back to the same group of tunnels over extended periods of time, even more than one year long (personal unpublished data).

As for these snails' diet, Varga (1989), analyzing the contents of the digestive tract and fecal pellets of *Helix mazzullii* specimens, found still recognizable plant fragments. In personal observations, we witnessed several times specimens of the *H. mazzullii* complex feeding on living or dried parts of different plants occurring on the rock face or at the rock base.

The taxa of the *H. mazzullii* group are strictly rupicolous (= inhabiting rocks) and possess saxicavous (= rock-boring) abilities and thus occupy an ecological niche different from that of the closely related genera *Cornu* and *Cantareus* whose very common and widespread species are strictly linked to the soil.

## Paleontology

Fossils attributable to *H. mazzullii* (*s. str.*) were recovered at the following sites:

- near Palermo (Philippi 1836; 1844)
- Billiemi (Palermo), in bony breccias (Calcara 1845a; 1845b: as *Helix retirugis*)
- Monte Pellegrino (Fig. 31) and Ficarazzi (Palermo province), in post-Pliocene fossiliferous deposits (Monte-rosato 1872; 1877; 1891)
- Monte Pellegrino, in quaternary deposits encrusting secondary rocks, including fossil deposits of rodents and carnivores from the mountain peak (of continental origin) and the post-Pliocene detrital limestone (calcareous arenite of marine origin) (De Gregorio 1886; 1895; 1916a; 1916b; 1927)
- Castellana (Boccadifalco, Palermo) (De Gregorio, 1886)
- near Palermo, in quaternary deposits encrusting secondary rocks of the mountains (quite frequent) (De Gregorio 1927)
- Pietrazzi county (Palermo), in a post-Pliocene deposit (doubtful records) beneath a marine shelly limestone layer (De Gregorio 1927)
- Bisaquino (Palermo), in quaternary rocks (one-specimen) (De Gregorio 1927)
- caves near Palermo (grotta dell'Addaura, grotta Perciata and grotta delle Vitelle), in prehistoric deposits (very frequent), or in the surroundings of caves [grotta della Conza (Sferracavallo) and grotta del Maccagnone (Carlini)] (Anca 1860a; 1860b; 1867; Falconer 1868; Tricomi 1926; 1929; De Stefani 1941)
- Luparello cave (Baida, Palermo) (De Gregorio 1895; 1899: as *Helix Luparellensis*) (Fig. 32). Although the fossil assemblages of the Luparello cave underwent considerable reworking over a long time (Burgio & Costanza 1999), *H. luparellensis* is, beyond any reasonable doubts, associated with the vertebrate fauna including *Elephas falconeri* Busk (fossil dwarf elephant) (De Gregorio 1895; 1899; 1927). This fauna, dating back to the Middle Pleistocene, around 550,000 years ago (Kotsakis 1979; Belluomini & Bada 1985; Bada *et al.* 1991; Bonfiglio & Burgio 1992; Burgio 1998), is the richest one in endemic species of the quaternary period in Sicily
- Alcamo, *Elephas falconeri* complex in continental sandy limestone (locally called "travertino") (Burgio & Cani 1988; Abate *et al.* 2006) (Fig. 33)

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- Poggio Schinaldo (Cinisi, Palermo), from the *Elephas falconeri* complex in cave deposit (Esu *et al.* 1986).

The shell of the sample from Alcamo is globose, weakly cancellated and rugose, and hence completely different from *H. luparellensis*. This evidence suggests that during the Middle Pleistocene at least three different shell morphotypes (*H. mazzullii* from Monte Pellegrino, *H. cf. mazzullii* from Alcamo and Poggio Schinaldo, *H. luparellensis* from Baida) belonging to the *H. mazzullii* group occurred in the Palermo area.

Fossil shells of *H. cephalaeeditana* have been observed in the talus slope of La Rocca of Cefalù (personal data). Notably, these specimens are much bigger (height 40 mm; maximum diameter 38 mm) than shells of modern specimens (Fig. 34).

Several paleontological reports of fossils of *H. mazzullii* attributable to the taxon *insolida* include:

- Grotta di Caciucavaddu (Levanzo, Egadi Islands, Trapani), quaternary deposits (De Gregorio 1927; Fiorentino *et al.* 2004)
- Monte San Giuliano (Trapani), quaternary rock (De Gregorio 1927)
- coastal zone of Cala Mancina (south-west of San Vito lo Capo, Trapani), near Grotta dei Cavalli, breccias (personal data) (Fig. 35)
- Favignana (Egadi Islands, Trapani) (Fig. 36), Lower Pleistocene, (personal data)
- Grotta dell'Uzzo, meso-neolithic (10–8.3 thousand years ago) (Compagnoni 1993; Piperno 1997).

## Discussion

This study represents the first comprehensive analysis of the *H. mazzullii* group from Sicily, one of the most interesting, yet least well-known Helicidae taxa. Data discussed herein confirm the existence of a certain degree of differentiation within the group, as already suggested by many authors on the grounds of shell morphology (Figs. 37–41) and genital features (Pirajno 1840; Benoit 1857; Monterosato 1892; 1894; Pilsbry 1895; Hesse 1919; Cockerell 1921; Giannuzzi-Savelli *et al.* 1986; Falconieri 1995; Colombara *et al.* 2008; Ryolo & Palazzi 2009; Liberto *et al.* 2010).

The presence of fossil shells of *mazzullii* s.l. specimens is well documented in many continental deposits of Sicily, which suggests that the group might have occurred at least in the late Pliocene–early Pleistocene (Monterosato 1872, 1877, 1891; De Gregorio 1886, 1895, 1899, 1916a, 1916b, 1927; Kotsakis 1979; Belluomini & Bada 1985; Esu *et al.* 1986; Burgio & Cani 1988; Bada *et al.* 1991; Bonfiglio & Burgio 1992; Burgio 1998; Fiorentino *et al.* 2004; Abate *et al.* 2006; personal data). Nevertheless, although these records indicate the terminus after which the *mazzullii* group certainly occurred, we cannot rule out the possibility of an earlier appearance. In fact, our BEAST analyses suggest that the origin of the *mazzullii* group can be dated about  $10.7 \pm 0.07$  million years ago and that the split of the sister taxa *mazzullii* and *cephalaeditana* likely occurred about  $8.98 \pm 0.07$  million years ago. Moreover, the BEAST and MEGA time estimates are largely congruent. With MEGA, node ages were dated to about 8.82 (origin of the *mazzullii* group) and 7.9 (divergence between *mazzullii* and *cephalaeditana*) million years ago. Hence our results indicate that both dichotomic events might have taken place long before the Messinian salinity crisis, suggesting a longer evolutionary history for the *mazzullii* group than expected. This agrees with the occurrence of a peculiar *H. mazzullii*-associated vertebrate fauna known as *Pellegrinia* fauna (after *Pellegrinia panormensis*, an endemic genus and species of the African family Ctenodactylidae [Mammalia, Rodentia] [De Gregorio, 1886] which may be a survivor of Oligocene–early Miocene faunas [Azzaroli & Guascone 1979; Azzaroli 1990]), in one of the oldest continental fossil deposits of Sicily (Monte Pellegrino).

Considering the specimen sampling which covered representative sites throughout the distribution area of the group and the robust phylogenetic reconstruction based on nucleotide patterns of two mitochondrial (16S and 12S rDNA) and one nuclear (ITS-2) gene fragments, we propose the following system: the *mazzullii* group consists of three species, *insolida*, *mazzullii* and *cephalaeditana*, with a sister relationship for the latter two.

Remarkably, intraspecific mtDNA sequence divergences observed within the *mazzullii* group (1.3% for 16S rDNA; 1.6% for 12S rDNA) are much lower than those reported for other land snails (up to 12.9%, see, e.g., Thomaz *et al.* 1996; Hayashi & Chiba 2000; Watanabe & Chiba 2001), and the mutation rate (0.48% per million years for both 16S rDNA and 12S rDNA) is not high. This might indicate that an accelerated mtDNA evolution

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found in some taxa of Mollusca (Pinceel *et al.* 2005 and references therein) but not in others (Murray *et al.* 1991; Douris *et al.* 1998; Guiller *et al.* 2001; Pfenninger & Posada 2002), does not occur in this case. Therefore, in our opinion, the mtDNA genetic differences assessed between groups (6–10% for 16S rDNA; 7–17% for 12S rDNA) can be considered a good value for species delimitation in Stylommatophora, suggesting strong isolation and strong divergence, i.e., the very low dispersal ability of these animals is likely to have drastically reduced gene flow. This justifies to consider the taxa under investigation as different phylogenetic species. Moreover, taking into account that in molluscs for the ITS region interspecific divergences rise up to 3.4% and divergences between different genera are up to 10% (e.g., Wade *et al.* 2001; Van Riel *et al.* 2005), ITS-2 distances between the *mazzullii* group and *C. aspersum* and *C. apertus* (7.6% and 6%, respectively) seem not only to substantiate our hypothesis that the *mazzullii* group comprises three species but also to strongly support the concept that these species belong to a distinct genus.

The morphological data demonstrate that the three taxa within the *mazzullii* group differ in many features including shell size, shape and sculpture, the ratio of the bursa copulatrix diverticulum (BCD) and duct (DBC) lengths, the number of digit-like appendices of the digitiform glands and the number of vaginal longitudinal pleats.

When compared to *Cornu aspersum*, *Cantareus apertus* and *Eobania vermiculata*, the genitalia of the three species of the *mazzullii* complex differ significantly in many morphological characters, although the general genitalic architecture is similar (see Giusti *et al.* 1995). Thus, genital morphology likewise indicates that the *mazzullii* complex may deserve to be treated as a separate genus.

To further investigate this point, twenty species, representing sixteen currently recognized Helicidae genera, together with the thirty-seven specimens analysed in this study, were used to construct a Maximum Likelihood molecular phylogeny. Partial DNA sequences of the mitochondrial 16S rDNA for the twenty species were retrieved from GenBank. Most genera are represented by a single species. *Helicella itala* (Hygromiidae) was used as an out-group. As shown in Figure 42, the ML consensus tree clearly indicates that (i) all genera are well separated from one another; (ii) 16S rDNA sequences of *C. aspersum* and *C. apertus* reported in the present paper are absolutely in line with those deposited in GenBank by other authors (i.e., all specimens of a given genus are in the same clade); (iii) the *mazzullii* complex, *Cantareus* and *Cornu* form a monophyletic group which is distinct from other related genera (e.g., *Eobania*, *Helix*); and (iv) the *mazzullii* group is supported by a high BP value (97%). The results of this “meta-analysis”, combined with the fact that the exclusive ecological features and very limited distribution area of *mazzullii* group make it very peculiar among Mediterranean Helicidae, once again support the idea that the complex might be attributed to a distinct genus.

More than a century ago, Monterosato (1894) introduced *Erctella* (from Ercta, the ancient name of Monte Pellegrino, near Palermo) as a new section in the subgenus *Helix* (*Pomatia*). He included two species group taxa of the *mazzullii* group, *H. mazzullii* and *H. costae*, in the new section. Monterosato didn't publish any other papers on these land snails, but in one of his manuscripts he wrote: "...Tutte queste quattro principali forme: *luparellensis* (estinta); *retirugis*, *mazzulli*, *insolida* (attuali); sono appartenenti al sottogenere *Erctella*..." ("...All four main types: *luparellensis* [extinct]; *retirugis*, *mazzulli*, *insolida* [extant]; belong to the subgenus *Erctella*..." [translation by the authors]) (see [http://www.sim-online.it/area.php?idarea=26, under "Manoscritti", file name: "su alcune helix/gs003-4 su alcune helix.jpg"](http://www.sim-online.it/area.php?idarea=26, under 'Manoscritti', file name: 'su alcune helix/gs003-4 su alcune helix.jpg')) . In an autographic note (Fig. 43) (Gigli collection, Natural History Museum of Milan) commenting on several *mazzullii* specimens caught on Monte Pellegrino in 1912, Monterosato says: "...It belongs to the genus *Erctella* Monts., introduced by me and adopted by Pilsbry. Because of its anatomical characters it firmly differs from *Cryptomphalus* (*aspersa*)".

The differences among *H. mazzullii* s.l., *Cantareus apertus* and *Cornu aspersum* were confirmed by Varga's (1989) detailed work on the digestive and other organ systems of these taxa (all included in the genus *Helix* by Varga) from Sicily. In addition, Varga (1989) recognized a distinct species within the *mazzullii* complex (as *Helix* sp.), collected between Palermo and Trapani.

The name *Erctella* has been mostly ignored or considered as synonym of related genera (Alzona 1971; Bank *et al.* 2001; Bank 2009) despite the rather high number of species included in this supraspecific taxon by Pilsbry (1895). Recently, Liberto *et al.* (2010) proposed to re-introduce the genus *Erctella* for this group of endemic Sicilian taxa with their peculiar rupicolous and saxicavous biology. Results discussed in this paper not only support the suggestion to re-introduce the genus *Erctella* including *E. mazzullii*, *E. cephalaeeditana* and *E. insolida*, but also complement and support Monterosato's opinion (1894; Ryolo & Palazzi 2009) that *Erctella* shows a high variability and is to be kept quite distinct from the closely related genera *Cantareus* and *Cornu*.

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Remarkably, Sacchi (1955a, b) arrived to a similar conclusion from analyses of ecological and biogeographical features. In fact, based on rock boring abilities and morphological similarities between Sicilian *Erctella* specimens and the also rupicolous and saxicavous North African *Helix subaperta* Ancey, 1893, he suggested that the two groups might have split during the Messinian salinity crisis. In our opinion, even if well-documented land connections surely occurred, the climatic and ecological conditions during that time made these helicids with very low active dispersal abilities unlikely candidates for the suggested land-based dispersal process. Hence, we hypothesize that the separation process between the Sicilian *Erctella* and similar North African helicids might date back much further than previously reported, maybe to the Oligocene–Miocene, when the split, dislocation and connection of microplates of the old Tyrrhenian plate resulted in allopatric speciation events and distribution of vicariant taxa within the southwestern Mediterranean basin (Giusti & Manganelli 1984; La Greca 1984, 1990a, 1990b; Gueguen *et al.* 1998; Rosenbaum *et al.* 2002; Pfenninger *et al.* 2010). Nevertheless, further studies of the phylogenetic relationships between Sicilian and Maghrebinian helicids are certainly needed to address this question, and, maybe, provide additional support to our speculative hypothesis.

Finally, we express our hope that all findings presented and discussed herein may provide a useful contribution to the knowledge of these peculiar and extremely vulnerable Sicilian species that will help to organize and manage adequate conservation measures and habitat protection programs.

## Systematics

### Helicidae

#### *Erctella* Monterosato, 1894

Type species: *Helix mazzullii* De Cristofori & Jan, 1832, by subsequent designation (Pilsbry 1895: 316).

**Shell** (Figs. 9–20). Dextral, medium-sized for a helicid (height 24–40 mm, maximum diameter 21–35 mm), globose-conical, light, uniformly yellowish or brownish, otherwise greenish for the presence of thin periostracum; sometimes with 3–5 brown spiral bands; external surface with thin growth lines, sometimes particularly wrinkled and with reticulated appearance; spire more or less elevated with 4–5 convex whorls, last whorl very large; aperture large, oval to round; sutures deep; umbilicus closed; peristome sharp, sometimes more or less thickened and/or reflected.

**Animal.** Yellow, sometimes with mantle border and foot lighter and head slightly darker, or entirely darker (Benoit 1857; Monterosato 1892).

**Genitalia** (Figs. 21–23). General scheme of the semidiaulic monotrematic type (incomplete triaulic monotrematic: Visser 1977; Giusti *et al.* 1995), characterised by diverticulum of bursa copulatrix as long as duct of bursa copulatrix, or longer; two digitiform glands, each of which is distally divided into 11–45 slender and branched digit-like appendages; vagina showing internally smooth and raised opening of free-oviduct, 4–8 small, raised, longitudinal pleats that disappear near dart-sac opening; a V-shaped pleat around dart-sac opening which continues with a little pilaster-shaped pleat ending in the genital atrium; penis of medium size, roundish; penial flagellum as long as penis and epiphallus together, or shorter; distal portion of penis internally divided into proximal and distal cavities by annular pad; proximal cavity with 14–20 longitudinal pleats, a small papilla on the internal wall and a very small penial papilla inside opening of proximal penis into distal penis; distal cavity smooth with a raised crest-like structure situated level with its opening into genital atrium. Inside the genital atrium is a characteristic bulge which is close to but separate from both the crest-like structure of the distal penis and the small pilaster-shaped pleat of the vagina.

**Remarks.** For differences characterizing *Erctella* with respect to the most closely related genera see Table 6 and the following dichotomous key to the genera *Cantareus*, *Eobania*, *Cornu* and *Erctella*.

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**TABLE 6.** Anatomical comparison of *Cantareus*, *Eobania*, *Cornu* and *Ercetella*.

Character	<i>Cantareus</i>	<i>Eobania</i>	<i>Cornu</i>	<i>Ercetella</i>
diverticulum of bursa copulatrix	variable in length	much longer than duct of bursa copulatrix	similar in length to duct of bursa copulatrix	variable in length
digitiform glands	short base, distal portion with many branched digit-like appendages	long base, distal portion bifurcate into two then three or more branches, each with tufts of branched digit-like appendices	short base, distal portion with many branched digit-like appendages	short base, distal portion with many branched digit-like appendages
penial flagellum	as long as penis and epiphallus together, or little shorter	as long as penis and epiphallus together, or little longer	much longer, 3-5 times as long as epiphallus	as long as penis and epiphallus together, or shorter
penis	oblong	oblong	oblong	roundish
penial papilla	rosette-like system of pleats	very small, rosette-like	small, rosette-like	very small, rosette-like
false penial papilla	cylindroconical, ending level with annular pad	long, slender, surpassing the annular pad	small, not reaching the annular pad	small, not reaching the annular pad
crest-like structure	irregularly shaped, situated level with its opening into genital atrium	large, located inside the genital atrium embracing the opening of penis	less irregularly shaped, situated level with its opening into genital atrium, extended to fuse with a knob-like structure which arises at the end of a pilaster-shaped pleat on vagina wall	small, situated level with its opening into genital atrium, near a robust and characteristic bulge of the inner wall of the atrium; bulge separated from the little pilaster-shaped pleat of distal vagina

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1. Shell naticoid, uniformly yellowish, greenish or brownish, completely without bands. Proximal cavity of distal penis containing a rosette-like system of pleats which imitates or forms very small, true penial papilla. Thermophilic and sciaphilous genus more common on argillaceous and marly soils, that aestivates by hiding deep in the soil and closing its shell aperture with a robust epiphramg ..... *Cantareus*
- Shell not naticoid. Proximal cavity of distal penis containing very short, rosette-like penial papilla ..... 2
2. Shell globose to sub-globose-conical, somewhat depressed, creamy-white, usually with marked brown bands or single, diluted, flecked, pale-brown and covering upper surface of whorls, peristome markedly reflected. Proximal cavity of distal penis containing also a long and slender false penial papilla that surpasses the annular pad. Thermophilic genus with wide ecological valence, living in many different kinds of biotopes that aestivates by attaching itself by its paper-like epiphram to rocks, walls, under surface of stones and other similar substrata ..... *Eobania*
- Shell globose-conical, uniformly greenish-yellow or with reddish-brown spiral bands. Proximal cavity of distal penis containing smaller false penial papilla not reaching the annular pad ..... 3
3. Penis oblong; penial flagellum 3–5 times as long as epiphallus; crest-like structure in distal cavity of penis extended to fuse with a knob-like structure which arises at the end of a pilaster-shaped pleat on vagina wall; genus with wide ecological valence (occurring in many different kinds of biotopes) ..... *Cornu*
- Penis roundish; penial flagellum as long as penis and epiphallus together, or shorter; distal cavity of penis with a little crest-like structure separated by the little found pilaster-shaped pleat on vagina wall; just before the genital atrium there is also a robust and characteristic bulge of the internal wall of the distal cavity. Genus with exclusively rupicolous species ... *Ercetella*

***Ercetella mazzullii* (De Cristofori & Jan, 1832)**

Figs. 9–14, 21

*Helix crispata* Costa 1829: 106, 111–112 (Sicily and near Naples); non *Helix crispata* Féruccac 1821: 29. — Costa 1836: 16 (Sicily); Scacchi 1836: 17.

*Helicogena mazzulli* Jan 1830: 3; *nomen nudum*.

*Helix retirugis* Menke 1830: 14; *nomen nudum*.

*Helix mazzullii* De Cristofori & Jan 1832: 1 (Sicily); *nomen nudum*.

*Helix mazzullii* De Cristofori & Jan 1832: 1. — Philippi 1836: 126, pl. 8 fig. 3 (“montibus circa Panormum”), 135 (fossil: “in limestone from Palermo”); Aradas & Maggiore 1841: 59–60 (Palermo; plaia of Catania); Calcara 1842: 23 (Termini Imerese: San Calogero, on the hills); Philippi 1844: 103 (Palermo), 112 (fossil: “in limestone from Palermo”); Pfeiffer 1848: 242 (in Sicilia, in montibus circa Panorum); Gaudin 1860: 4–6 (Monte Pellegrino, Monte Billiemi, Monte Gibilforno); Anca 1860a: 312 (fossil: Perciata cave on Monte Gallo); Anca 1860b: 688 (fossil: Perciata cave on Monte Gallo); Tryon 1888: 235–236, pl. 59 figs. 54–57; De Gregorio 1890: 79; Monterosato 1891: 12 (Monte Pellegrino, both fossil and extant); Monterosato 1892: 13; Ancey 1893: 137; Tricomi 1929: 9 (fossil: Addaura cave; Perciata cave; Vitelle cave; Conza cave); Varga 1989: 77.

*Helix mazzullii* var. *zonata* De Cristofori & Jan 1832: 1.

*Helix (Pomatia) Mazzullii* — Beck 1837: 44.

*Helix aspersa* var. *mazzullii* — Rossmässler 1837: 5, pl. 22 figs. 295–296.

*Helix retirugis* Rossmässler 1835: 56 (as a synonym of *Helix mazzullii*; see remarks) — Calcara 1845a: 22 (Monte Pellegrino; Monte Cuccio; Roccazzo; Baida; Pietrazze; fossil: Billiemi; Monte Pellegrino); Calcara 1845b: 21 (fossil: Billiemi; Monte Pellegrino); Dupuy, 1848: 112, pl. 5, fig. 4.

*Helix quinciacensis* — Mauduy 1839: 53, pl. 2, figs. 6–7.

*Helix ritirugis* — [Incorrect subsequent spelling for *retirugis*] Cantraine 1840: 100 (surroundings of Palermo).

*Helix costae* Benoit 1857: 72–76, pl. 1 fig. 10 (Barcellona; Cefalù; Monte Pellegrino; surroundings of Palermo; fossil: Monte Pellegrino and Billiemi, near Palermo).

*Helix Mazzullii* — [Incorrect subsequent spelling for *mazzullii*] Bourguignat 1860: 160–163, pl. 22, figs. 1–2 (Cefalù; Palermo; southern part of Italy, especially Calabria); Pilsbry 1895: 316, 318; Sequenza 1902: 450 (fossil: Santa Teresa di Riva, c.da Grotte, Messina province); Germain 1908: 154.

*Helix mazzulii* var. *zonata* Bourguignat 1860: 162, pl. 22 fig. 3.

*Helix Quincayensis* — [Incorrect subsequent spelling for *quinciacensis*] Bourguignat 1860: 163–166, pl. 22 figs. 4–6 (Palermo; Cefalù).

*Helix mazzulli* — [Incorrect subsequent spelling for *mazzullii*] Anca 1867: 5 (fossil: Perciata cave on Monte Gallo); Monterosato 1872: 10 (fossil: Monte Pellegrino; Ficarazzi); Benoit 1875: 133 (Barcellona; Cefalù; Monte Pellegrino); Monterosato 1877: 42 (fossil: Monte Pellegrino; Ficarazzi); Benoit 1882: 12–13 (Palermo; Cefalù; Barcellona); De Gregorio 1886: 14 (fossil: Castellana, near Bellolampo; Monte Pellegrino); Westerlund 1889: 451 (Sicilien, Süditalien = Sicily, southern Italy); De Gregorio 1916b: 91 (Monte Pellegrino); De Gregorio 1917: 381, pl. 124 fig. 5 (grotte di Palermo); De Gregorio 1927: 13 (fossil: Palermo; Pietrazzi), 17 (fossil: Bisaquino), 20 (fossil: at the base of Monte Pellegrino); De Stefani 1941: 19 (fossil: caves nearby Palermo); Burgio & Cani 1988: 90 (fossil: Alcamo); Abate *et al.* 2006: 27 (fossil: Alcamo).

*Helix Mazulli* — [Incorrect subsequent spelling for *mazzullii*] Falconer 1868: 550 (fossil: Maccagnone cave).

*Helicogena mazzullii* — Paulucci 1878: 8 (Sicily).

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*Helix mazzulli* var. *quincayensis* — Westerlund 1889: 451.

*Helix Costae* (*H. Mazzulli*) — Platania 1890: 45 (Monte Pellegrino; Monte Cuccio); Salvo 1932: 45 (Monte Pellegrino; Monte Cuccio).

*Helix (Ercetella) mazzulli* — [Incorrect subsequent spelling for *mazzullii*] Monterosato 1894: 168–169 (Monte Pellegrino; Capo Gallo).

*Helix (Ercetella) costae* monstr. *contraria* and monstr. *scalariformis* Monterosato 1894: 169.

*Helix (Ercetella) costae* var. ex col. *unicolor, fasciata, albina* Monterosato 1894: 169.

*Helix (Ercetella) costae* var. ex forma: *acuta, arietina, intorta* Monterosato 1894: 169.

*Helix Luparellensis* De Gregorio 1895: 192 (fossil: Luparello cave, near Baida; Palermo) — De Gregorio 1899: 4 (fossil: Luparello cave); Burgio & Costanza 1999: 368 (fossil: Luparello cave).

*Helix mazzulli* var. *lymneopsis* De Gregorio 1895: 193 (near Palermo, maybe Monte Pellegrino).

*Helix mazzulli* var. *normalis* De Gregorio 1895: 193 (Monte Pellegrino; Guadagna, near Palermo).

*Helix mazzulli* var. *supracristata* De Gregorio 1895: 193 (near Palermo).

*Helix mazzulli* var. *zonata* — De Gregorio 1895: 193 (Castelvetrano; San Vito; Marsala; Capo Gallo; Pachino).

*Helix (Helicogena) mazzullii* — Kobelt 1903: 100, pl. 320, figs 5–13.

*Helix (Ercetella) Mazulli* — [Incorrect subsequent spelling for *mazzullii*] De Gregorio 1927: 5 (fossil: Monte Pellegrino).

*Helix Mazzullii* var. *limneopsis* — [Incorrect subsequent spelling for *lymneopsis*] De Gregorio 1927: 16 (Palermo: Addaura cave).

*Helix Mazzullii* F. Luparellensis — De Gregorio 1927: 19 (fossil: Palermo: Luparello cave).

*Helix mazzulli* — [Incorrect subsequent spelling for *mazzullii*] Kuehnelt 1932: 132 (Monte Pellegrino); Rensch 1932a: 792 (Monte Pellegrino); Rensch 1937: 568 (Monte Pellegrino).

*Helix (Cryptomphalus) mazzullii* — Zilch 1952: 136; Sacchi 1955a: 83–87, pl. 3, fig. E (Monte Pellegrino; Cefalù; Monte Gallo; Monte Palmeto; Capo San Vito; Monte Cofano; Custonaci); Alzona 1971: 220 (northwestern Sicily, from Palermo to Trapani); Cesari 1978: 58 (Monte Pellegrino; Cefalù, La Rocca; Custonaci); Falconieri 1995: 131.

*Cryptomphalus mazzullii* — Sacchi 1955b: 635–642 (Capo Gallo; Capo San Vito; Custonaci; Cefalù; Isola delle Femmine).

*Helix (Cornu) mazzullii* — Esu et al. 1986: 235 (fossil: Poggio Schinaldo); Giusti & Castagnolo 1982: 89.

*Cantareus mazzullii* — Manganelli et al. 1995: 33; Manganelli et al. 2000: 41–42 (from Trapani to Cefalù); Vitturi et al. 2005: 351–357 (Monte Pellegrino); Lo Brano & Sparacio 2006: 584 (Monte Cuccio; Monte Pellegrino; Capo Gallo).

*Helix mazzulli mazzulli* — [Incorrect subsequent spelling for *mazzullii*] Cossignani & Cossignani 1995: 142–143 (northwestern Sicily, Monte Pellegrino).

*Cornu mazzullii* — Bank 2009 (Sicily); Colombo et al. 2008: 90.

*Ercetella mazzullii* — Liberto et al. 2010: 113.

*Helix mazzuli* — [Incorrect subsequent spelling for *mazzullii*] Benoit (date?, see Benoit 1857–1862): pl. 11 fig. 9 (Monte Pellegrino).

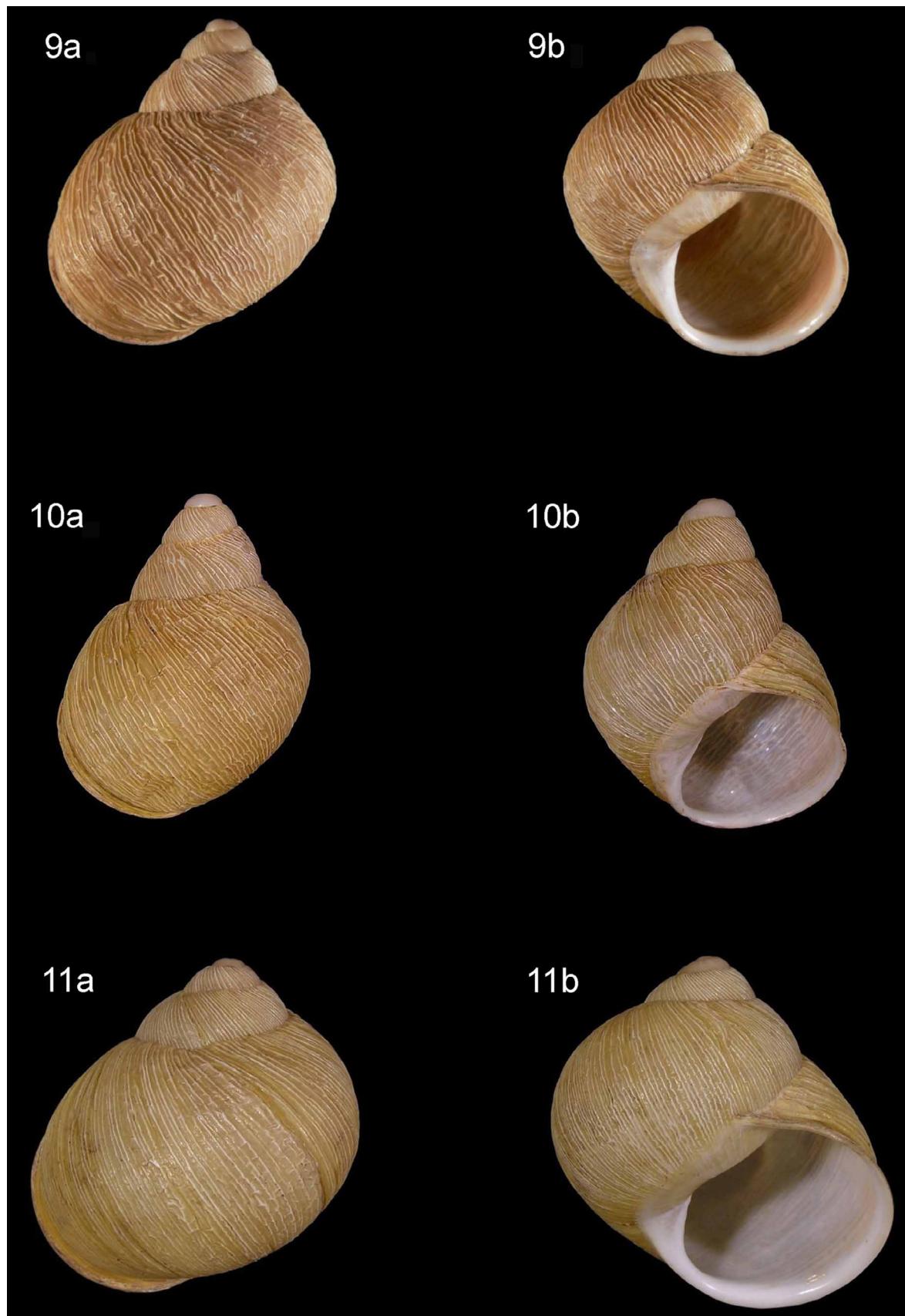
*Helix mazzuli* var. *zonata* — Benoit (date?, see Benoit 1857–1862): pl. 11 fig. 10.

**Shell** (Figs. 9–14). Globose-conical, light; height 25–36 mm (mean 31.5 mm); maximum diameter 24–33 mm (mean 27.5 mm); aperture height 17–26 mm (mean 20.7 mm); aperture maximum diameter 18–24 mm (mean 21.8 mm); uniformly yellowish or brownish, sometimes with 3–5 brown spiral bands; external surface with thin, uniform, axial wrinkles; aperture very large, round; peristome simple, sharp, little thickened and sometimes a little reflected. (Rossmässler 1837; Pfeiffer 1848; Benoit 1857; Westerlund 1889; Cesari 1978; Falconieri 1995)

**Radula.** Similar to that of *C. aspersum* (Pollonera 1887; Hesse 1919; Giusti et al. 1995) but differs in the less evident mesocone of the central tooth. (Hesse 1919; Cockerell 1921; Cesari 1978).

**Genitalia** (Fig. 21). Diverticulum of bursa copulatrix longer (nearly two-fold) than duct of bursa copulatrix; two digitiform glands each of which is divided into 20–35 slender and branched digit-like appendices; vagina internally with 5–6 longitudinal pleats; penial flagellum as long as penis and epiphallus together; proximal cavity of distal penis with some transversal pleats, a very small penial papilla and a small papilla on the internal wall (Hesse 1919; Cesari 1978; Giusti & Castagnolo 1982).

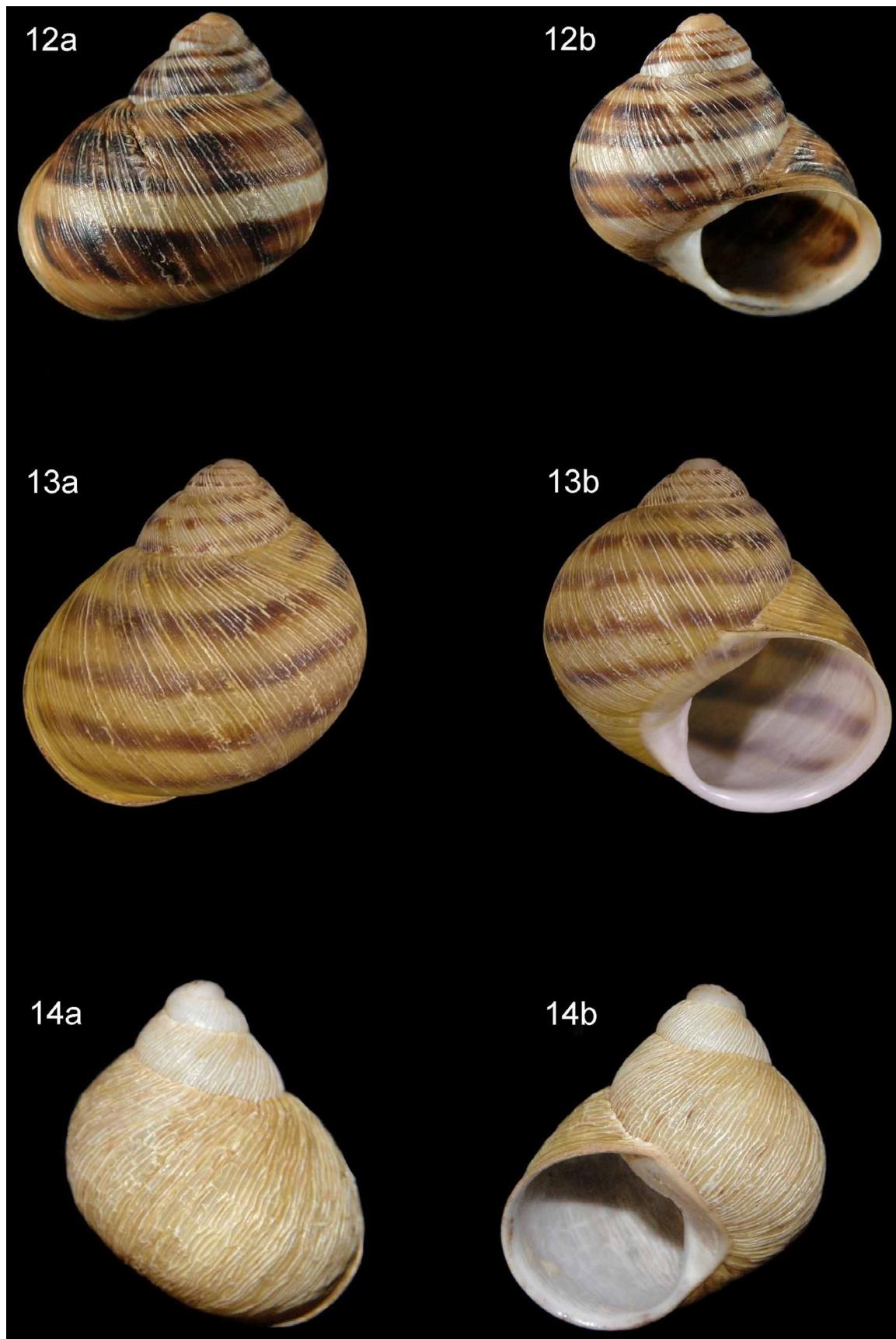
**Remarks.** Menke (1830) introduced the name *H. retirugis* as a *nomen nudum*. Rossmässler's citation of the name as a synonym of *Helix mazzullii* and its adoption as a valid name by Cantraine (1840) and subsequent authors make the name available and Rossmässler its nomenclatural author (ICZN 1999 Art. 11.6). The type material consists of the specimens “denoted by that name when it was adopted as the name of a taxon” (ICZN 1999 Art. 72.4.3). This was in 1840 by Cantraine (misspelled as *ritirugis*) who cited the species as being found “dans les environs de Palerme” (in the surroundings of Palermo). Until the nominal species has been typified (either with material of Cantraine or by a neotype) it remains unclear whether the name *retirugis* denotes the globose genuine *mazzullii* or the conical form of Monte Pellegrino.



**FIGURES 9–11.** *Erctella mazzullii* samples from various sites showing different morphotypes. Each shell shown in frontal (a) and dorsal (b) view. **9.** Palermo, Monte Pellegrino, height (H): 30 mm, maximum diameter (D): 27 mm. **10.** Palermo, Monte Pellegrino, H: 32 mm, D: 24 mm. **11.** Palermo, Cinisi, Monte Pecoraro, H: 35 mm, D: 33 mm.

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**FIGURES 12–14.** *Erctella mazzullii* samples from various sites showing different morphotypes. Each shell shown in frontal (a) and dorsal (b) view. **12.** Palermo, Sferracavallo, la Montagnola, H: 29 mm, D: 28 mm. **13.** Palermo, Cinisi, Monte Pecoraro, H: 36 mm, D: 34 mm. **14.** *Sine loco*, Paleontological Museum of University of Catania (MPUC), Monterosato collection, Lot 1187, H: 31 mm, D: 30 mm, sinistral shell.

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Bourguignat (1860), in line with Dupuy (1848), stressed that *Helix quinciacensis* Mauduyt 1839 was described on specimens of *C. mazzullii* introduced to Quinçay (France). Nevertheless, he considered it a valid species (with the surroundings of Palermo and Cefalù being the place of origin) for which he unjustifiedly emended Mauduyt's name to *Quincayensis*.

Zilch (1952), given that the entire De Cristofori & Jan malacological collection was destroyed (Zilch 1952), designated the specimen depicted by Rossmässler (1837: pl. 22 fig. 295) as the neotype of *Helix mazzullii*.

The neotype summarizes the main morphological features shown by the specimens of the populations inhabiting the surroundings of Palermo. However, the populations from Monte Pellegrino appear with a more conical, at times turreted shell, showing more evident sutures and an emphasized shell sculpture. Such morphological differences might account for elevating this population to the subspecies level. In that case, bearing in mind the description by Mauduyt (1839) and illustrations by Bourguignat (1860: pl. 22 figs. 4, 6 sub *quincayensis*), it should be reported under the name *Erectella mazzullii quinciacensis* (Mauduyt 1839).

De Gregorio described the fossil species *Helix Luparellensis* (Fig. 32) from the Luparello cave (Baida, Palermo) belonging to the *H. mazzullii* group (De Gregorio 1895; 1899). Later on, he re-considered this taxon as *C. mazzullii* (De Gregorio 1927), contrary to Monterosato's opinion who believed *H. luparellensis* to be a valid species (see Ryolo & Palazzi 2009).

***Erectella cephalaeeditana* (Giannuzzi-Savelli, Oliva & Sparacio, 1986)**

Figs. 15–17, 22

*Helix mazzullii* var. β Pirajno 1840: 13 (Cefalù, La Rocca; Rocca di San Nicola).

*Helix cephalaeeditana* Giannuzzi-Savelli et al. 1986: 203–205 (Cefalù, La Rocca).

*Cantareus cephalaeeditana* — Manganelli et al. 1995: 33.

*Helix mazzulli cephalaeeditana* — Cossignani & Cossignani 1995, pp. 27, 142–143 (Cefalù, La Rocca); Piazza 2003, pp. 3–30 (Cefalù, La Rocca).

*Helix mazzullii cephalaeeditana* var. *piazzensis* Piazza 2003: 31–32 (Cefalù, La Rocca).

*Cornu mazzullii cephalaeeditanum* — Colombara et al. 2008: 90.

*Erectella cephalaeeditana* — Liberto et al. 2010: 115, fig. 155.

**Shell** (Figs. 15–17). Small for an *Erectella*; height 24–32 mm (mean 26 mm); maximum diameter 21–28 mm (mean 23 mm); aperture height 15–20.5 mm (mean 16 mm); aperture maximum diameter 16–23 mm (mean 19 mm); spire more elevated, external surface of last two whorls, strongly wrinkled and irregularly reticulated; peristome thickened and well reflected; aperture oval (Pirajno 1840; Giannuzzi-Savelli et al. 1986).

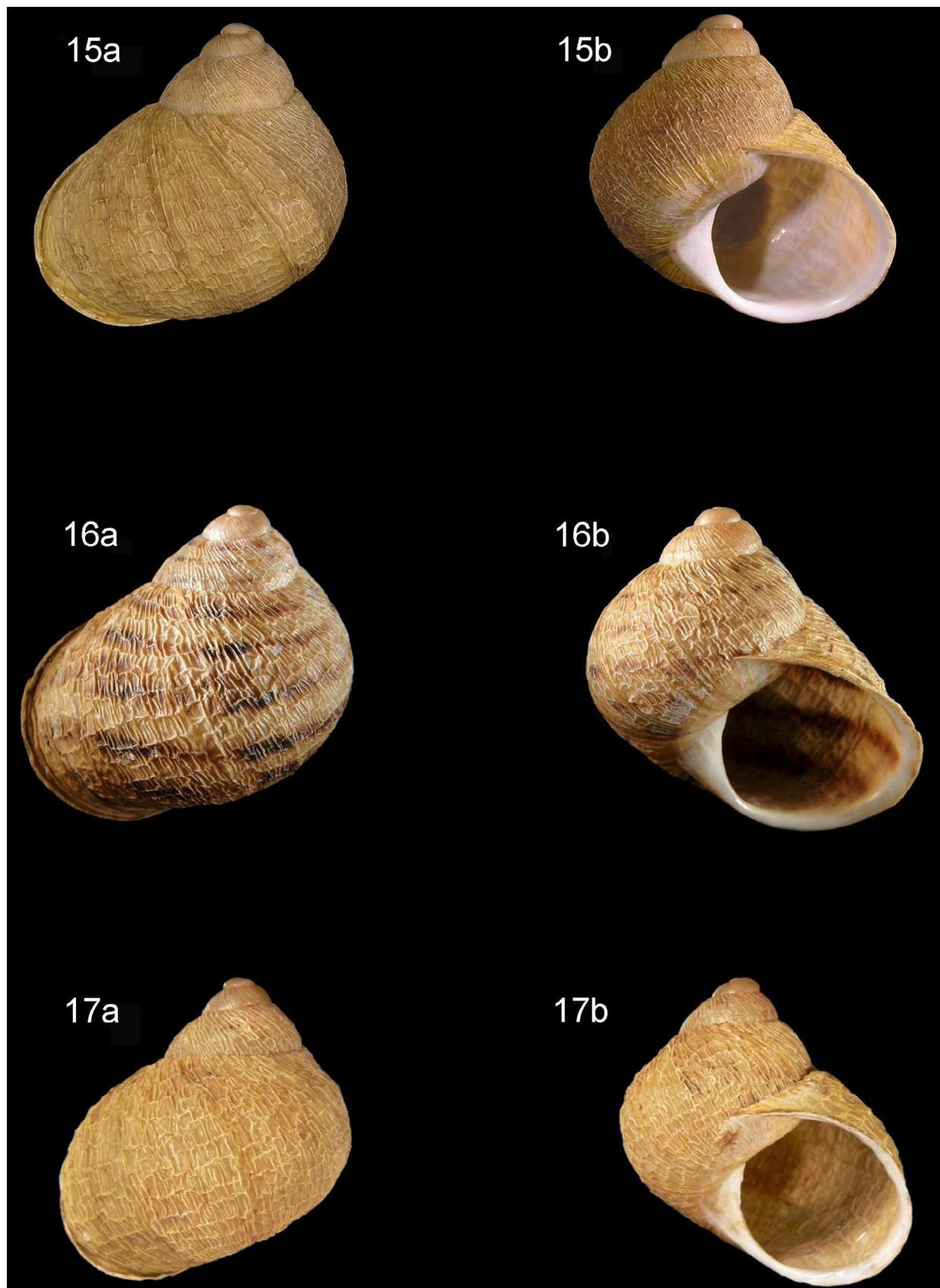
**Genitalia** (Fig. 22). Diverticulum of bursa copulatrix 1.2–1.5 times as long as duct of bursa copulatrix; two digitiform glands each of which is divided into 11–18 slender and branched digit-like appendices; vagina internally with 4–5 longitudinal pleats; penial flagellum usually shorter than penis and epiphallus together; proximal cavity of distal penis having some transversal, strongly raised pleats, a very small true penial papilla and a small papilla on the internal wall.

**Remarks.** For the taxon comprising specimens from Cefalù, La Rocca, formerly identified as *Helix mazzullii* var. β (Pirajno 1840), Giannuzzi-Savelli et al. (1986) introduced the name *H. cephalaeeditana* (as *Helix cephalaeeditana* Pirajno, ms.). Manganelli et al. (1995) although acknowledging the availability of the name (ICZN 1985 Articles 11d, 13a), nevertheless considered *cephalaeditana* a synonym of *mazzullii*.

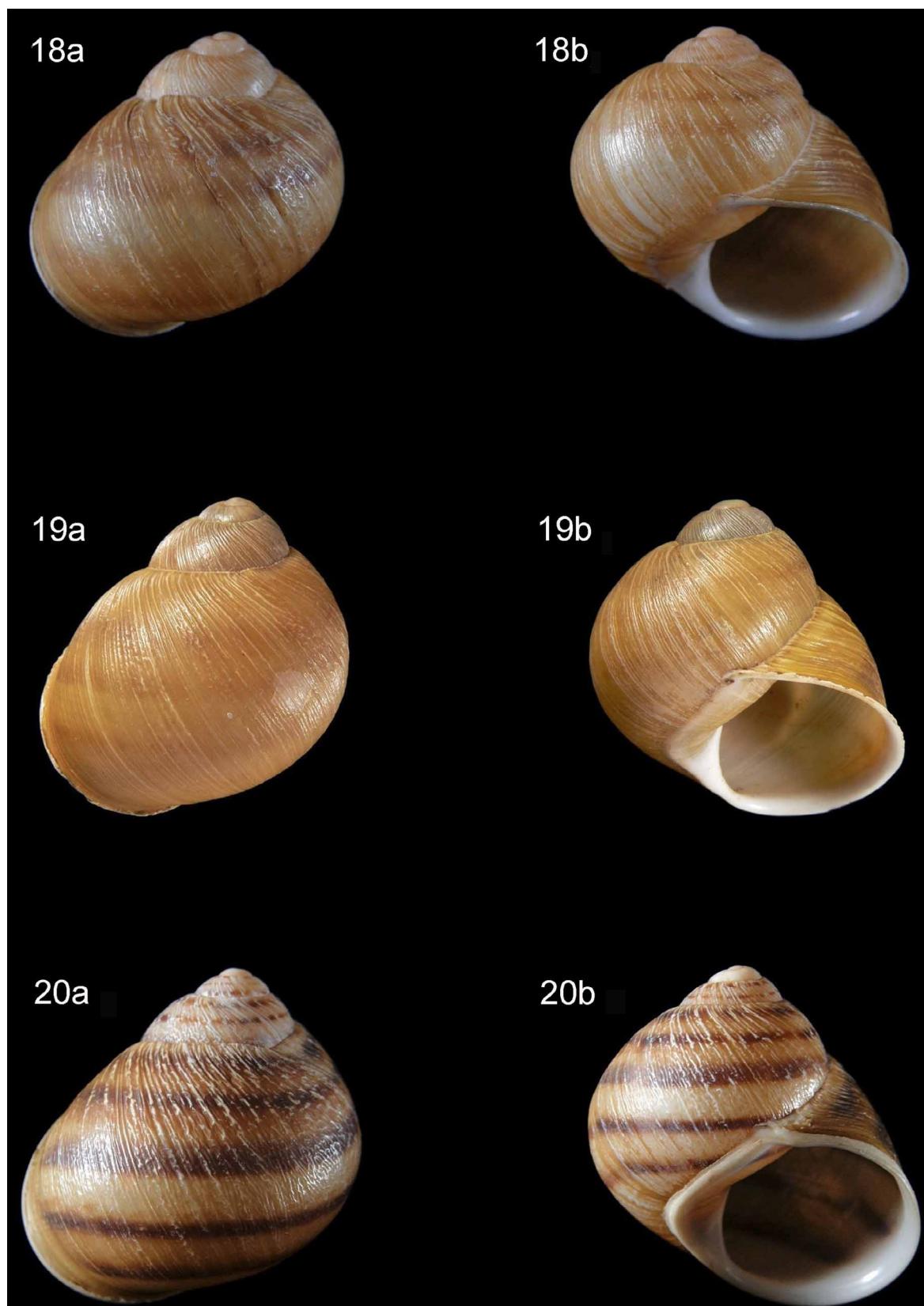
Bourguignat (1860), when reporting the distribution area of “*Helix quincayensis*” (= *H. quinciacensis*), mentioned also Cefalù in addition to the surroundings of Palermo; nevertheless, taking into account the description of the taxon provided by the author and illustrations included in his work (pl. 22 figs. 4 and 6) a synonymy between *H. quincayensis* and *cephalaeditana*, in our opinion, can be ruled out.

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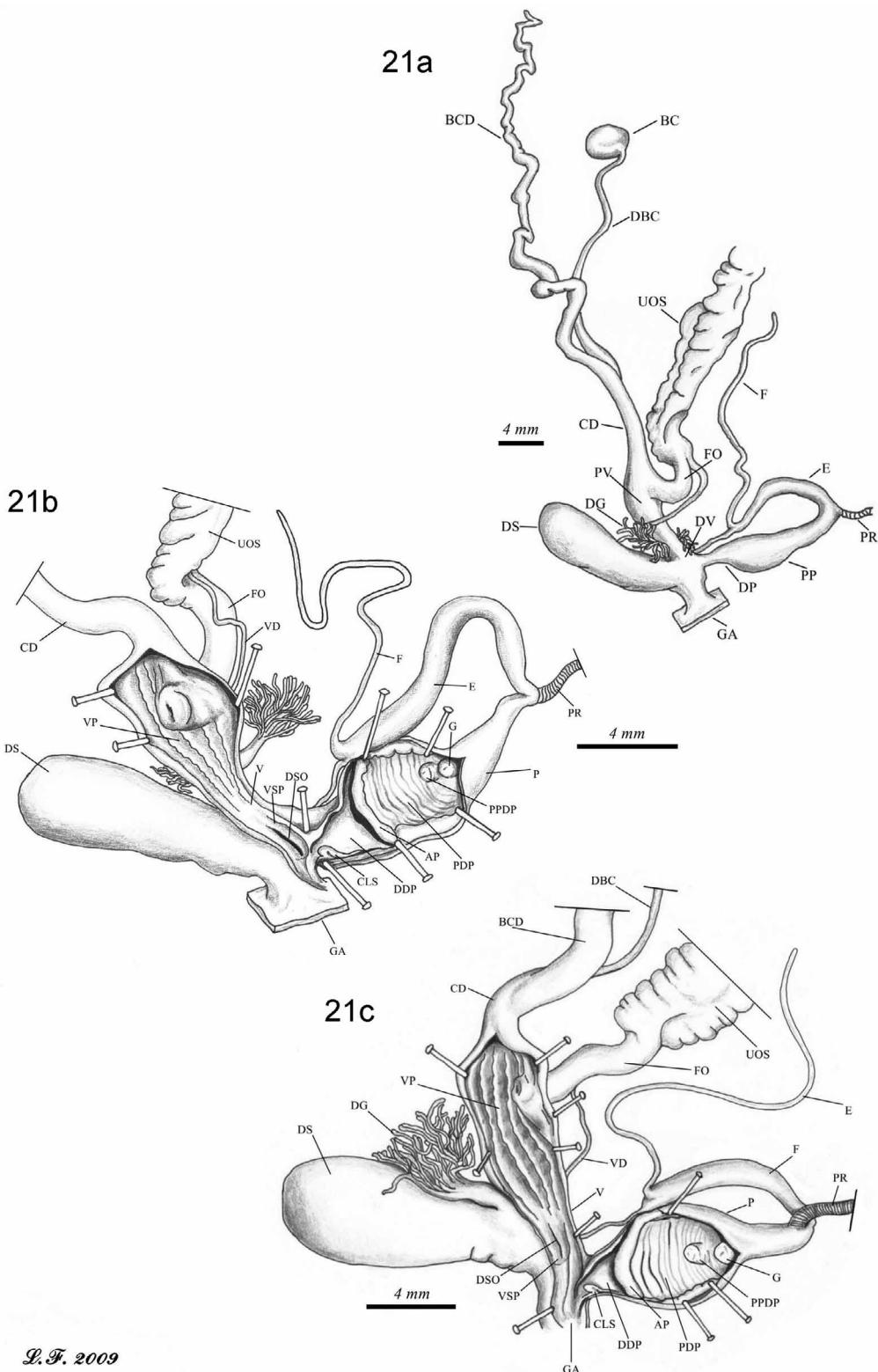
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**FIGURES 15–17.** *Erctella cephalaeeditana* samples from Cefalù, La Rocca showing different morphotypes. Each shell shown in frontal (a) and dorsal (b) view. **15.** height (H): 30 mm, maximum diameter (D): 29 mm. **16.** H: 31 mm, D: 26 mm. **17.** H: 25 mm, D: 23 mm.



**FIGURES 18–20.** *Erctella insolida* samples from several sites of the Trapani province showing different morphotypes. Each shell shown in frontal (a) and dorsal (b) view. **18.** San Vito lo Capo, Cala Mancina, height (H): 34 mm, maximum diameter (D): 30 mm. **19.** Custonaci, Monte S. Giovanni, H: 37 mm, D: 34 mm. **20.** San Vito lo Capo, Monte Passo del Lupo, H: 40 mm, D: 37 mm.



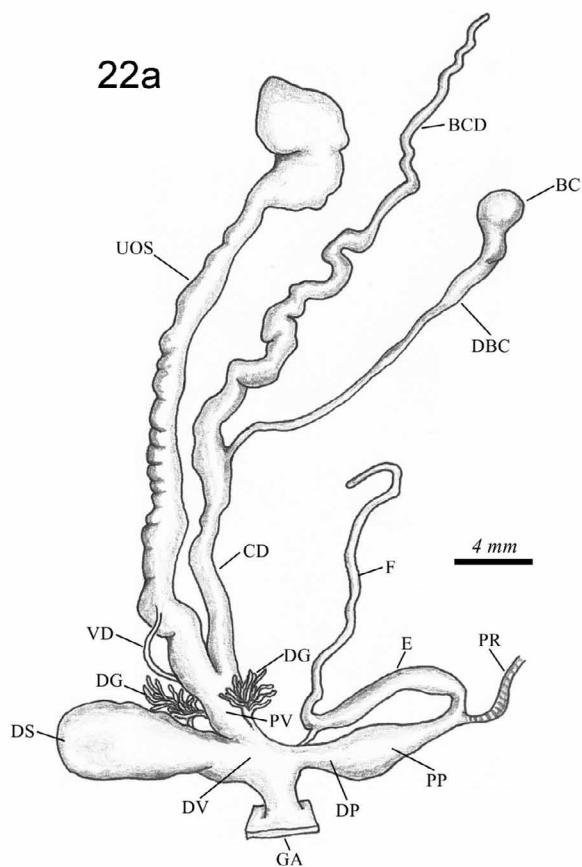
**FIGURE 21.** a. *Ercitella mazzullii* from Monte Pellegrino, genitalia. b. *Idem*, internal structures shown. c. *Ercitella mazzullii* from Monte Pecoraro, Genitalia with internal structure.

Acronyms in figures 21–26: AP = annular pad; BC = bursa copulatrix; BCD = diverticulum of bursa copulatrix; CD = copulatory duct; CLS = crest-like structure; DG = digitiform glands; DP = distal penis; DS = dart sac; DV = distal vagina; DBC = duct of the bursa copulatrix; DDP = distal cavity of the distal penis; DSO = dart sac opening; E = epiphallus; F = flagellum; FO = free oviduct; G = penial papilla; GA = genital atrium; P = penis; PDP = proximal cavity of the distal penis; PR = penial retractor muscle; PV = proximal vagina; UOS = uterine ovispermiduct; V = vagina; VD = vas deferens; VP = vaginal pleats; VSP = V-shaped pleat.

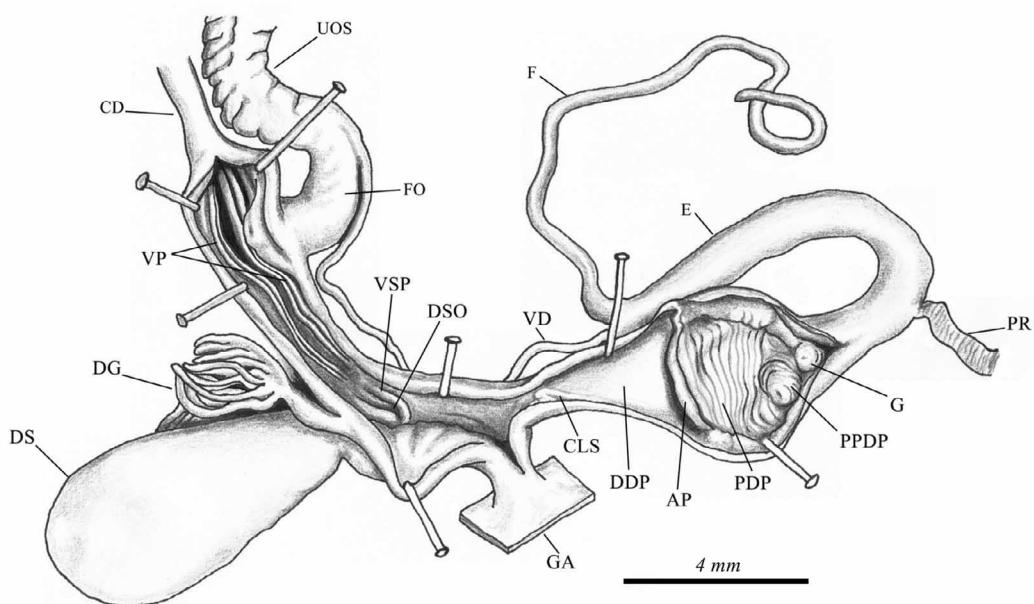
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22a



22b

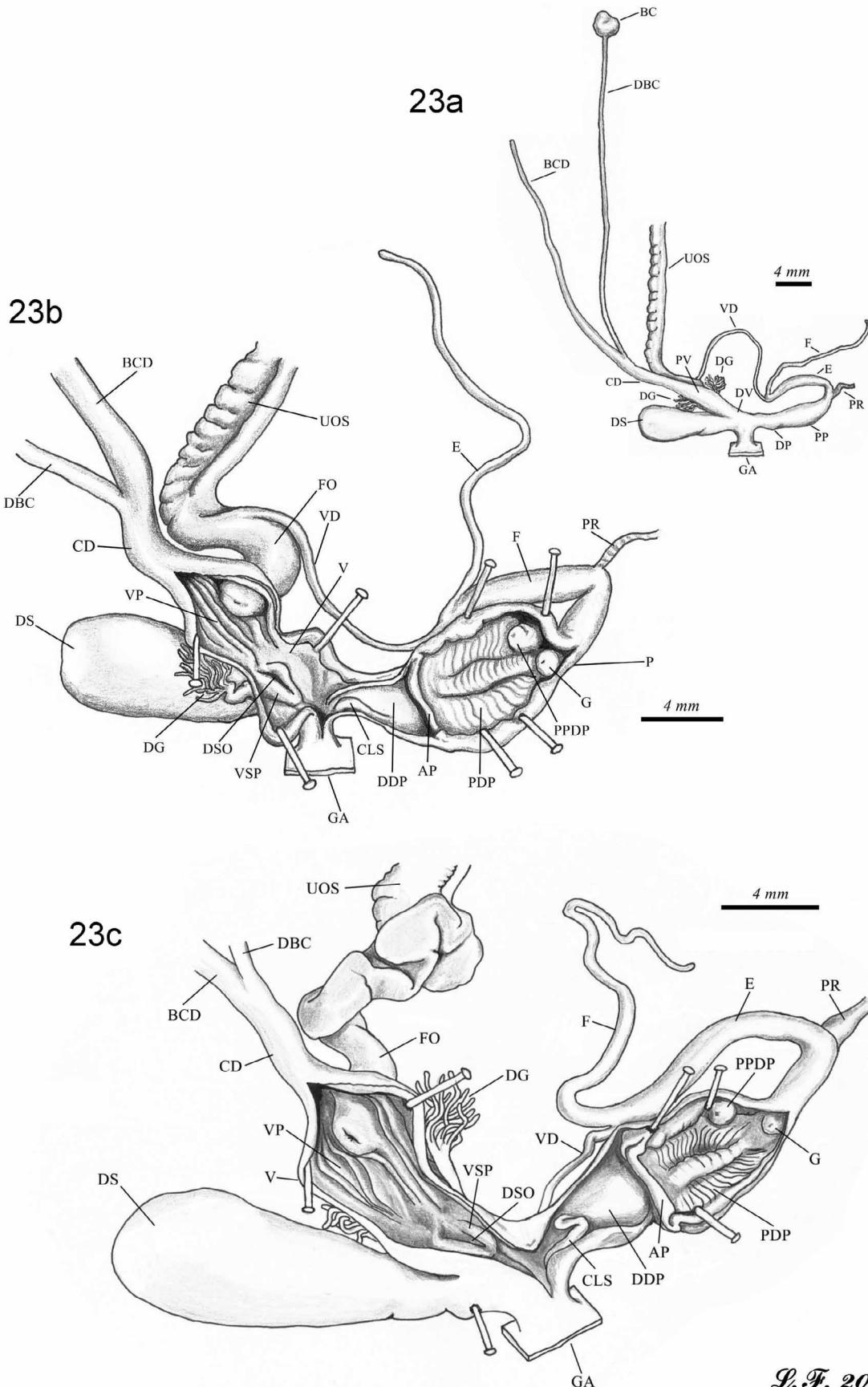


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**FIGURE 22. a.** *Erctella cephalaeeditana* from Cefalù, La Rocca, genitalia. **b.** *Idem*, internal structures shown.

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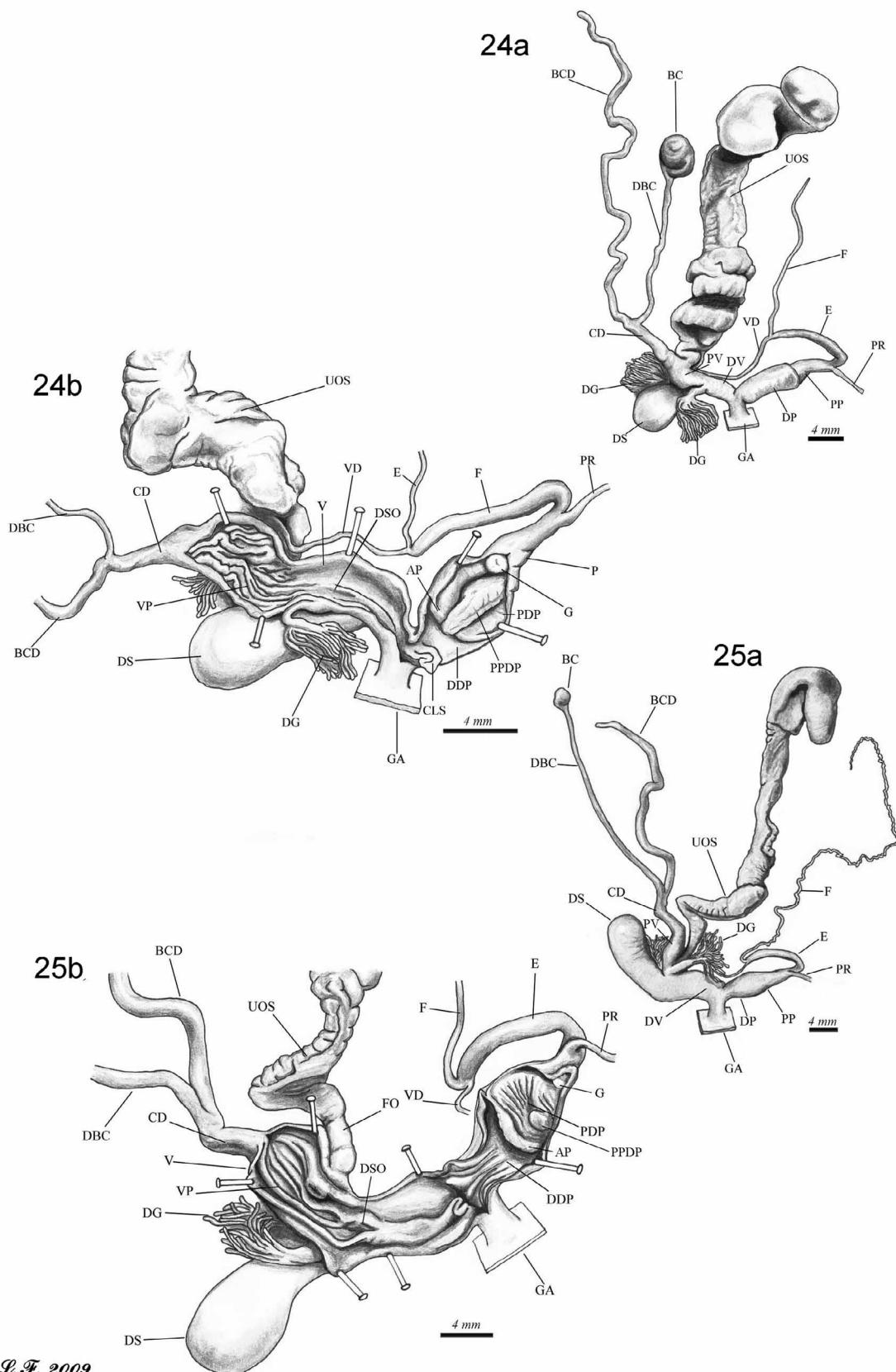


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**FIGURE 23.** **a.** *Erctella insolida* from Monte Cofano, genitalia. **b.** *Idem*, internal structures shown. **c.** *Erctella insolida* from San Vito lo Capo, Genitalia, internal structures shown.

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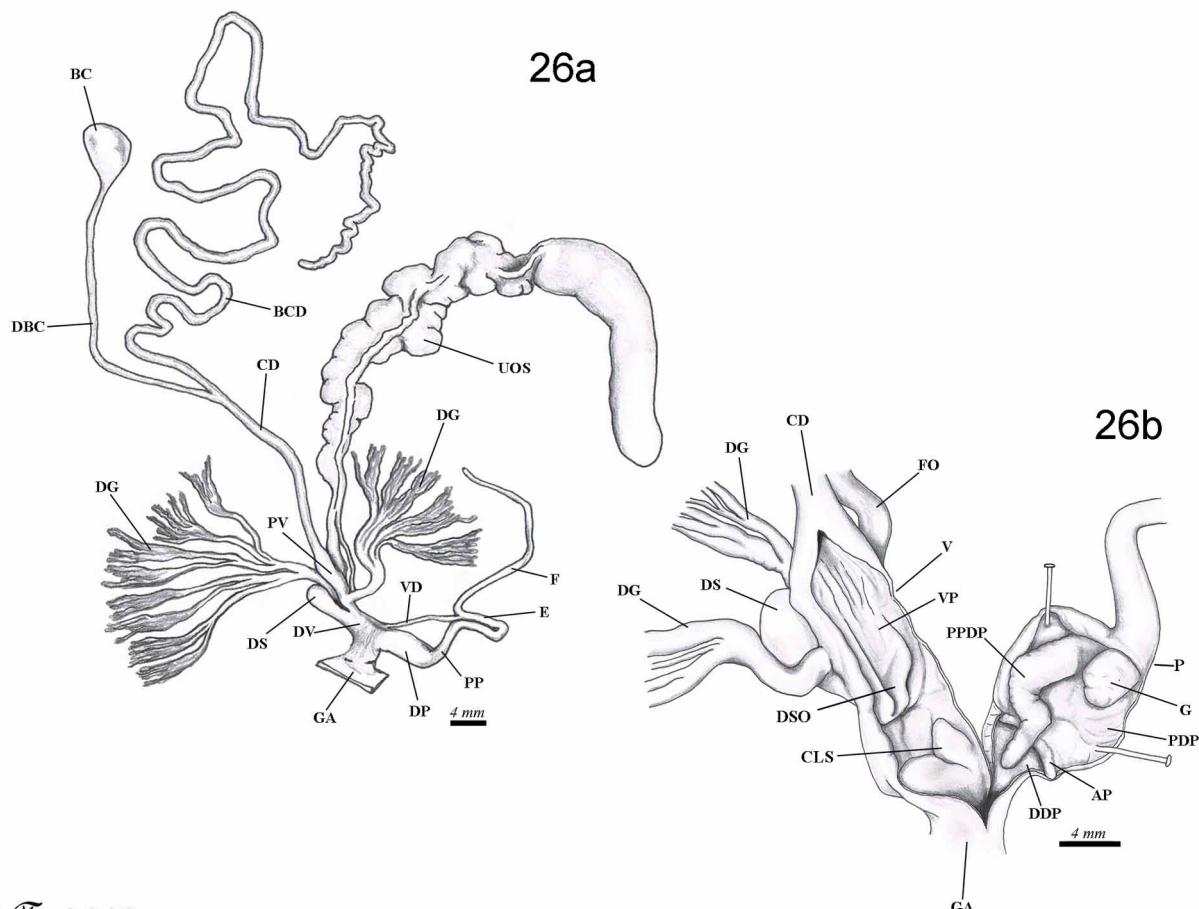


**FIGURE 24. a.** *Cornu aspersum* from Bosco Ficuzza (Palermo), genitalia. **b.** *Idem*, internal structures shown.

**FIGURE 25. a.** *Cantareus apertus* from Termini Imerese (Palermo), genitalia. **b.** *Idem*, internal structures shown.

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**FIGURE 26.** a. *Eobania vermiculata* from Collesano (Palermo) genitalia. b. *Idem*, internal structures shown.

**FIGURE 27.** *Cantareus apertus* from Termini Imerese (Palermo).

**FIGURE 28.** *Cornu aspersum* from Bosco Ficuzza (Palermo).

**FIGURE 29.** *Eobania vermiculata* from Collesano (Palermo).

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30a



30b



30c



30d



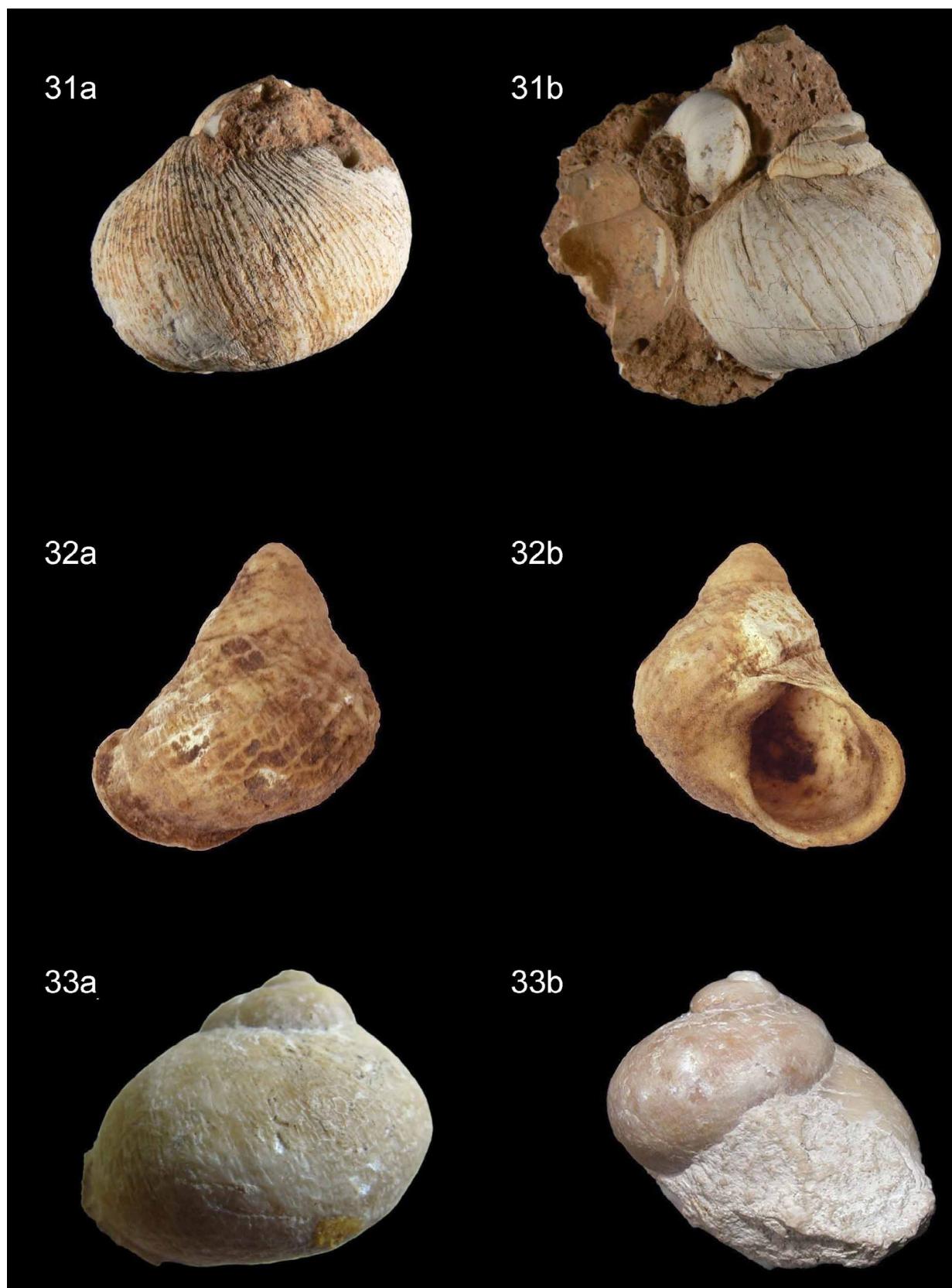
30e



30f



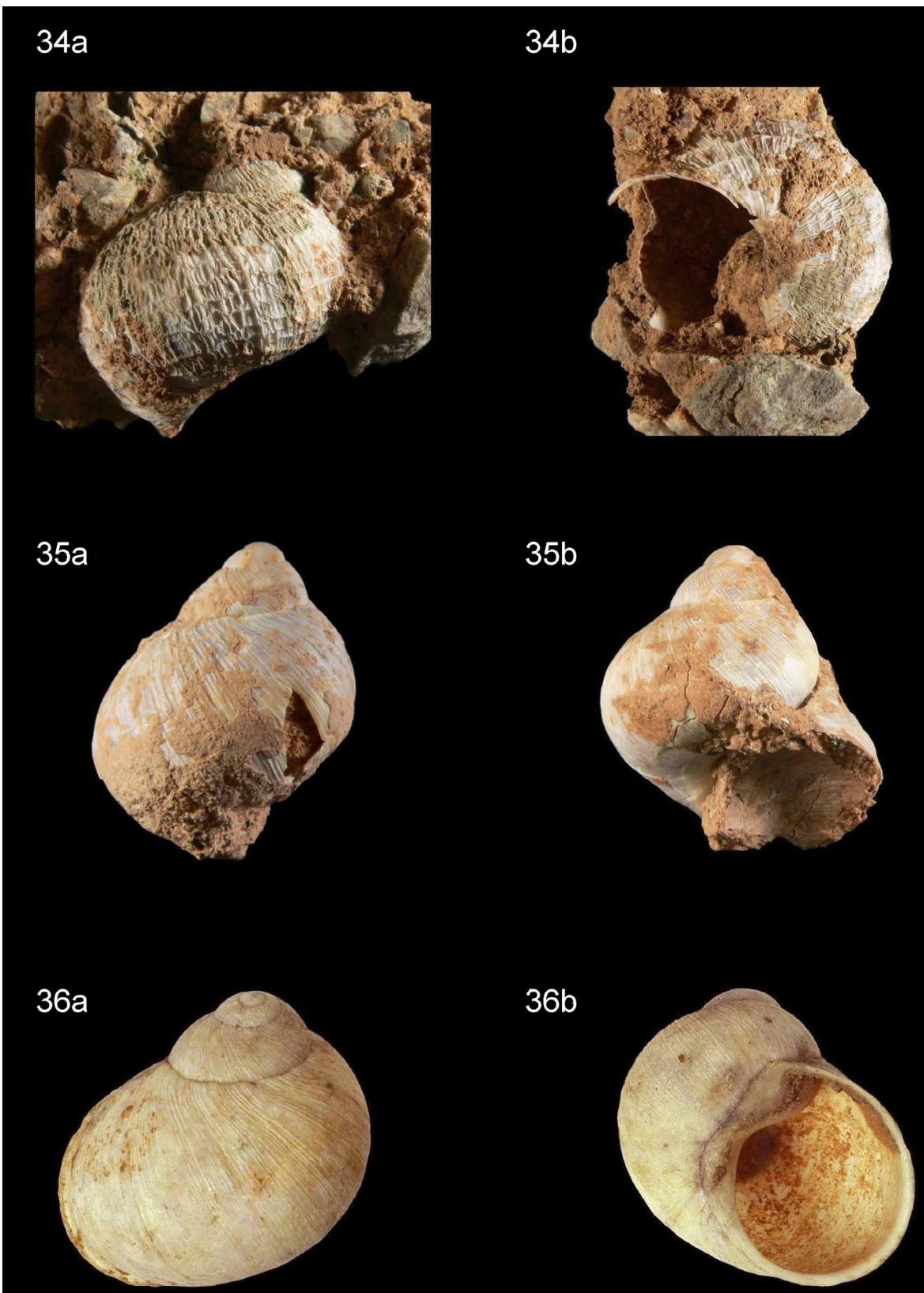
**FIGURE 30.** Tunnels dug into the limestone by *Erctella* specimens. **a.** Palermo, Monte Pellegrino, holes made by *E. mazzullii*. **b.** Trapani, Custonaci, Monte Cofano, a broad cavity in the rock, resulting from the progressive enlargement of single tunnels and consequent collapse of dividing walls, made by *E. insolida*. Hole size: 25x60 cm. **c.** Trapani, San Vito lo Capo, Cala Mancina, the innermost part of some tunnels. **d.** Cefalù, La Rocca: peculiar calcium carbonate concretions close to the opening of tunnels dug by *E. cephalaeeditana* specimens. These concretions are the result of the evaporation of water from the  $\text{H}_2\text{CO}_3$ -rich mucus used by the molluscs to dissolve the calcium carbonate of the rock. **e.** Messina, Militello Rosmarino, Monte Scurzi, holes in the limestone which can reasonably be attributed to the siccavous abilities of *Erctella* specimens occurring in that area some times ago. **f.** Pizzo Giurafelle, Gratteri (Palermo province), holes in the limestone probably made by *Erctella*.



**FIGURE 31.** *Ercatella mazzullii*, two fossils from Monte Pellegrino. **a.** H: 34 mm, D: 35 mm. **b.** H: 30 mm, D: 31 mm.

**FIGURE 32.** *Ercatella luparellensis*, one fossil shown in frontal (**a**) and dorsal (**b**) view, Luparello cave, Baida, Palermo (Museum of Terrasini, Palermo), H: 30 mm, D: 28 mm.

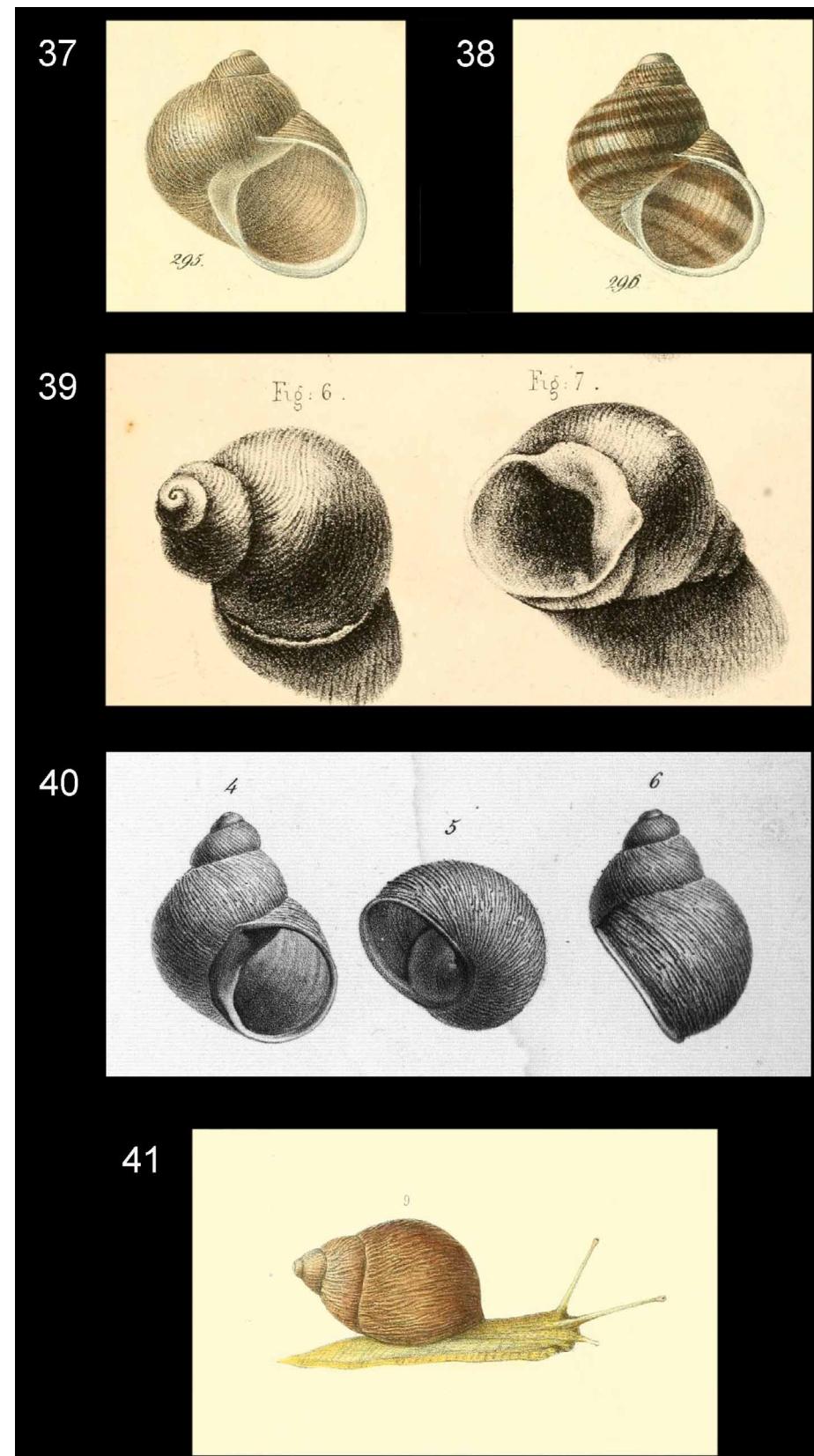
**FIGURE 33.** *Ercatella cf. mazzullii*, one fossil shown in frontal (**a**) and dorsal (**b**) view, Alcamo, Trapani (Geological Museum "G.G. Gemellaro", Palermo) H: 29 mm, D: 30 mm.



**FIGURE 34.** *Ercatella cephalaeeditana*, one fossil shown in frontal (**a**) and basal (**b**) view, Cefalù, La Rocca, H: 36 mm, D: 38 mm.

**FIGURE 35.** *Ercatella insolida*, two fossils from San Vito lo Capo, Cala Mancina. **a.** H: 32 mm, D: 27 mm. **b.** H: 34.5 mm, D: 31.5 mm.

**FIGURE 36.** *Ercatella insolida*, one fossil shown in frontal (**a**) and dorsal (**b**) view, Favignana, Egadi Islands, Trapani (Museum of Terrasini, Palermo), H: 42 mm, D: 37 mm.

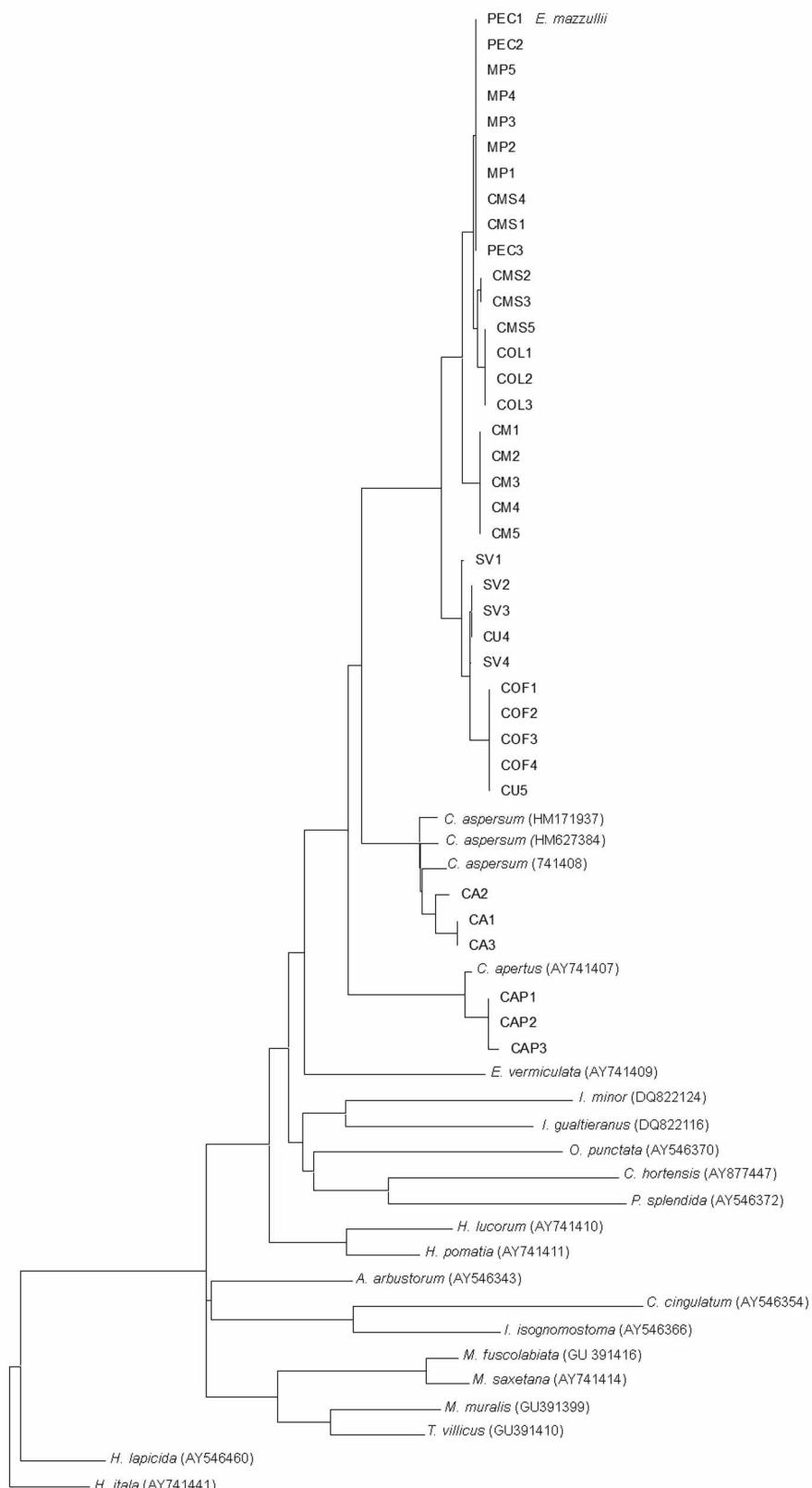


**FIGURES 37–38.** Reproduction of figures showing *Helix aspersa* var. *mazzullii* from Rossmässler (1837: pl. 22 figs. 295–296).

**FIGURE 39.** Reproduction of figures showing *Helix quinciacensis* from Mauduyt (1839: 53, pl. 2, figs. 6–7).

**FIGURE 40.** Reproduction of figures showing *Helix quincayensis* from Bourguignat (1860: pl. 22, figs. 4–6) (Palermo; Cefalù).

**FIGURE 41.** Reproduction of figure showing *Helix mazzuli* from Benoit (date? [see Benoit 1857–1862]: pl. 11 fig. 9).



**FIGURE 42.** Maximum Likelihood consensus tree inferred from the 16S rDNA sequence alignment representing a possible reconstruction of Helicidae phylogeny. Initial trees for the heuristic search were obtained automatically. A GTR +  $\Gamma$  model (alpha= 0.29) was employed. The analysis involved 58 nucleotide sequences. All positions containing gaps and missing data were eliminated.

er a varietate: erictina.  
 ... Appartiene al genere  
Erectella, Monts.  
 (da Erecta = P. pellegrini)  
 da me pubblicato e adottato  
 da Dr. Pilsbry, per  
 suoi caratteri anatomici  
 ci differisce notevolmente dal Cryptomphalus  
(aspersa)

**FIGURE 43.** Autographic note by Monterosato (Civic Museum of Natural History, Milan) commenting on several *mazzullii* specimens caught on Monte Pellegrino in 1912: "... Appartiene al genere *Erectella*, Monts. . da me pubblicato e adottato da Pilsbry. Pei suoi caratteri anatomici differisce notevolmente dal *Cryptomphalus (aspersa)*" (for English translation see Discussion section).

### *Erectella insolida* (Monterosato, 1892)

Figs. 18–20, 23

*Helix (Helicogena) aspersa* var. *insolida* Monterosato 1892: 26 (Favignana?; Monte San Giuliano).

*Helix (Erectella) mazzullii* forma *insolida* — Monterosato 1894: 168–169 (Monte San Giuliano or Erice, near Trapani).

*Helix mazzulli* — De Gregorio 1894: 7 (fossil, Levanzo island); Piperno 1997: 140 (fossil: Uzzo cave).

? *Helix* sp. — Varga 1989: 77 (*insolida*?).

*Helix mazzulli* var. *vitincola* De Gregorio 1895: 193 (San Vito).

*Helix (Cryptomphalus) aspersa* — Alzona 1971: 220 [partim].

*Cornu aspersum* — Bank 2009 [partim], (Sicily).

*Cantareus mazzullii* — Fiorentino et al. 2004: 89 (fossil: Levanzo island).

*Cornu mazzullii insolidum* — Colomba et al. 2008: 90.

*Erectella insolida* — Liberto et al. 2010: 115–116, fig. 156.

**Shell** (Figs. 18–20). Large for the genus; height 33–40 mm (mean 34 mm); maximum diameter 29–35 mm (mean 31.8 mm); aperture height 22–26 mm (mean 24 mm); aperture maximum diameter 23.5–28 mm (mean 25 mm); more globose; brownish or greenish; spire less elevated; external surface with very thin growth lines; peristome thickened, simple or little reflected; aperture rounded.

**Animal.** Generally dark.

**Genitalia** (Fig. 23). Diverticulum of bursa copulatrix approximately as long as duct of bursa copulatrix; two

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digitiform glands each of which is divided into 23–45 slender and branched digit-like appendices; vagina internally with 7–8 longitudinal pleats; penial flagellum as long as penis and epiphallus together; internal surface of proximal cavity of the distal penis with transversal pleats subdivided in 1–2 longitudinal and raised pleats, a very small true penial papilla and a small papilla on the internal wall.

**Remarks.** *Ercetella insolida* was described by Monterosato (1892) as a variety of *Helix (Helicogena) aspersa*, with the locus typicus: “Favignana ?” and Monte San Giuliano (Trapani province). The Sicilian naturalist himself later (1894) modified the position of the taxon and slightly re-defined its distribution area, reporting it as a variety of *H. (Ercetella) mazzullii* from Monte San Giuliano or Erice (Favignana being definitely ruled out). Afterward he changed his mind again and considered *insolida* a valid species [*H. (Ercetella) insolida*] distinct from other related species, particularly from *H. aspersa* (= *Cornu aspersum*) (Ryolo & Palazzi 2009).

Populations described by De Gregorio (1895) under the name *vitincola* (from San Vito Lo Capo, Trapani province) and those reported under *insolida* (from Monte Cofano) show minimal morphological differences and only a slight genetic divergence.

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## APPENDIX 1.

Additional material including samples or records from relevant private and/or museum collections.

Acronyms: CG — S. Giglio collection, Cefalù; CL — F. Liberto collection, Cefalù; CMT — Museo di Terrasini; CR — A. Reitano collection, Tremestieri Etneo; CS — I. Sparacio collection, Palermo; MM — Museo Civico di Storia Naturale di Milano; MSNG — Museo Civico di Storia Naturale di Genova “G. Doria”.

### *Ercetella mazzullii* (De Cristofori & Jan, 1832)

Records: Sicily, Palermo: Sicily, 5 exx., coll. C. Tapparone Canefri, ante 1891 (MSNG); Capaci (PA), 2 exx (the original note is by the Marquis Allery di Monterosato) (MSNG); Palermo, 1 ex, leg. Dufour, collection G. and M. Doria, ante 1867 (MSNG); Sicily, 2 exx, coll. of the Prince Oddone, ante 1866 (MSNG); Sicily, 1 ex (MSNG); Sicily, 2 exx (MSNG); north-western Sicily, ante 1900, ex coll. M.I.Z.U.P. in coll. V.E.Orlando (CMT); Monte Pellegrino, ante 1900, (quaternary period) ex coll. M.I.Z.U.P. in coll. V.E.Orlando (CMT); mountains nearby Palermo, ante 1950, leg. T.De Stefani, ex coll. V.E.Orlando (CMT); Sferracavallo and nearby mountains, ante 1950, leg. T.De Stefani, ex coll. V.E.Orlando (CMT); Sferracavallo, Grotta della Mandra, ante 1950, legit T.De Stefani, ex coll. V.E.Orlando (CMT); moutains nearby Sferracavallo, ante 1950, leg. T.De Stefani, ex coll. V.E.Orlando (CMT); Palermo surroundings, ante 1950, leg. T.De Stefani, ex coll. V.E.Orlando (CMT); Monte Cuccio, ante 1950, leg. T.De Stefani, ex coll. V.E.Orlando (CMT); Valdesi, ante 1950, leg. T.De Stefani, ex coll. V.E.Orlando (CMT); Monte Gallo, Grotta dei Vitelli, ante 1950, leg. T.De Stefani, ex coll. V.E.Orlando (CMT); base of Monte Pellegrino, ante 1960, fossils, leg. T.De Stefani, ex coll. V.E.Orlando (CMT); base of Monte Pellegrino, Belmonte mine, ante 1960, fossils, leg. T.De Stefani, ex coll. V.E.Orlando (CMT); Sferracavallo, Grotta Elia De Stefani, ante 1960, fossils, leg. T.De Stefani, ex coll. V.E.Orlando (CMT); Tommaso Natale, Grotta della Conza, ante 1960, fossils, leg. T.De Stefani, ex coll. V.E.Orlando (CMT); near Baida, continental sandy limestone, ante 1960, *H. luparellensis*, legit. T.De Stefani, ex coll. V.E.Orlando (CMT); Monte Pellegrino, 12.V.1974/XI.1970, ex coll. V.E.Orlando (CMT); Monte Pellegrino, 15.IV.1972, ex coll. V.E.Orlando (CMT); mountains nearby Palermo, “Vuc-ciria” market, X.1974, ex coll. V.E.Orlando (CMT); Monte Pecoraro, 27.03.1976, leg. S. Palazzi, ex coll. V.E.Orlando (CMT); Monte Pellegrino, 23.IX.1978, leg. S. Palazzi & V.E.Orlando, ex coll. V.E.Orlando (CMT); Monte Cuccio, 400 m slm, 15.IV.1979, ex coll. V.E.Orlando (CMT); Terrasini, Costa della Palazzola, 20.IX.1981, ex coll. V.E.Orlando (CMT); Carini, Monte Pecoraro, 16.IV.1983, leg. I.Sparacio, ex coll. V.E.Orlando (CMT); Monte Pellegrino, leg./coll. Alzona (MM); Capaci, coll. Alzona (PA); Monte Pellegrino, April 1918, leg./coll. Giorgi (MM); Carini, coll. Giorgi (MM); nearby Palermo, coll. Giorgi (MM); Monte Cuccio, coll. Giorgi (MM); Monte Pellegrino, March 1954, leg./coll. C. Sacchi (MM); Monte Gallo, coll. Priolo (MM); nearby Palermo, coll. Priolo (MM); Sferracavallo, La Montagnola, 23.III.1978 (CS); Monte Pellegrino, near Valdesi, 23.IV.1978, 15.II.2007 (CS); Monte Cuccio, Bellolampo, 600 m slm, 20.X.1978 (CS); Sferracavallo, La Fossa, 31.I.1981 (CS); Sferracavallo, Raffo Rosso, 15.VIII.1983 (CS); Carini, Monte Pecoraro, 16.IV.1983 (CS); Capaci loc. Zarcati, 6.IV.1985 (CS); Monte Pellegrino, Sanctuary of Saint Rosalia, 14.X.1989, 21.IV.2007, 22.IV.2008 (CS); Palermo: Sferracavallo, Capo Gallo, m 200, 05.XI.2006, (CL); Palermo: Pietrazzi, Grotta della Molara, m 90, 04.III.2007, (CL); Palermo: Monte Pellegrino, Pizzo Ferro, m 500, 11.VIII.2007, (CL); Palermo: Monte Pellegrino, Pizzo Rufiuliata, m 350, 14.VIII.2007, (CL); Palermo: Carini, Monte Columbrina, Grotta Zà Minica, m 130, 18.VIII.2008, (CL); Palermo: Cinisi, Monte Pecoraro, Pizzo Corvo, m 900, 18.VIII.2008, (CL); Sferracavallo, Capo Gallo, m 180, 05.XI.2006 (CG); Palermo: località Pietrazzi, m 90, 04.III.2007 (CG); Monte Pellegrino, (near the statue of Saint Rosalia) 10.XI.2006 (CG); Monte Pellegrino, Pizzo Ferro, m 500, 09.VIII.2007 (CG); Sferracavallo, La Montagnola, 10.VIII.1996, 5.X.2008 (CR); Palermo: Pietrazzi, Grotta della Molara, m 90, 10.XII.1994, 26.VIII.2001, 28.VIII.2005, 04.III.2007, 2.XII.2008 (CR); Monte Pellegrino, (near the statue of Saint Rosalia) 16.IX.1995, 17.XII.2005, 3.X.2007 (CR); Pizzo Ferro, m 500, 16.VIII.1999 (CR); Pizzo Rufiuliata, Monte Pellegrino, m 350, 3.X.2007 (CR); Pizzo di Mezzo, Monte Pellegrino, m 400, 3.X.2007 (CR); Grotta Addaura Caprara, Monte Pellegrino, m 210, 27.VIII.2001 (CR); La Croce, Monte Pellegrino, m 460 16.IX.1995 (CR); Monte Pellegrino peak, m 550 16.IX.1995 (CR); Mont.la S. Rosalia, m 130 16.IX.1995 (CR); C.da Costamazzone, Capo Gallo, m 150, 12.IV.2000 (CR);

### *Ercetella cephalaeeditana* (Giannuzzi-Savelli, Oliva & Sparacio, 1986)

Records: Sicily, Palermo: Cefalù, La Rocca coll. Priolo (MM); Cefalù, La Rocca, 20.XII.1980, leg. I.Sparacio, ex

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coll.V.E.Orlando (CMT); Cefalù, La Rocca, 20.XII.1980 (CS); idem, 12.II.2007 (CS); idem, 12.05.2007 (CS); Palermo: Cefalù, La Rocca, m 200, 19.X.2003, (CL); idem, m 70, 26.XII.2003, (CL); idem, m 70, 27.XII.2003, (CL); idem, m 80, 18.I.2004, (CL); idem, m 50, 14.XI.2004, (CL); idem, m 50, 28.XI.2004, (CL); idem, m 80, 29.X.2005, (CL); idem, m 80, 07.I.2006, (CL); idem, m 80, 21.X.2006, (CL); idem, m 160, VIII.2007, (CL); idem, m 60, 20.IV.2008, (CL); Palermo: Cefalù, La Rocca, m 200, southern slope, 13.VII.2003 (CG); idem, m 250, 16.X.2004 (CG); idem, m 200, northen slope, 16.IX.2006 (CG); idem, m 220, eastern slope, 18.IX.2006 (CG); idem, m 80, western slope, 20.IX.2006 (CG); idem, m 50, northen slope, 14.IX.2008 (CG); Palermo: Cefalù, La Rocca 1.V.1996 (CR).

*Erectella insolida* (Monterosato, 1892)

Records: Sicily, Trapani: Trapani, coll. Priolo (MM); Capo S. Vito, 1 ex (MSNG); S. Vito lo Capo, 9.XI.1975, leg. F.P.Romano, ex coll.V.E.Orlando (CMT); Monte Cofano, 1976, leg. S. Palazzi, ex coll.V.E.Orlando (CMT); Capo S. Vito, 2.VII.1978, leg. S.Palazzi, ex coll.V.E.Orlando (CMT); Favignana, Cala Galera, 25.VIII.1979, fossils, leg. F. & S. Palazzi, ex coll.V.E.Orlando (CMT); Levanzo island, 26.IV.1975, quaternary fossils, ex coll.V.E.Orlando (CMT); Monte Cofano (TP), 4.XI.1985 (CS); Scopello (TP), la Tonnara, July 1988 (CS); San Vito lo Capo (TP), loc. Belvedere, 1.XII.1992 (CS); Trapani: Custonaci, Monte Cofano, m 250, III.2005, (CL); Trapani: San Vito lo Capo, Monte Speziale, Cozzo Castelluzzo, m 368, 21.I.2007, (CL); Trapani: San Vito lo Capo, Cala Mancina, m 10, 21.I.2007, (CL); Trapani: Custonaci, Torre di Cofano, m 20, 21.I.2007, (CL); Trapani: Custonaci, Monte San Giovanni, m 290, 13.I.2008, (CL); Trapani: San Vito lo Capo, Cala Mancina, m 50, 13.I.2008, (CL); Trapani: Custonaci, Monte Cofano, Tonnara di Cofano, m 70, 13.I.2008, (CL); Trapani: Valderice, Rocca Giglio, m 150, III.2008, legit L. Barraco (CL); San Vito lo Capo, Monte Speziale, Cozzo Castelluzzo, m 360, 21.I.2007 (CG); San Vito lo Capo, Cala Mancina, m 20, 21.I.2007 (CG); Custonaci, Monte San Giovanni, m 290, 13.I.2008 (CG); San Vito lo Capo, Cala Mancina, m 50, 13.I.2008 (CG); Custonaci, Monte Cofano, Tonnara di Cofano, m 70, 13.I.2008 (CG); Trapani: San Vito lo Capo, Monte Passo del Lupo m 450 22.IX.1996 (CR); C.da Isolidda, San Vito lo Capo, m 65 22.IX.1996 (CR); Cala Mancina, m 10, 22.IX.1996, 21.I.2007, (CMT) Custonaci, Torre di Cofano, m 20, 22.IX.1996, 5.VI.1997, 21.I.2007, (CR), Monte Speziale, Cozzo Castelluzzo, m 360, 22.IX.1996, 21.I.2007 (CR), Monte Monaco, San Vito lo Capo, m 70, 19.IV.2009 (CR), Scopello, loc.tà Fraginesi, m 150, 22.X.2008, legit I. Sparacio (CS).