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SHELL AND BODY STRUCTURE OF THE PLESIOMORPHIC PULMONATE MARINE LIMPET *SIPHONARIA PECTINATA* (LINNAEUS, 1758) FROM PORTUGAL (GASTROPODA: HETEROBRANCHIA: SIPHONARIIDAE)

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ABSTRACT: The shell and body structure of the plesiomorphic pulmonate false-limpet *Siphonaria pectinata* (Linnaeus) from the south coast of Portugal is presented in detail. All the systems are described and discussed, in order to create a phenotypic database, since the siphonariid relationships are controversial, and most of the recent attempts at their resolution have been molecular. Most siphonariids show several morphological peculiarities (e.g., flap-like head without tentacles, interrupted shell muscle, anterior portion of genitalia surrounding shell muscle) compared to the remaining pulmonates or even to heterobranchs. On the other hand, the odontophore structure, and the genital system, including the penis retractor muscle connected with the shell muscle, place them close to higher pulmonates. The siphonariids present a new mode of limpetisation, with the visceral structures placed inside the haemocoel instead of being located below the shell apex.

KEY WORDS: morphology, anatomy, somatic evolution, limpetisation, systematics, comparative biology

INTRODUCTION

The siphonariids are marine pulmonate limpets with a combination of ancestral and derived characters (HUBENDICK 1946). They are herbivores (YONGE 1952), mainly inhabiting rocky shores of tropical and warm latitudes (DAYRAT et al. 2014). The detailed knowledge of their structure is crucial for the understanding of the evolution of Heterobranchia, the most heterogeneous group of gastropods, and the link between the opisthobranchs and the pulmonates.

The pulmonates are adapted to air breathing, using a pallial cavity modified into a "lung", connected to the exterior by an orifice called pneumostome and closed by a sphincter. Though most of them are entirely terrestrial, some primitive taxa are marine, and depend on the pelagic environment to complete their development. Within the family Siphonariidae, most taxa have planktotrophic veliger larvae (JABLONSKI & LUTZ 1983, GRAHAME & BRANCH 1985), although some species undergo intra-capsular larval stage – a mode of development sometimes called direct development (PAL & HODGSON 2002), a term that has been used for the siphonariids (CHAMBERS & MCQUAID 1994a, b, PAL & HODGSON 2003, 2005). Planktonic larval development is regarded as plesiomorphic within the family, and used as a systematic criterion to resolve their higher taxonomy and evolutionary relationships. The prevalence of this developmental mode supports the hypothesis that the siphonariids have a marine ancestry, rather than being derived from a terrestrial group which re-invaded the marine environment (HODGSON 1999 and references therein).

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The genus and species concepts within the Siphonariidae have been challenged as a result of the use of non-conchological, i.e., phenotypic and molecular, data (e.g., DAYRAT et al. 2011, 2014, WHITE & DAYRAT 2012). Forms of different shell structure have been proved to be conspecific variations, while conchologically similar forms have turned out to be different endemic species (e.g., HUBENDICK 1955). The anatomical information available is too tenuous to provide a strong support for a classifiction (e.g., HUBENDICK 1946); it leads to raising not easily recognisable or non-monophyletic taxa. There is a need for description of internal anatomical characters that could help delineating siphonariid species (DAYRAT et al. 2014). Currently, the research aiming at solution of the above mentioned problems focuses on molecular aspects rather than on anatomy. The relationships of the Siphonariidae, and of the Basommatophora in general, with the other Pulmonata, and their phylogenetic status within the clade Euthyneura, are also debateable; some molecular approaches indicate a closer relationship with the Sacoglossa opisthobranchs (GRANDE et al. 2004, 2008, DAYRAT et al. 2011, DINAPOLI et al. 2011, JENSEN 2011).

Siphonaria pectinata (Linnaeus, 1758) has long been regarded as a north amphi-Atlantic species, occurring along the Atlantic coasts of Europe and North America (VOSS 1959). However, molecular analysis of samples from both sides of the North Atlantic suggests that these regions may hold two different species (KAWAUCHI & GIRIBET 2011). Nevertheless, the species in its original sense has one of the broadest distributions in the genus, inhabiting the Mediterranean and the Eastern Atlantic Ocean from Portugal to Cameroon, with a gap at the Gulf of Guine (WHITE et al. 2011). There are some indications that the range of *S. pectinata* in the Northeast Atlantic might be extending, but it is common only as far north as the Iberian Peninsula (S. J. HAWKINS, pers. comm.).

Despite its importance, the morphological and anatomical knowledge of the siphonariids is relatively poor in view of their diversity and their nearly world-wide distribution. According to current estimates the genus *Siphonaria* comprises from 41 to over 100 recognised species (DAYRAT et al. 2014, WoRMS 2017). They are crucial for the understanding of the relationships of Pulmonata with the remaining higher heterobranchs. Inferring phylogeny within the pulmonates based on (mainly external) morphological characters is problematic due to the high incidence of homoplasy (WHITE et al. 2011), and a more complete, holistic anatomical description can certainly throw new light on the question. Besides, despite their remarkable resilience to habitat disturbance (HODGSON 1999), the siphonariids are still much less studied ecologically compared to other limpets (e.g., patellogastropods), even in Europe, where *S. pectinata* is the only species present. Therefore, information on its anatomy may be crucial for understanding of several aspects of the species' ecology and its possible adaptations to environmental changes. The main objective of this paper is to describe and compare the morpho-anatomical features of a common and widespread siphonariid species, in order to provide a baseline for complementary studies on comparative biology, and possibly on phylogeny and ecology.

MATERIAL AND METHODS

The samples included large animals (shell length ~30–40 mm) collected on the rocky shores of the region of Sines, Portugal (Praia da Oliveirinha: 37°53'19.21"N, 08°47'49.29"W and adjacent areas), preserved in 70% ethanol, and deposited in the malacological collection of the Museu de Zoologia da Universidade de São Paulo (MZSP). The specimens were extracted from their shells and dissected using the standard techniques, under a stereo-microscope, with the specimen immersed in ethanol. Digital photos of most dissection steps were taken; camera lucida drawings were made; the drawings and description are based on all the examined specimens (N= 20). The radula was examined in scanning electron microscope (SEM) in the Laboratório de Microscopia Eletrônica of MZSP. Additionally, to illustrate some details of the external morphology, digital photos of two live specimens were taken in the field.

The following abbreviations are used in the figures: aa – anterior aorta; ac – albumen chamber; ad – albumen gland duct; ag – albumen gland; an – anus; ap – terminal genital orifice; as – visceral (abdominal)-subintestinal ganglion; au – auricle; bc – bursa copulatrix; bd – bursa duct; bg – buccal ganglion; bm – buccal mass; br – subradular cartilage; cc – cerebral commissure; ce – cerebral ganglion; cv – ctenidial (efferent) vein; dd – duct of digestive gland; df – dorsal fold of buccal cavity; dg – digestive gland; ec – oesophageal crop; ef – oesophageal folds; es – oesophagus; ey – eye; fl – flap below pneumostome; fp – faecal pellets; ft – foot; gi – secondary gill; go – gonad; gp – genital pore; hd – hermaphrodite duct; he – head; in – intestine; im – isolated portion of shell muscle; jw – jaw; ki – kidney; m1–m8 – external and internal odontophore muscles; ma – buccal dilator muscle; mb – mantle edge; mj – peribuccal muscles; mn – mantle connection with nuchal re-

gion; mo – mouth; ms – gastric muscle; ne – nephropore; nr – nerve ring; nu – nuchal connection of mantle; nv – nerve; oc – odontophore cartilage; od – odontophore; pc – pericardium; pd – penis gland duct; pe – penis-like copulatory organ; pf – ventral pair of pulmonary folds; ph – buccal sphincter; pg – penis gland; pm – penial/copulatory organ muscle; pn – pneumostome; pp – pleuro-pedal ganglion; pt – prostate; pu – pulmonary (mantle) cavity; ra – radula; rn – radular nucleus; rs – radular sac; rt – rectum; sa – salivary gland aperture; sd – salivary duct; sg – salivary gland; sh – shell; sm – shell muscle; so – spermoviduct; sr – seminal receptacle; st – stomach; ta – terminal genital atrium; tg – integument; ur – urethra; ve – ventricle; vm – visceral mass; zr – parieto-supra-intestinal ganglion.

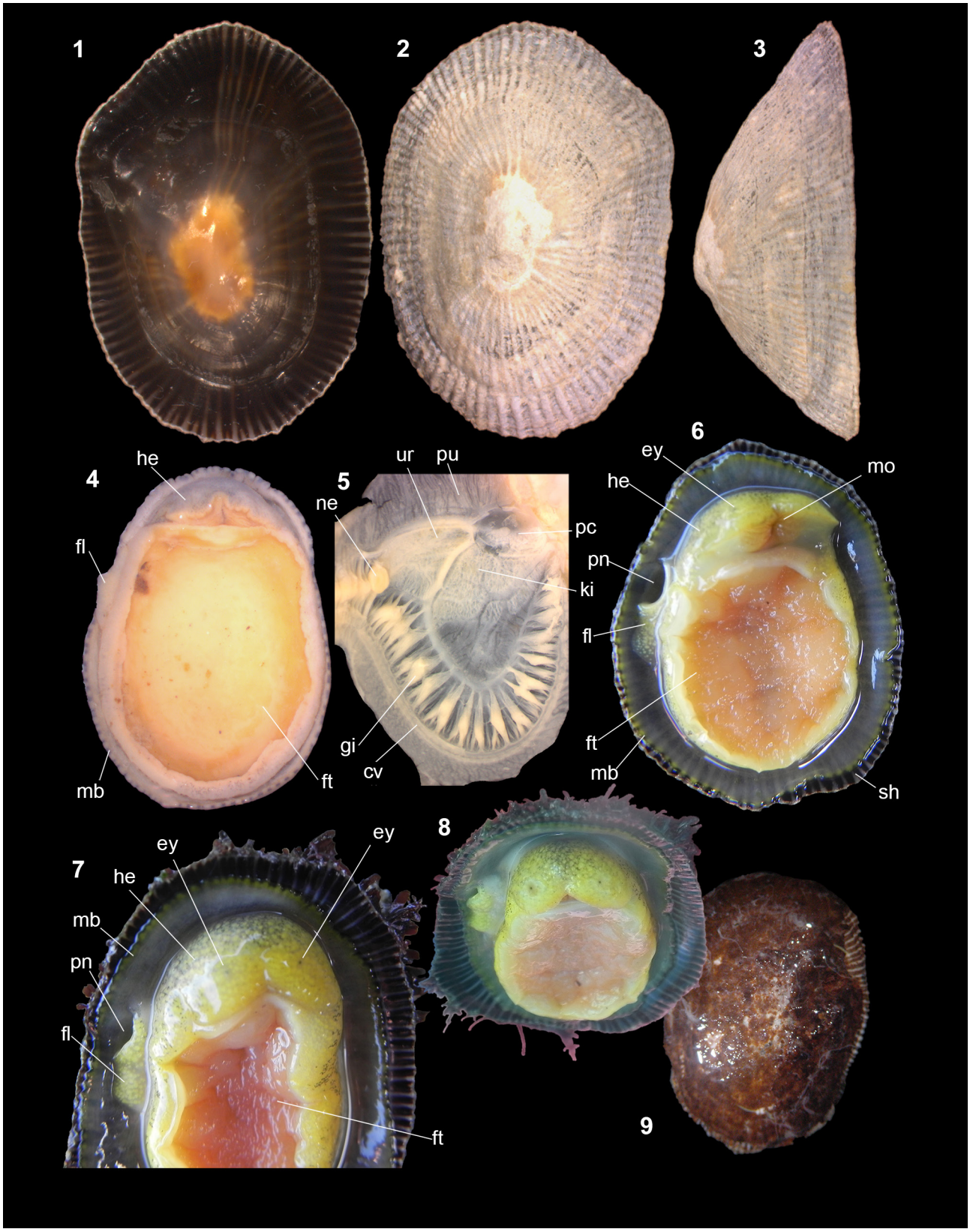
RESULTS

SIPHONARIA PECTINATA (LINNAEUS, 1758) (Figs 1–36)

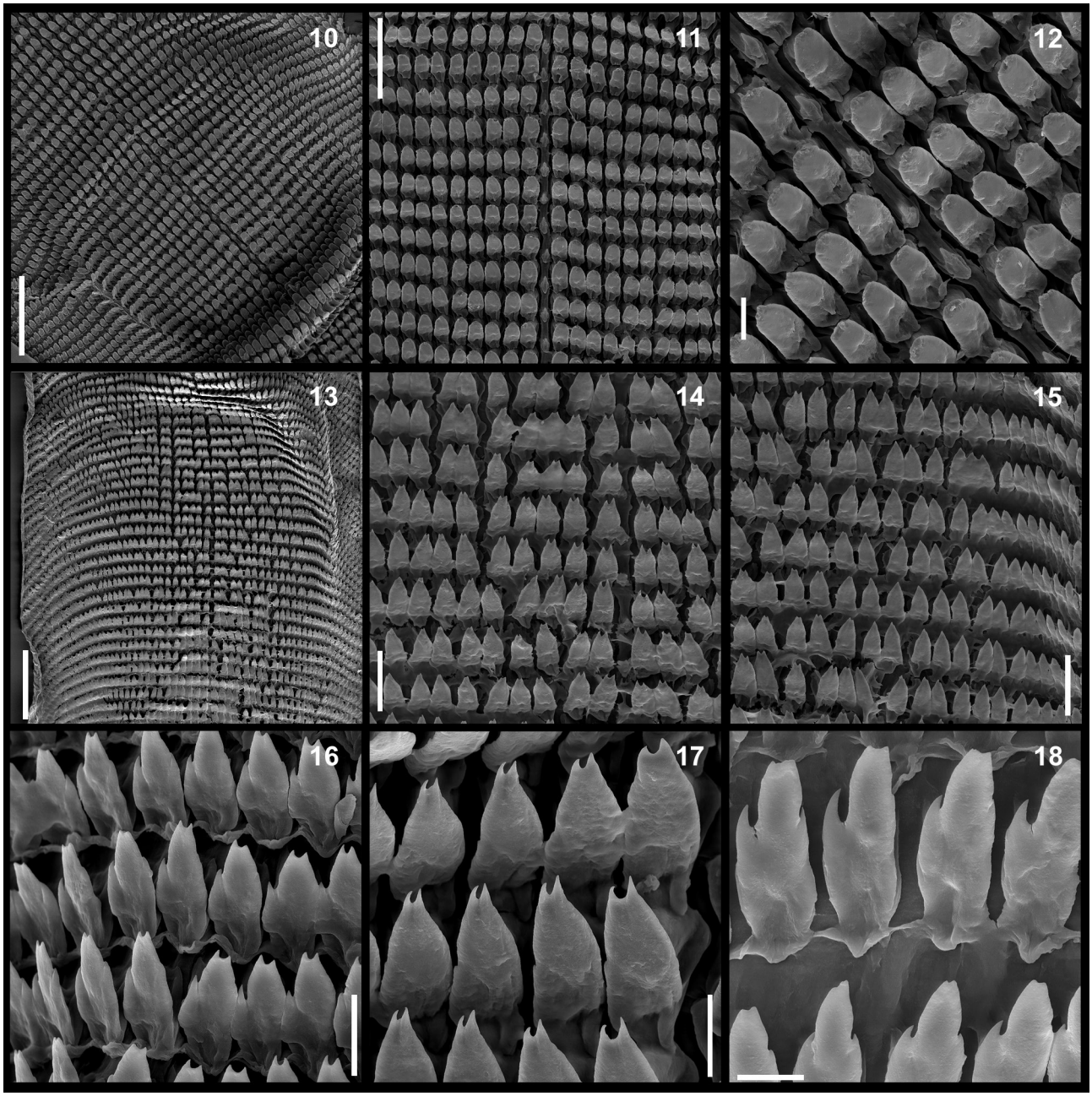
Shell (Figs 1–3, 9). Limpet-shaped, up to 40 mm. Outline elliptical, width ~70% of length; height ~40% of length. Colour brownish grey to beige. Sculpture mosaic of ~100 narrow radial ribs, with rounded profile, gradually and uniformly increasing towards edge; interspaces slightly narrower than threads; concentric undulations and growth lines. Apex sub-central, slightly displaced to left and posterior (Figs 2–3). Inner surface dark brown, glossy, with narrow beige radial bands corresponding to external threads; brownish-beige spot in apical region occupying ~15% of surface. Edges slightly irregular; wide radial groove in middle of right-anterior quadrant (Figs 1–2); this groove marks a gap in the horseshoe-shaped muscle scars, located half way between apex and edge (Fig. 1). Apex eroded to various degree in all the studied specimens, from ~10% (Fig. 2) to almost entire dorsal surface (Fig. 9), as a result of environmental influence. For other details see Voss (1959).

Head-foot (Figs 4, 6–8, 20, 22): of limpet shape, dorso-ventrally much flattened. Head uniformly yellow to cream-coloured with mosaic of greenish/brown and creamy spots on its dorsal surface (Figs 7–8). Head in the form of thick anterior flap, anterior edge widely bi-lobed, medially grooved, each lobe moving relatively independent from the other; pair of minute eyes (Figs 6, 8: ey) immersed in integument, each one located half way between median line and head lateral edge. No tentacles or ommatophores. Ventral head surface transversely folded, with mouth (Fig. 6: mo) in middle. Foot (ft) wide and ample, flattened, occupying most of shell ventral surface; colour pale orange to grey; no appendages, except for wide flap below pneumostome (Fig. 4: fl), with ~1/5 of head-foot length, located at the level of shell groove. Shell muscle thick, horseshoe-shaped, surrounding almost entire shell edge, close to mantle edge (Figs 20, 22: sm), except for anterior region (nuchal cavity) and right-anterior region (pneumostome) (both ~1/3 of shell width in length); left branch entire, right branch divided in anterior region, with anterior part of elliptical cross-section (Figs 19–20, 22; im), and posterior region half as long as right branch. Dorsal surface of foot relatively plain, forming pallial floor (Fig. 20: pu). Pneumostome ventral flap (Figs 4, 6–8: fl) described below. Haemocoel occupying ~6% of head-foot volume, restricted by shell muscles to central regions of head-foot (Fig. 22) (for more details see below).

Mantle organs (Figs 5, 19–21): mantle edge relatively thin, pigmented with a row of dark-brown spots parallel to edge (Figs 6–7, 19: mb). Pneumostome lacking sphincter or distinct adjacent muscles; protected by ventral flap (Figs 4, 6, 7: fl, 8), with ~¼ of shell length, closing or opening pneumostome. Pneumostome about 1/8 of shell length (Figs 19–22: pn), with anus in middle region (Figs 20, 22: an). "Lung" occupying ~80% of shell area (Fig 20: pu), slightly elliptical except for small incisions at pericardial area (Figs 20, 21: pc). Pulmonary vessels inconspicuous, well-developed secondary gill occupying ~1/3 of pallial roof (Figs 5, 19, 21: gi); gill filaments somewhat irregular, tall filaments alternating with shorter ones (Fig. 5); close to pneumostome anterior-right gill third straight, transverse, bearing pairs of relatively uniform, thick fold-like filaments; middle and posterior-left gill thirds curved (concavity anterior), with simple filaments, gradually diminishing to the left. Ctenidial vein (Figs 5, 20, 21, 23: cv) well-developed, anterior third running at mid-gill level, in middle and posterior thirds flanking posterior gill edge up to posterior-left corner of pericardium (Fig. 21). Pulmonary vein practically absent, except for pair of vessels flanking urethra (Fig. 21: ur), one coming from anterior edge of gill, another from middle and posterior edge of gill. Ventral floor of pallial cavity mostly flat, with three folds roughly following gill outline, roughly equidistant from pericardium and posterior end of cavity; a pair of folds running close to gill edges (Fig. 20: pf), with anterior fold slightly thicker, ending adjacent to anus, posterior fold flanking posterior edge of cavity, becoming fainter in posterior region of pneumostome; third fold bordering posterior margin of kidney ventral lobe (Fig. 20: ki); between consecutive folds wide furrow leading to pneumostome. Kidney pale cream,



Figs 1–9. *Siphonaria pectinata*. Shell morphology and gross anatomy: 1 – shell of specimen MZSP 115172-2, ventral view (L 36.3 mm); 2 – same, dorsal view; 3 – same, right lateral view; 4 – whole ventral view of specimen MZSP 115169-2 extracted from shell (L 21.6 mm); 5 – detail of pallial cavity, ventral-inner view, specimen MZSP 115172-1 (L 12.5 mm); 6 – live specimen, ventral view (L ~35.0 mm); 7 – another live specimen, ventral view, with head bent inwards to show face (L ~35.0 mm); 8 – same specimen, antero-ventral view; 9 – shell of MZSP 115170-1, dorsal view (L 32.3 mm)

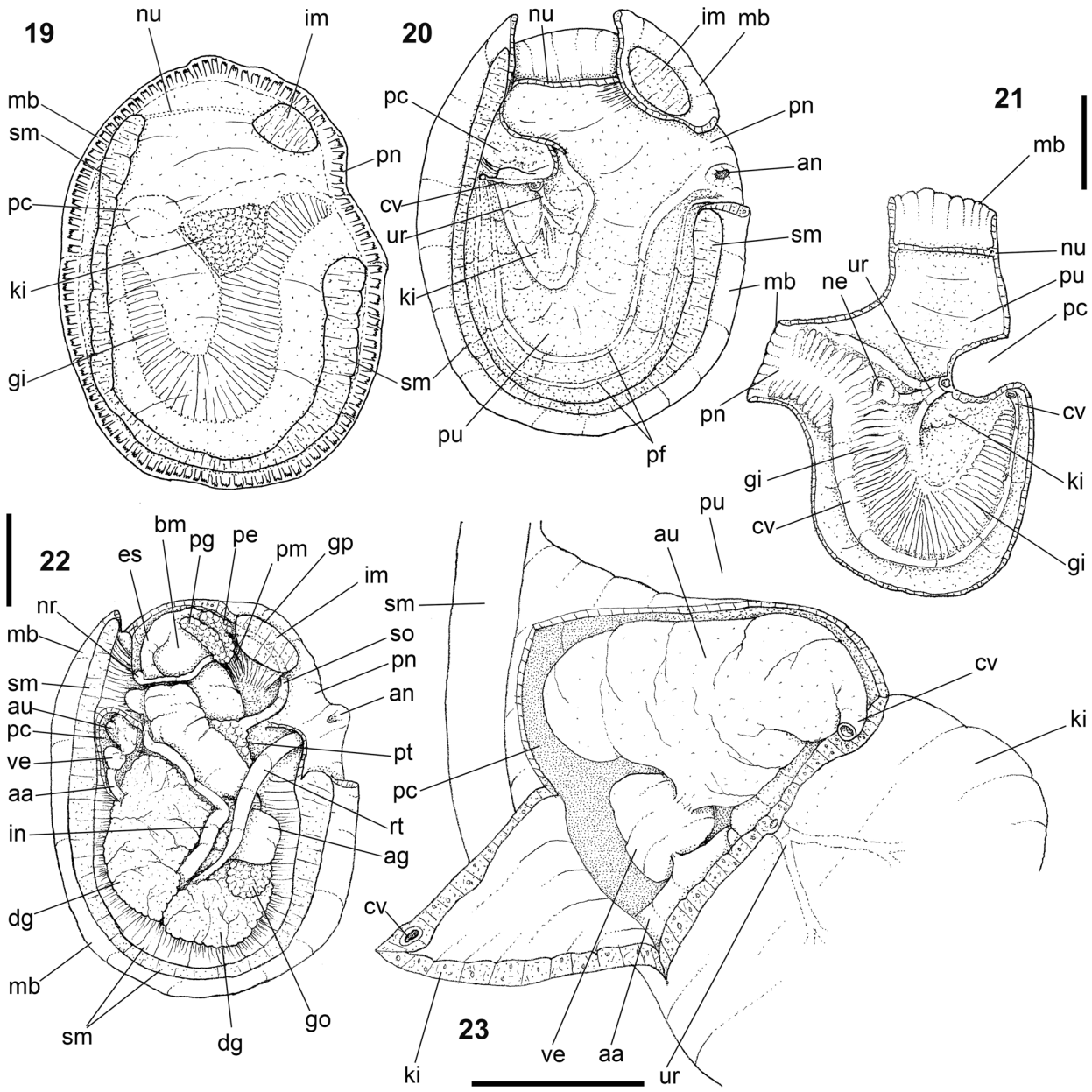


Figs 10–18. *Siphonaria pectinata*. Radulae in SEM (5 specimens): 10 – middle length, scale bar 200 μm; 11 – detail of central region, bar 100 μm; 12 – same, higher magnification, bar 20 μm; 13 – whole view, middle length, bar 200 μm; 14 – detail of central region (rachidian slightly to right), bar 50 μm; 15 – detail of lateral region, bar 50 μm; 16 – detail of middle of lateral region, bar 20 μm; 17 – detail of central border of lateral teeth (rachidian on right edge), bar 20 μm; 18 – same as Fig. 16, higher magnification, bar 10 μm

located slightly centrally-left, occupying ~25% of cavity area (for details see below). Rectum (Fig. 22: rt) not emerging in cavity; urethra (Figs 5, 20–21: ur) narrow, running form left to right in middle region of pallial cavity roof (for details see below).

Visceral mass (Figs 20, 22): practically entirely shifted to head-foot haemocoel. Buccal mass (bm) inside head, occupying ~15% of haemocoel volume. Midgut and hindgut occupying central areas, ~40% of haemocoel volume (Fig. 24). Digestive gland (dg) surrounding externally most middle and posterior structures, occupying ~20% of haemocoel volume. Genital system lying on the right side of haemocoel, though adjacent to digestive structures, occupying ~25% of haemocoel volume. Gonad (go) cream-coloured, immersed in middle-right side of digestive gland. For more details see below.

Circulatory and excretory systems (Figs 21, 23): Pericardium about as long as wide, located between anterior and middle thirds of left side (Figs 19–20: pc); occupying ~5% of total dorsal surface. Auricle located anterior and right of pericardial area



Figs 19–23. *Siphonaria pectinata*. Anatomy: 19 – whole dorsal view, shell removed; 20 – same, mantle and pallial cavity roof removed; 21 – pallial cavity roof removed from previous Figure, ventral-inner view; 22 – haemocoel, dorsal view, dorsal cover of integument (pallial floor) removed, structures seen as *in situ*; 23 – renopericardial region, dorsal view as in Fig. 20, anterior region of kidney sectioned and deflected to left. Scale bars 5 mm, except 23 – 2 mm

(Fig. 22: au), as continuation of pulmonary/ctenidial vein (Fig. 23: cv), ca. twice ventricle's size; walls thin, translucent, expanding as blind-sac anteriorly (Fig. 23: au). Ventricle thick-walled, connected to auricle on right side, connection with aorta trunk posterior (Fig. 23: ve). Anterior aorta ~4 times broader than posterior aorta, directed initially towards left, in short distance bent posteriorly, expanding near haemocoelic organs (Fig. 22: aa). Kidney simple, entirely solid (Figs 19–21: ki), with two lobes separated from each other by pulmonary cavity; both lobes solid and flattened; dorsal lobe lying in pallial roof right of pericardium, as large as pericardium (Figs 5, 19, 21: ki); ventral lobe located just ventral to dorsal lobe, elliptical (antero-posterior axis longer) in pallial floor, flanked by anterior pallial fold (Fig. 20: ki); middle region with branched urethra, converging to middle region of anterior edge. Urethra crossing from ventral to dorsal renal lobes on their anterior edge, running transversely to right for a distance equivalent to ¼ of pallial cavity width (Figs 5, 21: ur). Nephropore (or nephrostome) small, preceded by elevated papilla, with walls thick and somewhat hollow as urinary reservoir, bent ventrally and to pneumostome aperture (Figs 5, 21: ne). No detectable urethra or urinary gutter.

Digestive system (Figs 22, 24–31, 33). Oral tube short and muscular, developed as sphincter (Figs 26, 28: ph); nerve ring located posterior to buccal mass (Figs 22, 27: nr). Buccal mass spherical, occupying ~1/8 volume of haemocoel (Fig. 22: bm). Ventral region of buccal mass completely filled by odontophore; dorsal region mostly hollow, forming oral cavity (Fig. 26; showed opened in Fig. 25). Jaws simple, transversely folded; ~5 times wider than long, located in dorsal region of oral cavity, just posterior to sphincter (Fig. 25: jw). Pair of well-developed dorsal folds of buccal mass (Fig. 25: df) occupying most of dorsal surface of oral cavity; both flanking posterior edge of jaw; space between both dorsal folds relatively narrow but deep; each fold with narrow notch of oesophageal origin, corresponding to orifices of salivary ducts (Fig. 25: sa). Odontophore and peri-oral muscles (Figs 25–31):

mj – pair of jaw and peribuccal muscles (Figs 25, 30) very thick, originating on both sides of odontophore cartilages (oc) (Fig. 31), extending along entire buccal cavity towards dorsal side as thick layer of circular fibres (Fig. 25); ma, pair of buccal abductor muscles (Figs 27–28), originating in antero-lateral surface of haemocoel, running anteriorly along lateral surface of buccal mass, inserting in both lateral regions of mouth;

m1v – pair of ventral median retractor muscles of mouth (Figs 26, 28), originating in ventral surface of haemocoel at the level just posterior to buccal mass, running anteriorly close to median line, inserting in ventral region of mouth;

m2 – pair of buccal mass retractor muscles or radular muscles absent;

m3 – single transverse thin muscle covering anterior region of oral cavity (Figs 26, 29), as part of oral cavity walls anterior to mj, inserting on m4;

m4 – main pair of dorsal tensor muscles of radula (Figs 29, 31), very thick, originating in lateral and latero-dorsal regions of odontophore cartilages (oc), surrounding these cartilages anteriorly and medially; inserting in radular sac (rs) along its region immediately posterior to buccal cavity (Fig. 29);

m5 – pair of thin and narrow auxiliary dorsal tensor muscles (Figs 29–31), originating in outer-posterior surface of m4, running medially and anteriorly, inserting in radular sac just ventral to m4 insertion (Fig. 29);

m6 – horizontal muscle (Figs 29–31) thin and narrow, located between both odontophore cartilages posterior to their fusion, along ~2/3 their length;

m7 – pair of narrow and thin muscles (Figs 29–31), originating in medial edge of m11a pair, separating from them at their middle level, penetrating though m6 in its anterior third (Figs 30, 31) crossing from ventral to dorsal region of cartilages, running in ventral surface inside radular sac towards posterior (Fig. 29), inserting along posterior region of radular sac;

m10 – two pairs of ventral protractor muscles of buccal mass (Figs 26, 28), lateral part originating in ventral region of mouth and lateral part originating from adjacent surface of haemocoel (Fig. 28), running posteriorly close to median line, inserting in ventro-posterior region of odontophore, close to region of radular sac;

m10d – pair of small auxiliary protractor muscles of buccal mass (Fig. 26), originating in lateral side of mouth, running posteriorly for short distance, inserting in lateral surface of buccal mass;

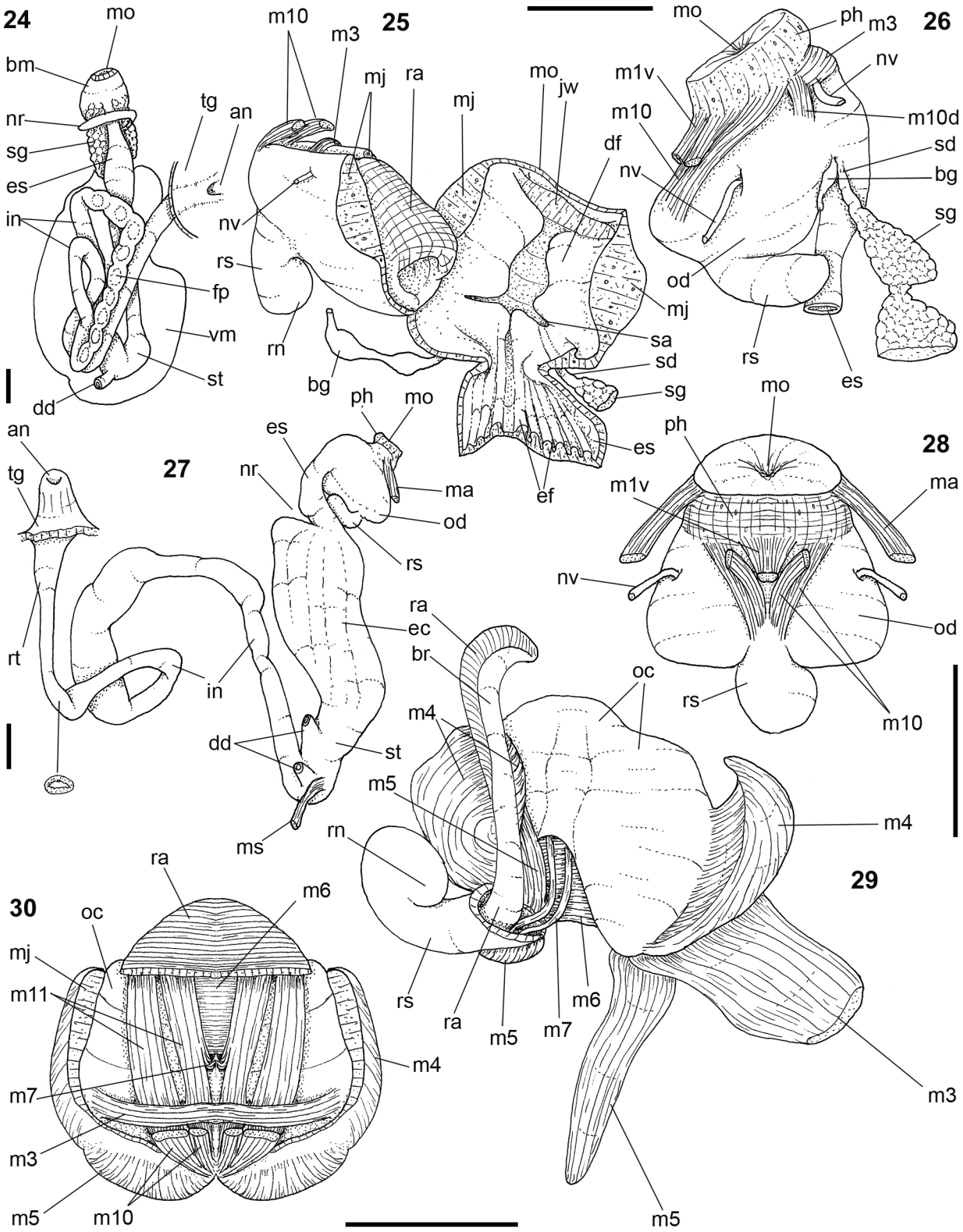
m11 – pair of narrow ventral tensor muscles of radula (Figs 30–31), originating in postero-ventral surface of odontophore cartilages, running anteriorly, inserting in ventral end of subradular cartilage (br);

m11a – median pair of auxiliary ventral tensor muscles of radula, slightly broader than m11 pair, located more medially, of similar origin and insertion as m11 (except being more medial), medial part of m11a originating from posterior region of m6 (Fig. 31).

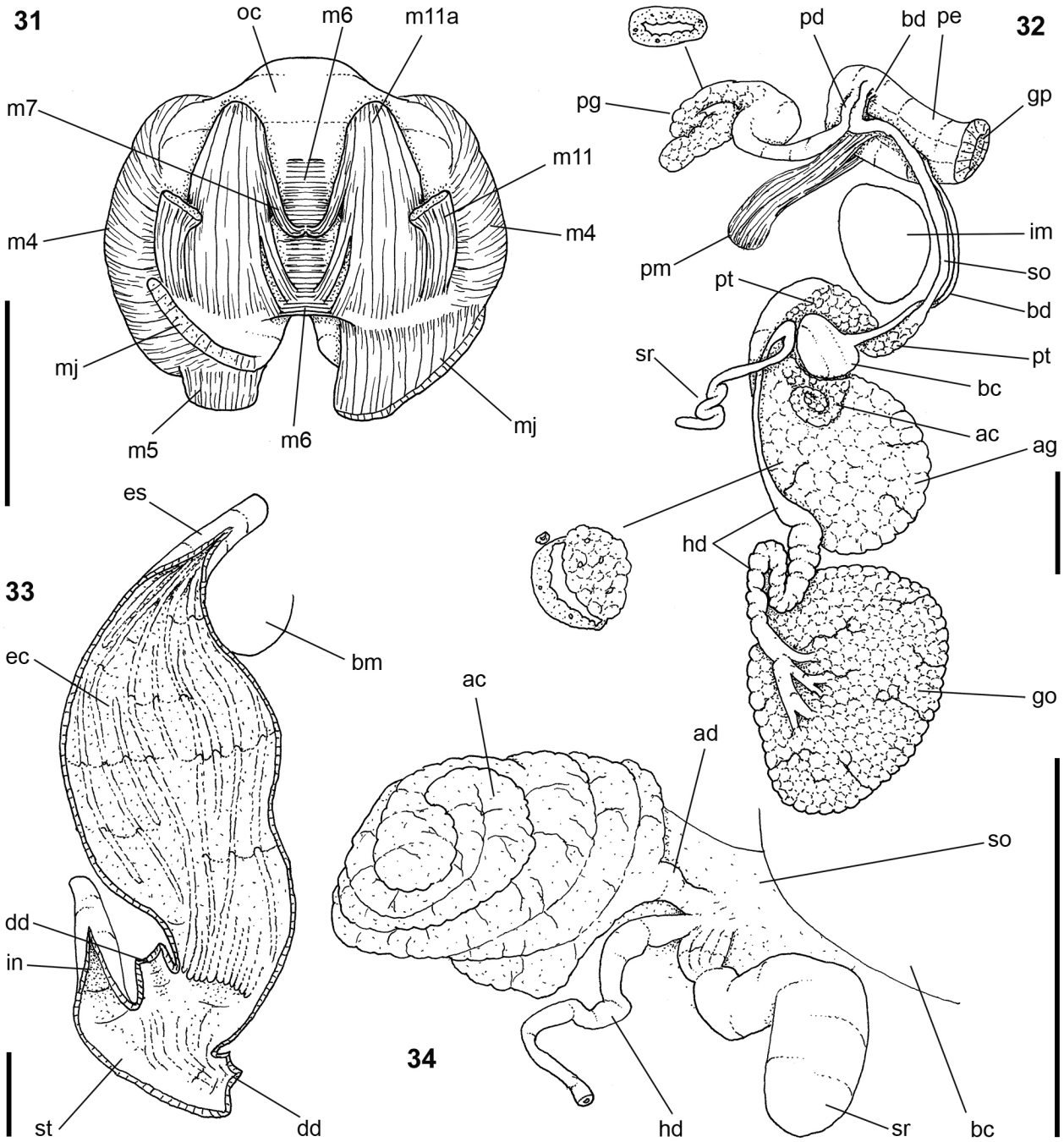
Odontophore non-muscular structures (Figs 29–31):

oc – odontophore cartilages, flattened, about half fused with each other along median line (Fig. 29), anterior end roughly rounded, remaining with elliptical shape, ~twice longer than wide, posterior end rounded; br, subradular cartilage, with expanding region in buccal cavity protecting subradular membrane (Fig. 29).

Radula (Figs 10–18), slightly longer than odontophore (Fig. 29), with rachidian teeth, plus ~35 (34–37, N=5) pairs of lateral teeth; no clear boundary with marginal teeth. Each radular row relatively straight in middle 2/3, marginal region slightly curved backwards (Figs 10, 13, 15). Rachidian tooth (Figs 11–12, 14) small, ~0.2% of radular width, ~3 times longer than wide; base long and rectangular, cutting edge hook-like, elevating from posterior end, directed forwards; sharp pointed terminal cusp restricted, in dorsal view, to posterior third of tooth; pair of small but broad expansions at base of terminal cusp (Fig. 12). Lateral teeth similar to rachidian tooth (Figs 10–11, 13), but twice as wide and cutting edge twice as long; form slightly asymmetrical, weakly bent internally. Cutting edge tip of lateral teeth varying from blunt (Figs 11–12) to bifid (Figs 14–18); in bifid cases, both terminal cusps similar, small, equidistant, turned forwards (Figs 16–17). Cutting edge of lateral teeth triangular, ~twice longer than wide; no basal cusp, but with longitudinal reinforcement as middle fold (Fig. 10, inferior region); 11–12 more central pairs of teeth relatively uniformly shaped; re-



Figs 24–30. *Siphonaria pectinata*. Anatomy: 24 – digestive tubes as *in situ*, dorsal view, topology of some structures also shown; 25 – buccal mass, dorsal view, inner surface exposed, odontophore and buccal ganglia (bg) sectioned and deflected to left, oesophagus opened longitudinally; 26 – buccal mass, left lateral view; 27 – expanded digestive tubes, dorsal view, transverse section at indicated level of intestine also shown; 28 – buccal mass, ventral view; 29 – odontophore, dorsal view, superficial muscles and membrane removed, both cartilages deflected, radula deflected to left, right muscles sectioned at their insertions and deflected to right; 30 – odontophore, ventral view, superficial membrane and terminal region of radula removed. Scale bars 2 mm



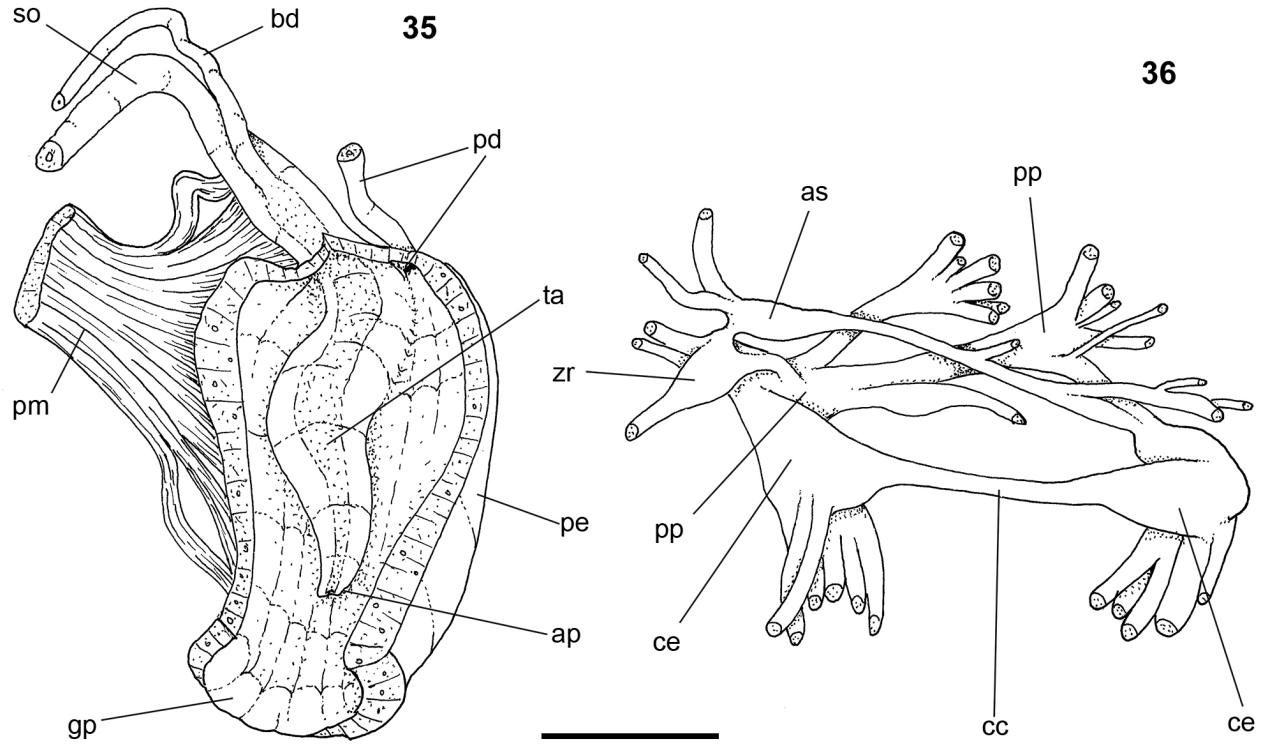
Figs 31–34. *Siphonaria pectinata*. Anatomy: 31 – odontophore, ventral view, superficial membrane and muscles removed, right peribuccal muscle (mj, left in Fig.) removed; 32 – expanded genital system, dorsal view, topology of some adjacent structures and transverse section in indicated regions also shown; 33 – midgut, dorsal view, mostly opened longitudinally to show inner surface, topology of buccal mass shown; 34 – detail of middle region of genital system, dorsal view, showing insertion of some structures. Scale bars 2 mm

maining, more marginal, ~23 pairs of teeth gradually becoming narrower, with cutting edge slightly shorter, constituting marginal teeth. Marginal teeth similar to lateral teeth, but with secondary cusp gradually appearing on lateral edge (Figs 16–18), being in most lateral teeth with ~half size of main, terminal cusp.

Salivary glands covering anterior half of oesophagus (Fig. 24: sg), forming single, white, thin mass passing through nerve ring. Pair of salivary ducts gradually distinguishable anteriorly, close to region of penetration in latero-posterior region of buccal mass (Fig. 26: sd). Salivary ducts running for short distance immersed in dorsal wall of buccal mass; opening as described above in posterior dorsal folds notch (Fig. 25: sa).

Oesophagus of ~80% haemocoel length, with thin, flaccid walls; anterior portion narrow passing through nerve ring (Figs 24, 26: es); inner surface of this anterior region with 8–10 longitudinal, narrow folds, as continuation of buccal cavity dorsal folds (Fig. 25: ef). Oesophageal crop occupying ~80% of oesophagus (Fig. 27: ec); inner surface with 8–10 narrow and low longitudinal folds, well-separated from each other (Fig. 33: ec). Stomach position and size described above (midgut in visceral mass section) (Fig. 24), relatively narrow and short, curved, mostly marked by pair of ducts of digestive gland (Figs 27, 33: dd) on boundary between end of oesophageal crop (ec) and intestine (in); gastric walls thin, flaccid; inner surface smooth, except for set of ends of oesophageal folds preceding gastric area (Fig. 33). Oesophageal insertion right and anterior, intestinal origin on left side and posterior, both close to posterior end of haemocoel (Fig. 24). Both ducts of lobes of digestive gland located close to each other, bent to left (Figs 27, 33: dd). Intestine initially narrow, making two tight loops as shown in Fig. 24 (in); middle portion slightly broader (Fig. 27: in), bearing aligned set of spherical faecal pellets (Fig. 24: fp). Rectum running obliquely from middle posterior end of haemocoel to right (Fig. 22: rt), short region preceding anus penetrating integument (tg) (Figs 22, 24: an). Anus simple, lacking sphincter, located between middle and anterior thirds of right margin, in middle and ventral to pneumostome (Figs 20, 22, 24: an). For other details, mainly histology, see KÖHLER (1894).

Genital system (Figs 32, 34, 35). Genital system mostly located along right side of haemocoel, compressed by digestive tubes and gland; occupying ~1/4 of haemocoel volume. Gonad described above (visceral mass), located in posterior-right region of haemocoel, bulging in digestive gland (Fig. 22: go); general form spherical-oval, with 4–5 branches of hermaphroditic duct originating on its ventral-left side (Fig. 32: go). Hermaphroditic duct (Fig. 32: hd) thick and weakly coiled, walls thick-glandular; running sigmoid-fashion along 1/3 haemocoel length; its anterior third distinctly narrow, flanking albumen gland; opening to spermoviduct side by side with albumen duct (Fig. 34: hd). Seminal receptacle (Figs 32, 34: sr) elongated, sac-like, as wide as to twice wider than hermaphroditic duct; sometimes very long and coiled (Fig. 32: sr) and sometimes shorter and curved (Fig. 34: sr); inner surface mostly smooth, except for anterior third, with 5–8 longitudinal folds. Albumen gland mostly slightly smaller than gonad, spherical (Fig. 32: ag), located just anterior to gonad (Fig. 22: ag); lumen flattened and wide (Fig. 32). Albumen chamber coiled as 3–4 increasingly larger whorls (Figs 32, 34: ac), occupying ~1/4 of albumen gland volume, located at anterior end of this gland; albumen duct opening side by side with hermaphroditic duct (Fig. 34: ad). Fertilisation complex (Fig. 34) with outlets of albumen gland duct, hermaphro-



Figs 35–36. *Siphonaria pectinata*. Anatomy: 35 – terminal portion of genital system, penis (pe) opened longitudinally; 36 – central nervous system (nerve ring), dorsal view. Scale bar 1 mm

ditic duct and duct of seminal receptacle close to each other on dorsal side of posterior region of spermoviduct (so). Spermoviduct (Figs 32, 35: so) curving along ~1/3 of haemocoel length; its posterior region located level with pneumostome, gradually tapering anteriorly, penetrating deeply in integument, at right surrounding isolated portion of shell muscle (Figs 22, 32: im); opening to posterior end of copulatory organ (Figs 32, 35: so). Prostate gland (Fig. 32: pt) occupying ~1/2 of posterior third of spermoviduct; ending before spermoviduct narrow portion. Anterior 2/3 of spermoviduct as single, narrow duct. Bursa copulatrix spherical (Fig. 32: bc), roughly as large as albumen chamber, located just anterior to it; bursa duct very narrow, running parallel to spermoviduct, in same course and position (Fig. 32: bd); opening to posterior end of copulatory organ (Fig. 35: bd) side by side with spermoviduct. Copulatory organ similar to penis or proboscis (Figs 32, 35: pe), located just anterior to isolated portion of shell muscle (Fig. 22: pe), with outer portion composed of thick muscular walls, length equivalent to that of isolated portion of shell muscle; also internal portion papilla-like, as projection starting in posterior end of outer portion, protruding internally towards exterior, tapering to sharp pointed tip; duct of bursa copulatrix (bd) and spermoviduct (so) inserting at base of this internal portion (Fig. 35: ta), opening in its tip (ap). Strong copulatory organ retractor muscle, originating in ventral-median region of isolated portion of shell muscle (Fig. 22: pm), surrounding its median and anterior surface, inserting fin-like along posterior and left sides of copulatory organ (Fig. 35: pm) in 4–5 different bundles (middle bundle broadest). Gland of copulatory organ elongated and twisted (Figs 22, 32: pg), compressed between buccal mass and adjacent shell muscle (Fig. 22: pg); lumen relatively wide, walls distinctly glandular in posterior 2/3, and smooth in anterior 1/3 (Fig. 32: pg); gradually narrowing, inserting in posterior region of copulatory organ between its outer and inner portions (Fig. 35: pd). Genital orifice very small, located at middle level of isolated component of shell muscle (Figs 22, 35: gp). For other details, mainly histology, see KÖHLER (1894).

Central nervous system (Fig. 36): nerve ring located at base of buccal mass (Figs 22, 27: nr). Pair of cerebral ganglia (ce) elliptical, about twice as long as wide; cerebral commissure (cc) as long as each ganglion. Each cerebral ganglion of about ¼ width of oral tube. Pair of pedal or pleuro-pedal ganglia (pp) slightly smaller than cerebral ganglion; pedal commissure as long as cerebral commissure, but slightly thicker; left pedal-cerebral connective shorter than right commissure, almost undetectable. Visceral (abdominal)-subintestinal ganglion (as) located close to left pedal ganglion, with about half its size. Parieto-supra-intestinal (zr) ganglion located close to visceral ganglion, connected to it by short connective; ce-zr connective 4–5 times shorter than ce-as connective. For other details see SALEUDDIN et al. (1997).

Habitat. Rocky intertidal zone – emerged-rocks at mid/low-shore levels and rock-pools at high-shore levels.

Distribution. Mediterranean; in Atlantic from Portugal to Gabon, Canaries (GIRIBET & KAWAUCHI 2016).

Measurements (in mm): MZSP 114172(2): 36.3 by 18.1; MZSP 115170(1): 32.3 by 23.8.

Material examined: Portugal, Setúbal, Sines, Oliveirinha Beach, 37°53'19.21"N 08°47'49.29"W, MZSP 115167, 3 specimens, MZSP 115169, 3 specimens, MZSP 115170, 2 specimens, MZSP 115171, 2 specimens, MZSP 115172, 2 specimens, MSP 115173, 2 specimens, 115174, 2 specimens (Maria Inês Seabra col, 5.ix.2013).

DISCUSSION

As in the case of most molluscs, the shell has for a long time been the main source of taxonomical knowledge on *Siphonaria* (HUBENDICK 1946). The shell of *S. pectinata* is characteristic and easy to identify due to its narrow and numerous radial ribs; despite this, it varies relatively widely. The variation (Figs 2, 9) shows that erosion by waves can modify the shell surface and, to some degree, the shell height. However, the variation is comparable to that found in other siphonariids (e.g., TABLADO & GAPPA 2001). The intertidal environment, with its high-energy water flow and wave action, influences the siphonariid shell shape and sculpture through some degree of phenotypic plasticity (COOKE 1911, TESKE et al. 2007). The phenotypic plasticity of the siphonariid shell shape and sculpture is regarded as ecologically adaptive in terms of intertidal zonation and geographical distribution: high-domed, light-coloured and more sculptured shells are found at higher shore levels and in tropical species (HODGSON 1999 and references therein); most of the samples analysed in this study came from high-energy sites. It is thought that some characters of the shield-shaped shells of *Siphonaria* can be influenced by the environment (WHITE et al. 2011).

The siphonariids (for specific taxa see below) have some anatomical peculiarities that are so far exclusive; some of them are discussed below. Despite the limpet-like shell shape, the usual somatic modifications involved in the "normal limpetisation pro-

cess" are not found in the siphonariids. The so-called "normal limpetisation process" will be dealt with in another paper, presenting the evolution from a typically coiled gastropod to an uncoiled, limpet-shaped one, in which all the structures are functionally restricted to the apertural region of the coiled snails. Most of the limpets examined within the project, for example patellogastropods (e.g., LEAL & SIMONE 1998); cocculiniforms (e.g., SIMONE 1996, LEAL & SIMONE 2000, SIMONE & CUNHA 2003); vetigastropod fissurellids (e.g., COSTA & SIMONE 2006, SIMONE 2008); caenogastropod hipponicids and capulids (e.g., SIMONE 2002); and heterobranch ancylids (e.g., SIMONE et al. 2012) (for synthesis see SIMONE 2011), have retained the visceral mass, which occupies the region just below and behind the shell apex. As all these groups are obviously a result of convergence, differences and convergent features appear. However, in the siphonariids (COTTRELL 1910, YONGE 1952, MARCUS & MARCUS 1960; this study), only the pallial roof is present below the shell apex (Figs 5, 19, 21); except for a small portion of the reno-pericardial structures (Fig 19: pc, ki), no visceral structure is located below the inner surface of the apex. Most of the siphonariid visceral structures are actually placed inside the haemocoel. This situation is unlike that found in any of the other above-mentioned limpets. In those groups the visceral hump is conical, as an internal cast of the apical shell region. In fact, the placement of visceral structures in the head-foot haemocoel is normal in another process which is common among gastropods – the limacisation – in which a typically coiled snail becomes a crawling, shell-lacking head-foot. While in the so-called "true limpets" the respiratory cavity consists of a space between the mantle and the visceral mass opening to the external environment at its anterior end, the respiratory cavity of siphonariids, the "false limpets", is almost entirely closed, with a small contractile orifice on the right side, where the characteristic siphonal groove is located. The generic name *Siphonaria* was inspired by this furrow, certainly produced by the flap which protects the pneumostome (Figs 6–7: fl).

Another interesting difference between the siphonariid way of limpetisation and that of the remaining above-mentioned limpets is the shell muscle. In the so called "true limpets" it is horseshoe-shaped (in top view), with the muscle scar as an unbroken semiring on the internal side of the shell. Although the siphonariid shell muscle also looks like a horseshoe, its right branch is interrupted at about ¼ of its length (Figs 19–20, 22: sm, im) in order to shelter the pneumostome (Figs 20, 22: pn) and the anus (an); a corresponding interruption is present in the muscle scar on the inner surface of the shell. This conformation of the shell muscle is associated with the characteristically smaller tenacity (force of attachment to the substratum) and greater foot flexibility of siphonariids compared to the other limpets, and thus it has ecological implications, such as their restriction to habitats protected from direct wave action and their mobility as highly active grazers (HODGSON 1999 and references therein). It is easy to interpret the horseshoe-shaped shell muscle as derived from the columellar muscle of the coiled gastropods. This is probably the case for the siphonariids, as at least the main portion of the shell muscle may be homologous to the columellar muscle. However, its isolated portion located at the right-anterior end (Figs 20, 22: im) cannot be derived from the columellar muscle, since the genital structures surround it externally (Fig. 22: so). Thus there is no way to envisage modifying of the columellar muscle in order to derive the siphonariid organisation, with some haemocoelic structures contouring a part of the muscle externally. To our knowledge, no developmental studies have furnished a clue on this issue (e.g., KNOX 1955). The best interpretation is that the main portion of the siphonariid shell muscle (sm) is actually derived from the columellar muscle; however, the isolated portion (im) may be a new acquisition, without any correlation with that of other gastropods. Unlike our observations of *S. pectinata*, in *S. tristensis* ("Leach" in Sowerby I, 1823 = *S. lessonii* Blainville, 1827, see GÜLLER et al. 2016) the genital structures have been illustrated as running directly anteriorly, not surrounding the isolated branch of the shell muscle (im) (DALL 1870: pl. 5, fig. 3); if this is confirmed, it will represent an additional siphonariid feature that merits further investigation. A possible misinterpretation is the shell muscle shape in *S. gigas* Sowerby I, 1825, which has been illustrated as continuous, only opening in the pneumostome area (HALLER 1893: figs 11, 12). On the other hand, and despite the above argumentation, the connection of the penis muscle with the isolated component of the shell muscle in *S. pectinata* (Fig. 22: in, pm) may be regarded as an indication that this portion of the shell muscle may be homologous to the columellar muscle. This contradictory issue certainly requires additional studies. In fact, the arrangement found in the heterobranch limpets has been mentioned by HASZPRUNAR (1985: 26) as being probably a result of the semi-detorted situation of the mantle/heart complex, also reflected by the euthyneuran nervous system. However, no conformation is found in the heterobranch ancylids (SIMONE et al. 2012). On the other hand, there is a strange similarity between the isolated component of the shell muscle (Fig. 22: pm) and the "adductor" muscle of some Sacoglossa, used for occluding the shell aperture by the flexible outer lip. This "adductor" muscle occurs, for example, in the genera *Ascobulla* Marcus, 1972 and *Cylindrobulla* Fischer, 1857, and is surrounded by some components of the genital ducts (MARCUS &

MARCUS 1970; personal observation); in other words, it might be easier to explain the siphonariid limpetisation starting from a sacoglossan ancestor. Indeed, some molecular approaches have shown a close relationship between the siphonariids and the sacoglossans (GRANDE et al. 2004, 2008, DAYRAT et al. 2011, DINAPOLI et al. 2011, JENSEN 2011).

The siphonariid head is also peculiar. It is a widely bi-lobed anterior flap (Figs 6–8: he), with a pair of eyes immersed in the integument (Figs 6–7: ey, 8) (DALL 1870, YONGE 1952). No other group of pulmonates has a similar head structure (TILLIER et al. 1992, SIMONE 2011). Normally the head is convex and provided either with a pair of non-retractile lateral tentacles (basommatophorans) or two pairs of retractile ones (stylommatophorans). The eyes are normally associated with those structures, being located at the base of tentacles in basommatophorans, and on the tip of dorsal tentacles in stylommatophorans. The siphonariid model, in fact, resembles that found in some opisthobranchs, such as some doridaceans and their allies (e.g., ALVIN et al. 2014), apart from the presence of rhinophores.

The dorso-lateral pedal defensive glands, epidermal multicellular structures not described here, appear to be an interesting characteristic of the siphonariids (PINCHUCK & HODGSON 2009). This kind of gland resembles that found in some calyptraeoidean caenogastropods (SIMONE 2002), although in that case the glands are distributed along the mantle border. In *S. pectinata*, an anti-predatory secretion is produced and exuded as a thick and white mucus (OCAÑA 2003), containing metabolites described as toxic (PAUL et al. 1997) and in live specimens its lateral release is commonly observed when the animal is disturbed. Effective chemical defence against predation has been demonstrated for *S. capensis* Quoy et Gaimard, 1833 in South Africa (MCQUAID et al. 1999), and is probably widespread in the genus, since at least 12 species have been found to biosynthesise polypropionates, compounds associated with the highly glandular nature of the foot-tissue and with their unpalatability (HODGSON 1999, MCQUAID et al. 1999, and references therein). A defensive response to predation is also present in intertidal pulmonate limpets of the allied family Trimusculidae, including release of a white secretion through the extended mantle, although in this case diterpenes have been identified as the predator deterring chemicals (SAN-MARTÍN et al. 2009). Though the ecological role of these secretions is mostly unknown, predation avoidance in an environment susceptible to both terrestrial and aquatic predators certainly has implications for several aspects of the snail's biology, such as longevity and foraging behaviour, and thus it may have an adaptive value for the siphonariids.

As air breathing pulmonates, the siphonariids have a well-developed "lung" (Fig 20: pu), in which the air is inhaled and exhaled though the pneumostome (pn) (YONGE 1952), and the flap ventral to it controls the breathing (Figs 6–7: fl, which is indicated in Figs 20, 22 as "pn"; "siphonal notch" by COTTRELL 1910; "anal lobe" by MARCUS & MARCUS 1960). The normal pulmonate lung bears well-developed vessels, however this is not true of the siphonariids, which, instead, have a well-developed gill (Figs 5, 19, 21: gi), with a ctenidial vein running to the auricle (Fig. 21: cv). The location, structure and variation of the thick gill filaments suggest that the siphonariid gill is secondary, and non-homologous to the monopectinate gill of other monotocardians (SIMONE 2011). This secondary condition of the siphonariid gill has long been accepted in the literature (VILLIERS & HODGSON 1987). Besides, the siphonariid secondary gill appears to vary between species; those of *S. pectinata* (studied here) and *S. hispida* Hubendick, 1946 (MARCUS & MARCUS 1960) appear to have a simpler structure than that of *S. obliquata* Sowerby I, 1825 (COTTRELL 1910: fig. 1) and *S. capensis* (VILLIERS & HODGSON 1987), in which the filaments have secondary folds; the gill of *S. gigas* is peculiar in being displaced to the periphery of the pallial cavity (HALLER 1893: figs 11, 12). The combination of gill and lung indicates that the animal is amphibious, breathing in both water and air (WELLS & WONG 1978, HODGSON 1999). Along with this ability, the respiratory physiology of siphonariids provides other advantages to cope with the harsh intertidal conditions, such as the facultative depression of aerobic metabolism in response to prolonged air exposure and desiccation; and the tolerance to hypoxic conditions and possible anaerobiosis on shores subjected to episodic sand inundation (HODGSON 1999). The anatomical basis for this physiological resilience is the withdrawal of the mantle skirt and the closure of the pneumostome, which close the connection of their vascular cavity with the external environment. These physiological adaptations enable the animal to survive and save energy in adverse habitats. As described above, the pneumostome of *S. pectinata*, and the siphonariids in general, lacks sphincter and cannot be closed (except by ventral flap).

The siphonariid reno-pericardial structures are relatively small for a limpet of such size, except for *S. gigas*, which has a relatively large kidney (HALLER 1893). The renal component touching the roof of the pallial cavity in *S. pectinata* (Fig. 19: ki) looks much smaller than the corresponding structures in its congeners (*S. obliquata* from New Zealand – COTTRELL 1911; seven species of *Siphonaria* from South Africa – ALLANSON 1958; *S. hispida* – MARCUS & MARCUS 1960). However, the dimensions of the pericardium are similar across these taxa.

The siphonariid digestive system also shows some interesting peculiarities. The odontophore is basically typical of most Pulmonata, bearing a well-developed pair of hard odontophore cartilages, widely fused with each other along the ventral-medial edge (Figs 28–31: oc); and the basic arrangement of intrinsic and extrinsic muscles. However, despite the relatively high degree of cartilage fusion, *S. pectinata* has a well-developed horizontal muscle (Figs 30–31: m6), in which the anterior region overlaps with the cartilage fusion (Fig. 31). The pair of ventral tensor muscles of the radula (Figs 30–31: m11) is extraordinarily wide, even duplicated (m11, m11a); while in other pulmonates the pair of m11 is narrow and thin. This thick m11 conformation indicates that the radula is used in a way characteristic of hard-scraper snails. Besides the strong pair m11, most of the odontophore muscles are also relatively strong and thick (Fig. 20), showing an intense use of the radula, which has been also observed in other siphonariids (Black et al. 1988). Nonetheless, this contrasts with the shortening of the radula (Figs 25–26, 28: rs). As a rule, the intense use of the radula results in its considerable elongation, as shown by littorinid caenogastropods (e.g., Simone 1998), in which the radula is longer than the shell. The most modified pair of odontophore muscles in *S. pectinata* is the m7 (Figs 30–31). The pair m7 connects the medial and middle edge of the pair m11a with the internal surface of the radular sac (Fig. 29: m7), crossing the middle level of the m6 (Figs 30–31: m7). The function and homologies of the m7 pairs are enigmatic (Simone 2011), and in this respect *S. pectinata* is no exception. As the odontophore muscles of other siphonariids have not been subject to any other study, the interpretation of such differences is impossible, and they may be relevant at any taxonomic level.

The radular morphology of the siphonariids is typical of pulmonates. The radula is a carpet of small and relatively uniform teeth. In contrast to the normal pulmonate radula, the distinction between marginal and lateral teeth is difficult in most siphonariids, showing a gradual change from medial to lateral regions. In other siphonariids lateral and marginal teeth differ slightly, for example in *Williamia radiata* (Pease, 1860) in which the former teeth are strongly bicuspid, while the marginal ones are single plates (Marshall 1981); this pattern is very different from the more homogeneous radula of *S. pectinata*. The radula of *S. pectinata* differs from that of *S. hispida* (Marcus & Marcus 1960: fig. 15) in having the rachidian still narrower, and in lacking multiple cusps at the cutting edge of the marginal teeth; it differs from that of *S. obliquata* (Cottrell 1910: fig. 2) in having a narrower and sharper pointed rachidian; and it also differs from *S. thersites* Carpenter, 1864 and *S. tristensis* (actually *S. lessonii*) in having the secondary cusps of the lateral and marginal teeth not so well-developed (Dall 1870: pl. 5, fig. 1). It should be emphasised that in *S. pectinata* the radula can bear simple (Figs 10–12) or bifid (Figs 13–18) tips, with possibly variable excavating ability into the rocky substrate while foraging. Overall, the fine-toothed radula should be comparatively weaker than the large and strongly mineralised teeth typical of patellogastropod radulae (Hodgson 1999). These differences in the radular structure of microphagous grazers are probably translated to grazing capacity, influencing interactions among sympatric limpet taxa. In SW. Portugal, *S. pectinata* co-occurs with *Patella depressa* Pennant, 1777 and *Patella ulyssiponensis* Gmelin, 1791, the dominant limpet species of mid and low intertidal zone, respectively. Compared to these patellids, *S. pectinata* is generally much less abundant and its distribution is more variable in space (Seabra in prep.). One of the possible mechanisms explaining this pattern might be an inferior competitive ability of *S. pectinata* in exploiting limited food resources (Seabra in prep.). Ocaña & Fa (2003) found that *S. pectinata* in Gilbraltar grazed exclusively on superficial soft algae, suggesting that this feeding specialisation could reduce competition with *Patella* limpets, which graze by deep scraping of the rocks and often include calcareous encrusting algae in their diet. In fact, siphonariids are often out-competed but not completely eliminated by patellogastropods (Hodgson 1999).

The siphonariid oesophagus is wide and has thick muscular walls (Figs 27, 33: ec). This shows that the organ serves to store food, being a gizzard, and is capable of distension and additional mechanical food processing. The siphonariid stomach, on the other hand, is relatively small, being a single corner in which both ducts of the digestive gland lobes open (Fig. 27: st); the stomach of *S. pectinata* looks proportionally smaller than that of *S. obliquata* (Cottrell 1910) and of *S. hispida* (Marcus & Marcus 1960). The remaining digestive structures are typical of herbivore or microphagous snails, with several intestinal loops compressed by the remaining head-foot structures (Fig. 24: in). The formation of faecal pellets (Fig. 24: fp), instead of a single faecal string, is another interesting character in most siphonariids, as well as the location of the anus on the floor of the pallial cavity. In most gastropods which have pallial cavity the anus is located in its roof. The rectal region in *S. gigas* has a large bulging chamber (Haller 1893: fig. 16: ed), which apparently is unique in that species.

The simplicity of the intestinal loops in the siphonariids is noteworthy, compared to other herbivore limpets, being related to the fact that the digestive gland rather than the intestine performs most of the enzymatic and absorption functions (Murty et al. 2013). However, these functions of the digestive gland appear to be normal in Mollusca. In about

half of the examined specimens the contents, apart from vegetal matter pulverised by the radula, included whole specimens of the minute caenogastropod *Skeneopsis planorbis* (Fabricius, 1780), and sometimes young specimens of the mussel *Mytilus* sp. These organisms were found in the gastric and intestinal contents, with the soft parts apparently intact. They might be merely contaminants, although it cannot be excluded that they serve as additional nutritive matter, which could explain the relative shortness of the intestine.

The genital system of *S. pectinata* has the same general structure as in the remaining siphonariids, having roughly the same main components (Hutton 1882). However, each known species appears to bear the following distinctive characters: *S. pectinata* (Fig. 32) differs from *S. hispida* (Marcus & Marcus 1960) in having proportionally smaller middle glands (albumen and capsule glands: ac, ag, which remain constant in specimens collected throughout the year) compared to the gonad; in the hermaphrodite duct (hd) being wider and blunter, and mainly in the structure of the copulatory organ, lacking flagellum and epiphallus, being a single bulk, while in *S. hispida* it is Y-shaped. It differs from *S capensis* (Allanson 1958, Pal 2007) in having the hermaphrodite duct longer and more coiled, more divided in middle, and in a more elongated shape of the penis gland (pg). It differs from *S. anneae* Tomlin, 1944 (= *S. carbo* Hanley, 1858, see: Teske et al. 2007) and from *S. deflexa* Born, 1778 (Allanson 1958) in having the penis retractor muscle (pm) much wider and in a more elongated shape of the penis gland (pg). It differs from *S. obliquata* (Cottrell 1910: fig. 6) in having the hermaphrodite duct (hd) much thicker, the seminal receptacle (sr) much longer and wider, and the penis gland (pg) longer and coiled. It differs from *S. virgulata* Hedley, 1915 and from *S. luzonica* Reeve, 1856 in having the penis gland inserted in the posterior instead of the anterior region of penis (Hubendick 1955), the latter situation appearing to be the commonest (Hutton 1882). It differs from *S. serrata* (Waldheim, 1807) (Pal 2007) in having the penis gland inserted in the posterior end of the penis and not on its base, and in lacking flagellum in this gland. It differs from *Williamia radiata* (Ruthensteiner et al. 2007) in having a much smaller and single penis gland, inserted on the penis tip. For additional details of the siphonariid genital structures see Pal & Hodgson (2002, 2003, 2005) and Pal (2007).

The genital orifice of most siphonariids is mostly single and simple, located in the right-anterior region of the body edge, as described for *S. pectinata* (Fig. 22: gp). So far, the genital system of *S. gigas* is the only one with two orifices (Haller 1893: fig 19). Preceding the genital aperture, there is in *S. pectinata* what used to be called penis (Fig. 32: pe), but actually it serves both female and male branches of the genital system. Inside (Fig. 35) it has a main, central, papilla-like component as outlets of the spermoviduct and of the bursa (ta), while the outlet of the penis gland is located posteriorly, outside the papilla (pd). This whole penis resembles those of the remaining pulmonates in being retractile, proboscis-like, and certainly its main body (Fig. 35: pe) may be everted during copulation, with the central papilla as the tip (ta, ap). The retraction of the structure is certainly provided by the penis retractor muscle (pm), which is very similar to those of the remaining pulmonates, mostly originating from columellar muscle. The penis muscle of the siphonariids originates from the isolated component of the shell muscle (Fig. 22: in, pm). It is interesting to note that protandry has been documented in *S. pectinata* (e.g., Marcus & Marcus 1960).

The siphonariid central nervous system is relatively uniform amongst the species, sharing some distinctive characters compared to other pulmonates, such as the wide distance between both cerebral ganglia (Fig. 36: ce); the asymmetry of the shape of the left and right pedal and pleural ganglia (pp); the proximity of the abdominal or parieto-supra-intestinal ganglia (zr); the different length of the connectives between the cerebral and the pleuro-pedal ganglion; and the particular conformation of the connective of the visceral (abdominal)-subintestinal ganglion (as). The nerve ring of *S. pectinata* is different from that of other siphonariids in having a single pair of pedal commissures, which is relatively thick, as it is also the case of *Williamia gussoni* (Costa, 1829) (Ruthensteiner 2006), while *S. hispida* (Marcus & Marcus 1960) and *S. obliquata* (Cottrell 1910) have this commissure thinner and duplicated. Another inter-specifically contrasting aspect is that *S. pectinata* and *S. hispida* (Marcus & Marcus 1960) have the pedal and pleural ganglia completely fused (PP), while they are close, but separated in *S. obliquata* (Cottrell 1910). The arrangement of connectives of the visceral (abdominal)-subintestinal ganglion (as) and the parieto-supra-intestinal ganglion (zr) appears to be slightly different in all species in which this feature was examined.

As mentioned above, living intertidally, the siphonariids are part of a complex ecosystem that highly influences their shell morphology. Their outer surface becomes a garden of epibionts, an example can be seen in Fig. 9 with the fouling of non-calcareous algae *Ralfsia verrucosa* (Areschoug, 1845). Besides, it is relatively common to find epibiotic fouling or filamentous algae covering the specimens living in more damp habitats, such as *Caulacanthus ustulatus* (Mertens ex Turner) Kützing, 1843 in the specimen in Figs 7–8. The colour of the head-foot also varies to some extent. This pertains mainly to the foot (rath-

er than the head, which is usually yellowish) whose colour varies from yellowish orange to grey, possibly depending on the diet, habitat or the animal's size. Some of the spots are most probably related to the dorso-lateral pedal defensive glands (PINCHUCK & HODGSON 2009) which were not investigated here.

The phylogenetic placement of the family Siphonariidae is debateable, with supporters of all rational possibilities, from the traditional Basommatophora, to Archaeopulmonata, Opisthobrancha and Stylommatophora. For example, *Siphonaria* may be nested within opisthobranchs (GRANDE et al. 2008, WHITE et al. 2011) or constitute the basal-most lineage of pulmonates (KLUSSMANN-KOLB et al. 2008, DAYRAT et al. 2011). Of course, the contradictory results have been provided by molecular approaches, which in themselves are somewhat discordant and still under analysis. Concerning the morphological characters, any possible similarity to each of the above-mentioned taxonomic groups does not bear a closer examination. An example is the pallial cavity, whose resemblance across some of these groups has been interpreted as superficial, and explained as homoplasies in response to similar environmental pressure (JENSEN 2011).

A discussion on the phylogenetic uniformity and the problems related to the siphonariids in particular, can be read in SCHRÖDL (2014). The issue will be further analysed in an ongoing paper (senior author). So far, at least from a morphological point of view (see: GIRIBET 2015), some basic synapomorphies of Pulmonata can be evoked, which are present in both Basommatophora and Stylommatophora, such as the unique dorsal jaw plate; the retractile shape of penis; the penis retractor muscle originating from columellar muscle; the bursa copulatrix with elongated duct, inserting close to the genital orifice and placed close to the pericardium. Besides, though some siphonariids appear to have no penis, the general organisation of the genital system of the so far investigated siphonariids indicates an arrangement which is mostly similar to that found in as other plesiomorphic pulmonates (MORTON 1955, RUTHENSTEINER & STOCKER 2009).

Though the phylogenetic relationships of the Siphonariidae are not within the scope of this paper, the facts and arguments briefly presented above should be at least considered. The problem will be dealt with in a future paper which will discuss representatives of several basal and higher pulmonates in a formal phylogenetic scenario.

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