J. Moll. Stud. (1998), **64**, 355–386

MORPHOLOGY, ANATOMY AND HISTOLOGY OF FOUR SPECIES OF *ARMINA* RAFINESQUE, 1814 (NUDIBRANCHIA, ARMINOIDEA, ARMINIDAE) FROM THE MEDITERRANEAN SEA AND THE ATLANTIC OCEAN

ANNETTE KOLB
Lehrstuhl für Spezielle Zoologie, Ruhr-Universität, 44780 Bochum, Germany
(Received 4 December 1996; accepted 1 June 1997)

ABSTRACT

Three Mediterranean arminids (*Armina neapolitana* (Delle Chiaje, 1824); *Armina tigrina* Rafinesque, 1814 and *Armina maculata* Rafinesque, 1814) and one Atlantic species (*Armina loveni* (Bergh, 1860)) were examined morphologically, anatomically and histologically. Detailed descriptions of all organ systems are given. Marginal sacs are described for all four species.

All four species are very similar in their morphology, anatomy and histology. Differences are apparent in the colour and structure of the notum, in the shape of the radula teeth, in the structure of the digestive gland and in the anterior genital complex.

The data gathered in the examinations are compared to existing literature. Additionally, the four species are compared to other species of *Armina*. Since the existing data about these are sparse, the comparison is rather incomplete. More information especially about anatomy needs to be gathered for a better understanding of the genus.

INTRODUCTION

The Arminidae are poorly known nudibranchs. They burrow in the sediment (Bergh 1891), live mainly in deep water (Bergh, 1891; Marcus & Marcus, 1966) and little is known of their biology. Knowledge of the anatomy of arminid nudibranchs, especially other than *Armina* is very fragmentary and the phylogeny of the Arminidae (i.e. the genera *Armina*, *Dermatobranchus, Histiomena, Linguella, Pleurophyllidiella* and *Pleurophyllidiopsis*) is unclear. Characters shared by members of the family are an elongate, flattened body narrowing towards the tail (Schmekel & Portmann, 1982; Lance, 1962); longitudinal ridges or pustules on the notum (Pruvot-Fol, 1954); a distinct oral veil and retractile rhinophores (Schmekel & Portmann, 1982); a caruncle in front of the rhinophores (Bergh, 1891); and a radula typically having a broad and denticulated rachidian tooth and partly denticulated, falciform laterals (Odhner, 1939). All these characters, except for the caruncle and the subnotal lamellae, which probably have evolved within the Arminidae and are typical for some but not all genera (Kolb & Wägele, in press), are plesiomorphic and no autapomorphies have yet been recognized for the Arminidae.

The genus *Armina* with more than fifty nominal species has a worldwide distribution. Species typically have two different types of subnotal lamellae: branchial and hyponotal lamellae (e.g. Miller & Willan, 1986). The rhinophores are situated close together in front of the continuous notal margin. Most studies on *Armina* have concentrated on the morphology and the structure of the radula (e.g. Baba, 1949, 1955; Pruvot-Fol, 1955; Thompson, Cattaneo & Wong, 1990). Our knowledge about the anatomy of *Armina* is sparse and so far details are available for less than one third of all nominal species. The anatomical descriptions mainly concentrate on the digestive and the genital system. Hardly anything is known about the nervous, circulatory and the excretory system of the animals. A few detailed papers describing the anatomy of tropical species were published by Marcus & Marcus (1960, 1967) and Marcus (1971). Only Bergh (1866–67, 1869, 1879, 1890) presented the morphology and anatomy of some European and tropical *Armina* species in considerable detail, including three of the species studied here—*Armina maculata* Rafinesque, 1814, *Armina tigrina* Rafinesque, 1814 and *Armina loveni* (Bergh, 1860). Since then further publications on these species have been by Pruvot-Fol (1937, 1954), Schmekel & Portmann (1982) and Ballesteros (1981, 1983). Garcia & Garcia-Gomez (1988, 1990a, 1990b) presented some anatomical details of *A. maculata*.

So far nothing has been published on the histology of the organ systems of these animals. The present paper concentrates on the redescription of *Armina neapolitana, Armina tigrina, Armina maculata* and *Armina loveni* including the anatomy and histology of all the organ systems. Those data may contribute to a better understanding of these exotic nudibranchs and of the phylogeny of the Arminidae.

MATERIAL AND METHODS

Specimens were fixed in 4% formaldehyde/seawater after relaxation in magnesium chloride. Later they were transferred to 70% ethanol for storage. One or two specimens of each species were used for macropreparations. For the histological examinations of the other specimens, the products of Kulzer Histo-Technik (hydroxyethylmetacrylat) were used. The histological sections (2.5 μm thick) were stained with toluidine blue. The buccal armature was examined with a Hitachi S-450 scanning electron microscope. Detailed information about the specimens is given in Table 1.

RESULTS

Armina neapolitana (Delle Chiaje, 1824)

Synonymy:

Pleurophyllidia neapolitana Delle Chiaje, 1824
Pleurophyllidia lineolata Delle Chiaje, 1841
Pleurophyllidia vasconica Cuenot, 1914

External morphology:

The colour of the living animals is unknown. The body is elongate, flattened and narrows towards the posterior. Dorsal notal ridges (maximum of 16) are indistinct in preserved animals (Fig. 1A, C, D). Black pigment is visible through the epidermis. The oral veil is distinct (Fig. 1A) and behind it (only visible in specimen No. 1) two club-shaped rhinophores are present, each with 9 to 11 vertical lamellae (Fig. 1B). They contain the rhinophoral ganglia and nerves, which are embedded in muscular and connective tissue. Eyes are visible through the epidermis at the outer base of each rhinophore. A strong retractor muscle inserts just beneath. A triangularly shaped caruncle lies in front of the rhinophores (Fig. 1D), with spots of black pigment. It consists of irregularly arranged connective tissue fibres and does not contain any nerves. The genital opening is on the right side, typically just behind the branchial lamellae

Table 1. Length, location, date and preparation methods of specimens.

Species	Specimen	Length (mm)	Location	Date	Preparation
Armina neapolitana	No. 1	20	Banyuls-sur-mer, depth 70 m	June 1993	macroprep., SEM
	No. 2	22.5	Banyuls-sur-mer, depth 70 m	June 1993	histology
	No. 3	31	Banyuls-sur-mer, depth 70 m	June 1993	histology
Armina maculata	No. 1	27	Banyuls-sur-mer, depth 70 m	June 1993	histology
	No. 2	35	Banyuls-sur-mer,	May 1994	macroprep., SEM, genital complex for histology
	No. 3 (from the MNHN Paris)	37	unknown	unknown	morphology
	No. 4	63	Banyuls, depth 40–60 m	September 1978	macroprep., SEM
Armina loveni	No. 1	37	Irish Sea	July 1995	macroprep., SEM
Armina tigrina	No. 1 (from the MNHN Paris)	30	French coast	unknown	histology
	No. 2 (from the BMNH London)	53	Italy, Liguran Sea depth 160 m	unknown	macroprep. of genital complex

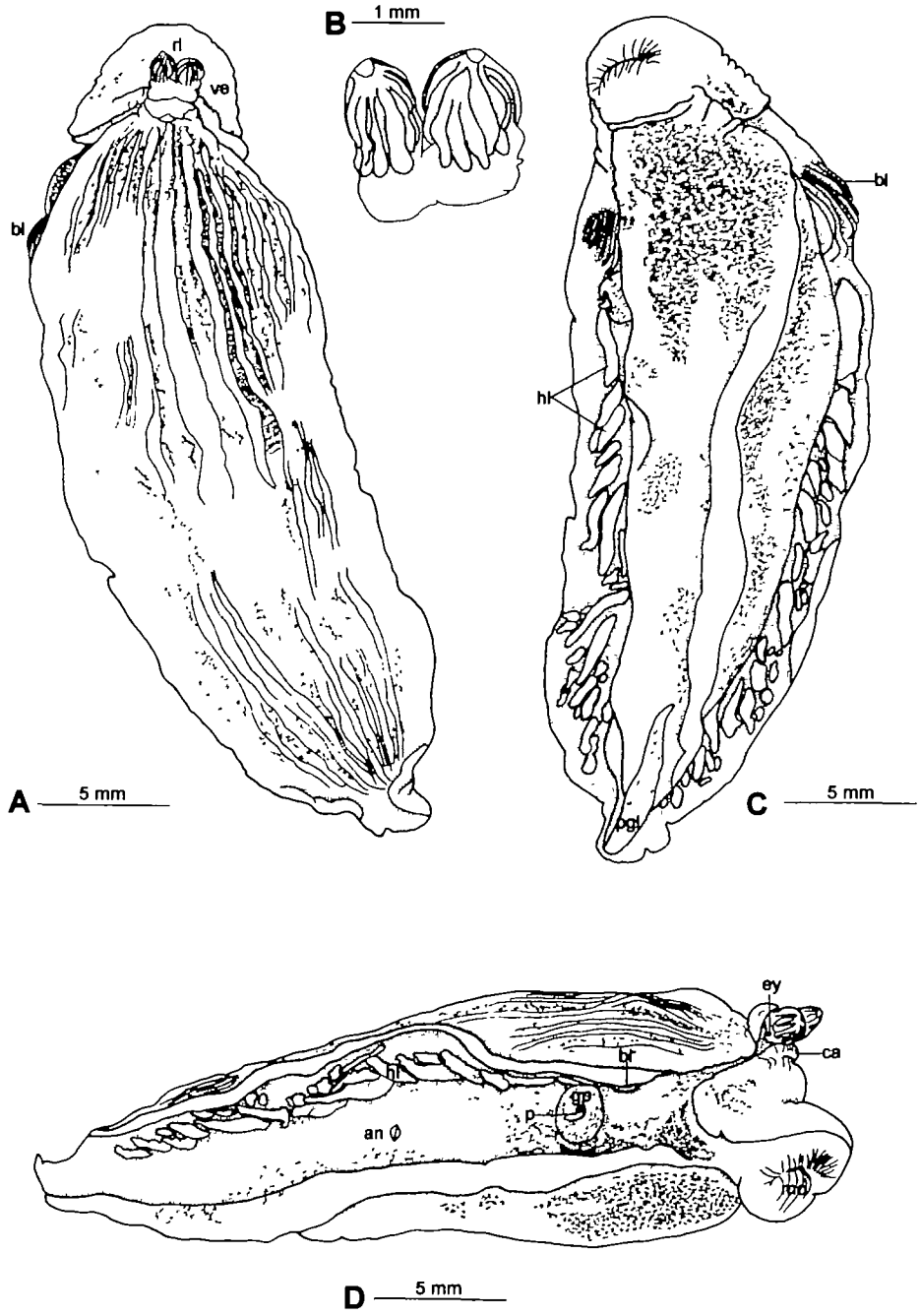


Figure 1. *Armina neapolitana*. **A.** Dorsal view. **B.** Rhinophores. **C.** Ventral view. **D.** Lateral view. Abbreviations: an, anus; bl, branchial lamellae; ca, caruncle; ey, eye; gp, genital papilla; hl, hyponotal lamellae; mo, mouth opening; p, penis; pgl, pedal gland; ri, rhinophores; ve, oral veil.

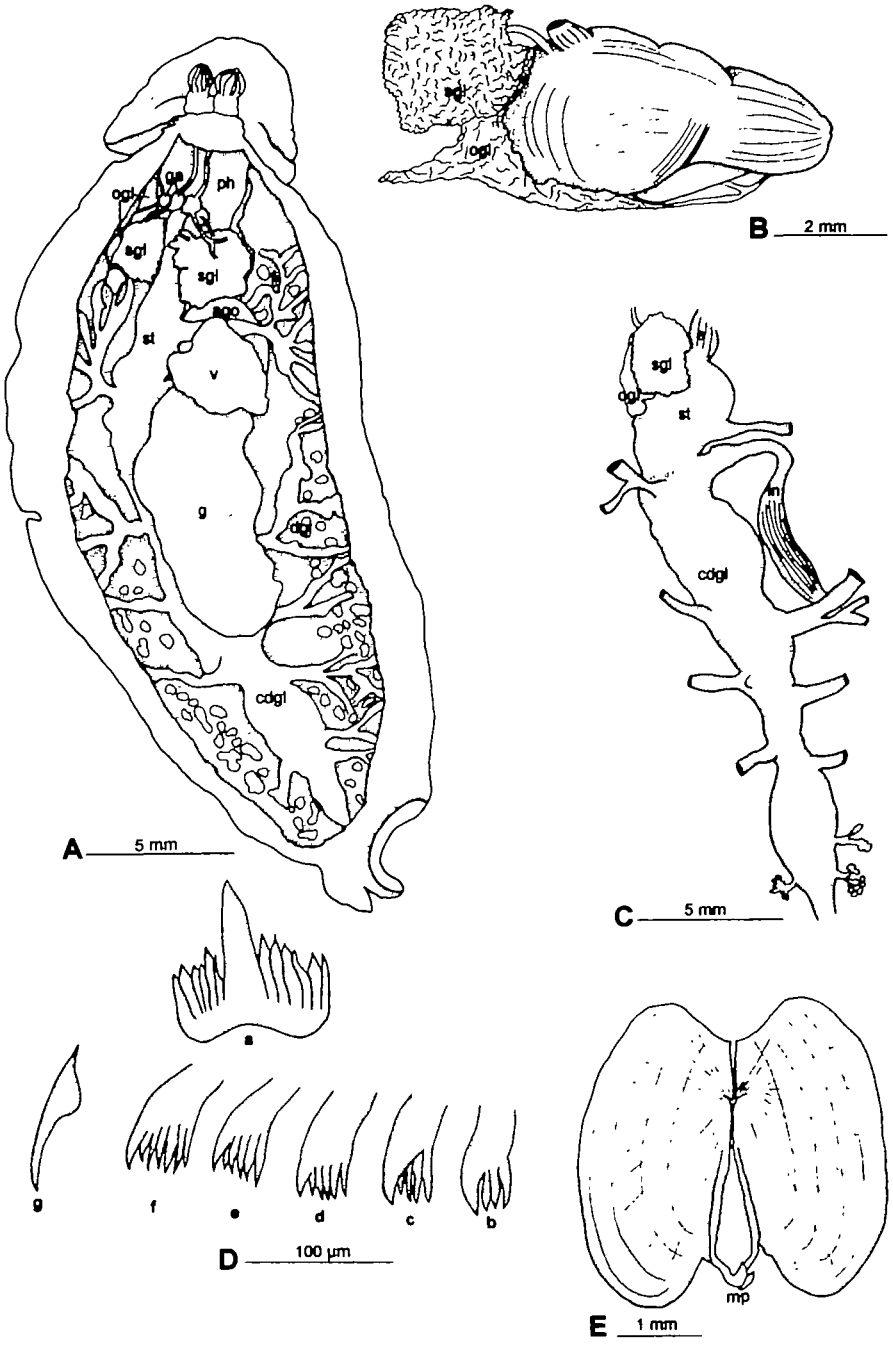


Figure 2. *Armina neapolitana*—Anatomy, **A.** Dorsal view after removal of the notum. **B.** Lateral view of oral tube and pharynx with adjacent glands. **C.** Dorsal view of the digestive tract without glandular parts of the digestive gland. **D.** Radula teeth, **a.** rachidian tooth, **b.–f.** lateral teeth, **g.** marginal tooth. **E.** Jaws. Abbreviations: ago, anterior genital organs; cdgl, central canal of digestive gland; dgl, digestive gland; e, oesophagus; g, gonad; ga, ganglia; in, intestine; mp, masticatory process; ogl, oral gland; ph, pharynx; sgl, salivary gland; st, stomach; v, ventricle.

(Fig. 1D). The position of the anus varies between individuals from the middle to posterior third of the right side. The renal opening lies between the genital aperture and the anus (Fig. 1D).

On the ventral side of the notal brim, there are 18 to 32 branchial lamellae (primary and secondary) present on each side (Fig. 1C). Figures 4D and 5A show histological sections of the branchial lamellae. The epithelium consists of cuboidal cells mainly with basal nuclei. Spherical ciliated cells with a large central nucleus (probably glandular cells) are located in the distal part of the lamellae. The epihelium is underlaid by a layer of connective tissue. Nine to 19 hyponotal lamellae are present on each side, arranged transverse to the edge of the foot. Branches of the digestive gland are partly visible through the epithelium of the hyponotal lamellae. It is composed of cuboidal cells. Cilia are not visible (Fig. 4B). The digestive gland does not extend into the branchial lamellae. The foot is distinct from the mantle edge. The pedal gland (Fig. 1C) is white and located at the posterior end of the foot sole. The gland is located directly beneath the epidermis. It is composed of a few layers of cells containing large nuclei and numerous light-staining granules (Fig. 4E). Irregular gaps and muscle fibres are interspersed between the glandular cells.

Marginal sacs are only distinguishable in the histological sections. They are spherical and located on the lateral notal edge of the mantle brim. Only immature stages could be found. They contain irregularly arranged cells. Some of these have large nuclei, others contain only a single droplet of violet secretion. Other secretions are not visible (see marginal sacs of *Armina loveni*).

The epidermis is formed by a highly vacuolized epithelium consisting of prismatic cells with a basal nucleus. The vacuoles do not stain with toluidine blue ('Spezialvakuolenzellen' Schmekel, 1982). In certain areas, especially around the oral tube, the epidermis is heavily ciliated. Subepidermal glandular cells are also present here. Glandular cells are scattered throughout the dorsal and ventral epidermis and are absent in the lateral epithelium.

Anatomy

Digestive system: The oral tube is short and lined by a prismatic epithelium in its ventral part, similar to the epidermis. The dorsal epithelium is heavily folded and lined by a thin layer of cuticle. Small, densely ciliated ducts of the oral glands enter the oral tube with a common duct in the middle of its ventral side (Fig. 2B). The oral glands are composed of white to yellowish follicles, which stain light purple with toluidine blue. They are located at the sides and especially beneath the pharynx. In the follicles of the oral glands two different types of glandular cells are present: one type contains a large basally lying nucleus and many non-staining vesicles of different sizes (Fig. 4Cb); the other type contains one large grey vesicle and generally a few small ones. A nucleus is not visible. The muscular buccal mass is large, 4.25 × 3.3 mm (Fig. 2B). The jaws are large and heavily cuticularized, with a thickened masticatory border and a denticulated masticatory process (Fig. 2E). The radula is multiseriate, with the formula 48 × 45.1.1.1.45. The rachidian tooth is broad (breadth 100 μm) with 6–7 denticles on either side of a median cusp (Fig. 2Da). The first lateral is hook-shaped with approximately 3 sharp denticles on the outer side (Fig. 7B, C). Succeeding laterals are elongate and fork-like with 4–7 long cusps (Fig. 2Db–f; 7A, D). The outermost lateral is narrow and smooth with a single blade (Fig. 2Dg).

The salivary glands are large and orange, consisting of numerous tube-like multicellular follicles. The cells of the follicles contain a nucleus at the base and spherical blue-staining granules (Fig. 4Ca). The structure of the follicles is not always homogenous, some cells also contain non-staining vesicles similar to the first type of cells of the oral glands. The salivary glands cover the buccal bulb and part of the oesophagus (Fig. 2A). The non-ciliated efferent ducts of the salivary glands enter the pharynx dorsally in front of the opening to the oesophagus. The oesophagus is tube-like, leading through the nerve ring towards the stomach. The epithelium between the pharynx and oesophagus is highly folded and lined with a cuticle. The oesophageal epithelium consists of ciliated columnar cells. The stomach is lined by cuboidal cells with large, basally-lying nuclei. In some parts the cells are elongate, becoming cylindrical in shape. Cilia are not visible. Two ducts from the digestive gland enter the stomach (Fig. 2A, C). The first (right digestive gland) opens on the right side anterior to the intestine. The second (left anterior digestive gland) leads from the left side across the opening of the intestine. The transition between the stomach and the posterior left part of the digestive gland is continuous, the latter forming a central canal of 16 mm length. Its epithelial lining is similar to that of the stomach. In the posterior part of the central canal ciliated cylindrical cells are present. The lateral tubes of the digestive gland open to the sides of the central canal, 6 to the right side and 5 to the left. All lateral tubes ramify into branches, consisting of small follicles. The cells of the follicles contain many vacuoles and granules. At least two different cells types can be distinguished: 1. Cylindrical cells with basal nuclei partly containing blue-staining vacuoles or vacuoles without secretions. These cells are located at the distal edges of the follicles. 2. Cells with a nucleus which contains a visible nucleolus. Vacuoles are not present. This truly digestive part of the gland is located only in the lateral parts of the body within the notal tissue (Fig. 4A) and in the hyponotal lamellae. The intestine is tube-like, with a dorsal typhlosole, running posteriorly past the reproductive system, along the ventral side of the anterior gonad. It is lined by an epithelium consisting of cylindrical ciliated cells.

Nervous system: Cerebropleural ganglia (0.8 × 0.5 mm) are located on the dorsal side of the pharynx at the anterior end of the oesophagus (Fig. 2A). They

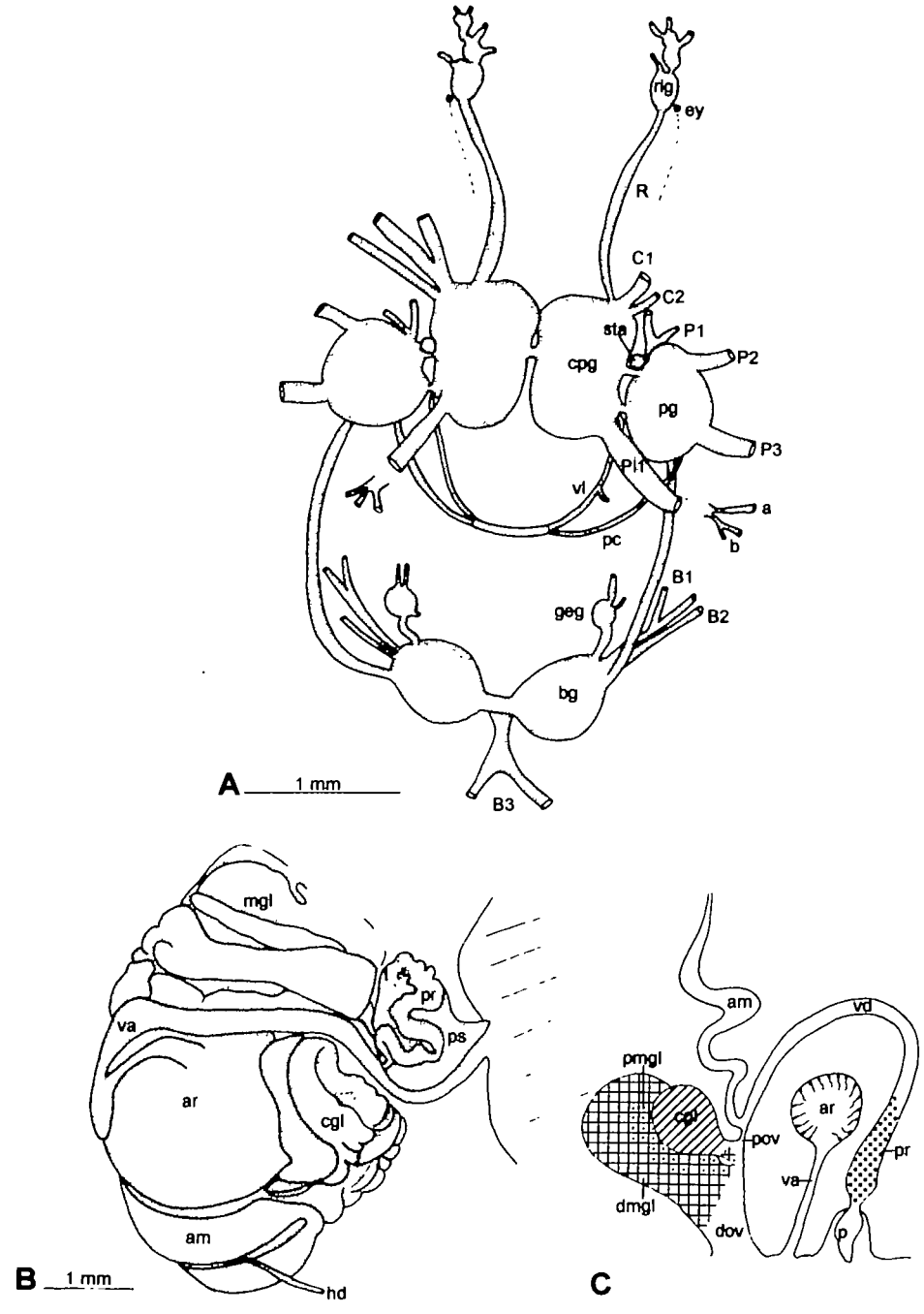


Figure 3. *Armina neapolitana*— Anatomy. **A.** Nervous system. **B.** Anterior genital complex *in-situ*. **C.** Schematic outline of genital complex. Abbreviations: am, ampulla; ar, allosperm receptacle; bg, buccal ganglion; cgl, capsule gland; cpg, cerebropleural ganglion; dmgl, distal mucus gland; dov, distal oviduct; ey, eye; geg, gastro-oesophageal ganglion; hd, hermaphrodite duct; mgl, mucus gland; p, penis; pc, pedal commissure; pg, pedal ganglion; pmgl, proximal mucus gland; pov, proximal oviduct; pr, prostate; rig, rhinophoral ganglion; sta, statocyst; va, vagina; vd, vas deferens; vl, visceral loop.

are surrounded by a thick layer of connective tissue and contain nerve cells of different sizes (as all other ganglia do, too). The nerve cells are located mainly in the periphery of the ganglia. The pedal ganglia are spherical (0.57 × 0.55 mm) and lie close to the outer sides of the cerebropleural ganglia. There are two connectives between the pedal and cerebropleural ganglia. A long visceral loop with the visceral nerve originates on the right side. The pedal commisure, visceral loop and probably also the parapedal commissure run in a common connective sheath, and therefore the parapedal commissure is not visible. There is a single connective between the cerebropleural and buccal ganglia. The latter (0.5 × 0.6 mm) lie underneath the oesophagus just behind its opening into the pharynx. The gastro-oesophageal ganglia are small, lying anteriorly to the buccal ganglia upon the pharynx. The rhinophoral ganglia are located at the base of the rhinophores. The eyes lie close to the base of the rhinophoral ganglia but are not innervated by them. The eyes are formed by a nearly spherical lens with a diameter of approximately 40 μm and a layer of black pigment. No optical ganglia are visible. The statocyst is spherical to elliptical and located close to the cerebropleural-pedal connective. It is lined by an epithelium with large ciliated cells containing granules. Few small statocones of different sizes are also present (Fig. 5C).

The following innervation areas could be identified (see Fig. 3A): R: rhinophores; C1: oral tube; C2: right/left margin of oral veil; Pl1: a: dorsal notum; b: caudal body region; P1: frontal part of foot; P2: lateral part of foot; P3: caudal part of foot; B1: pharynx; B2: pharynx B3*: radula sheath

Reproductive system: The gonad is compact, large, and subdivided into several follicular bundles, lying on top of the central canal of the digestive gland (Fig. 2A). The follicles contain oogonia and spermatogonia, the latter are located in the centre of a follicle, the oogonia in the periphery (Fig. 6C). A ciliated hermaphrodite duct runs along the ventral side of the gonad, leading anteriorly towards a tube-like, coiled ampulla (Fig. 3B), which is lined by a flat epitelium partly containing large vacuoles. The ampulla is filled with autosperm. In its distal part the epithelial cells are elongate and the nuclei become visible. Some cells are ciliated. The ampulla contains more sperm in this region. A heavily ciliated postampullary duct lies within the mass of the mucus gland. The duct is surrounded by a thick muscular layer and divides into the vas deferens and the proximal oviduct. The vas deferens has few coils, and runs towards the right side of the genital complex, entering the penial sheath dorsally. The proximal vas deferens consists of ciliated cuboidal cells containing large nuclei. It is filled with sperm. A prostate is present (Fig. 6B). It is coiled and consists of blue-staining (toluidine blue) cylindrical glandular cells which alternate with ciliated supporting cells. The penis is muscular and unarmed. The proximal oviduct is very short and lined by cylindrical cells with long cilia containing few vacuoles and a basal nucleus. It enters the white capsule gland, which has narrow coils and is smaller than the mucus gland. The cells of the capsule gland stain blue. They are elongated, cylindrical. The glandular cells contain dark blue granules and a basal nucleus. They alternate with slender, ciliated supporting cells which contain an apical nucleus. The mucus gland is very large, yellowish in colour, with large coils. Histologically it can be divided into two parts: the proximal mucus gland and the distal mucus gland. The cells of the proximal mucus gland are pink with heterogeneous contents and heavily ciliated. The glandular cells alternate with supporting cells. The distal mucus gland shows different parts representing different functions (according to Schmekel 1971): a) light blue-staining cells with pygnotic nuclei representing 'old' areas of the gland (the secretions of the cells are emptied out); b) long, slender cells with dark violet vesicles, cilia are not visible; these are 'active' zones of the gland; c) pink cells, partly filled with vesicles, partly no granules visible, instead the secretions form irregular fibers; this is the 'Kleberegion' (Schmekel 1971: 134). The allosperm receptacle is spherical, lying at the right side of the genital complex. It is composed of a thick, folded epithelium consisting of cuboidal to cylindrical cells. Nuclei are hardly visible. It is packed with sperm, which are found in two different arrangements: in the periphery the heads of the sperm are directed towards or even embedded in the epithelial cells. In the centre lies a mass of irregularly arranged sperm (Fig. 6D). The receptacle opens via a long vagina into the genital papilla. A schematic outline of the distal genital system is shown in Fig. 3C.

Heart and circulatory system: The pericardium was damaged during preparation. It is composed of a very thin epithelium. Cells are hardly visible, only the nuclei are distinct. The atrium consists of a thin, delicate muscular tissue, lying to the left of the muscular ventricle. The latter is located in the anterior region of the gonad and partly covers it (Fig. 2A). The main aorta proceeds from the ventricle in a ventral direction.

Excretory system: The kidney covers the gonad. It is composed of a delicate tissue consisting of highly vacuolized cells and blue-staining granules. Cells with larger nuclei and a distinct nucleolus are interspersed. The renopericardial duct is lined with cuboidal cells bearing long cilia. Some cells contain a vacuole and few small, blue-staining vesicles.

Discussion

Morphology: Previous morphological descriptions of *Armina neapolitana* are very uniform. The colour of the living animal is described as yellow to light brownish with white ridges on the notum and a pale grey background by Pruvot-Fol (1937) and Schmekel & Portmann (1982). Pruvot-Fol (1937) interprets the small elevation on which the rhinophores are standing as the caruncle. This does not correspond to

*. nerve is not symmetrical

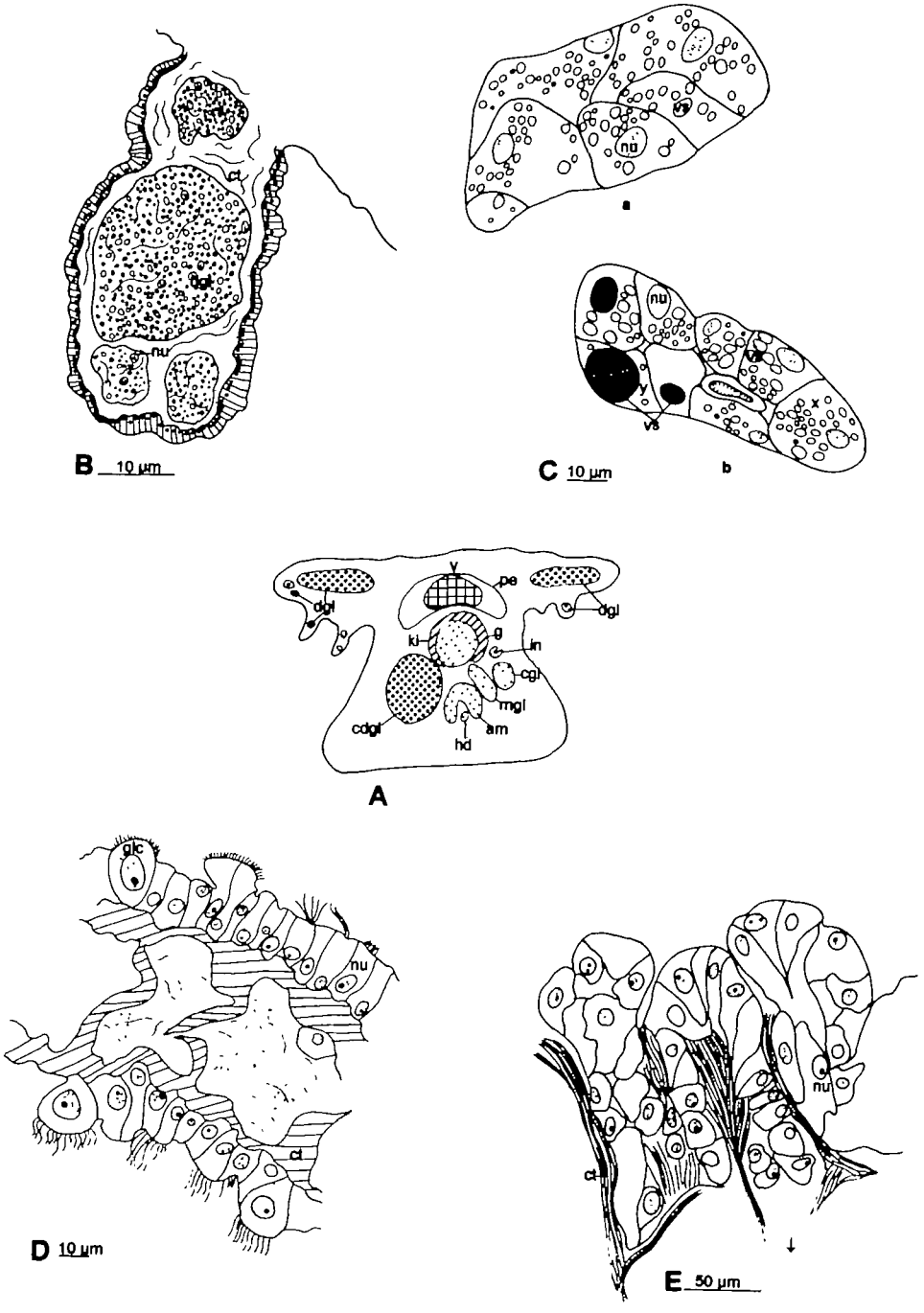


Figure 4. *Armina neapolitana*—Histology. **A.** Schematic drawing of cross section of animal in region of anterior genital complex. **B.** Cross section of hyponotal lamellae. **C.** Follicles of salivary gland (**a**) and oral gland (**b**). **D.** Section of branchial lamellae. **E.** Cross section of pedal gland, arrow: direction of foot epithelium. Abbreviations: am, ampulla; cdgl, central canal of digestive gland; cgl, capsule gland; ct, connective tissue; dgl, digestive gland; g, gonad; glc, gland cell; hd, hermaphrodite duct; in, intestine; ki, kidney; mgl, mucus gland; nu, nucleus; pe, pericardium; v, ventricle; vs, vesicle; x, cell type 1; y, cell type 2.

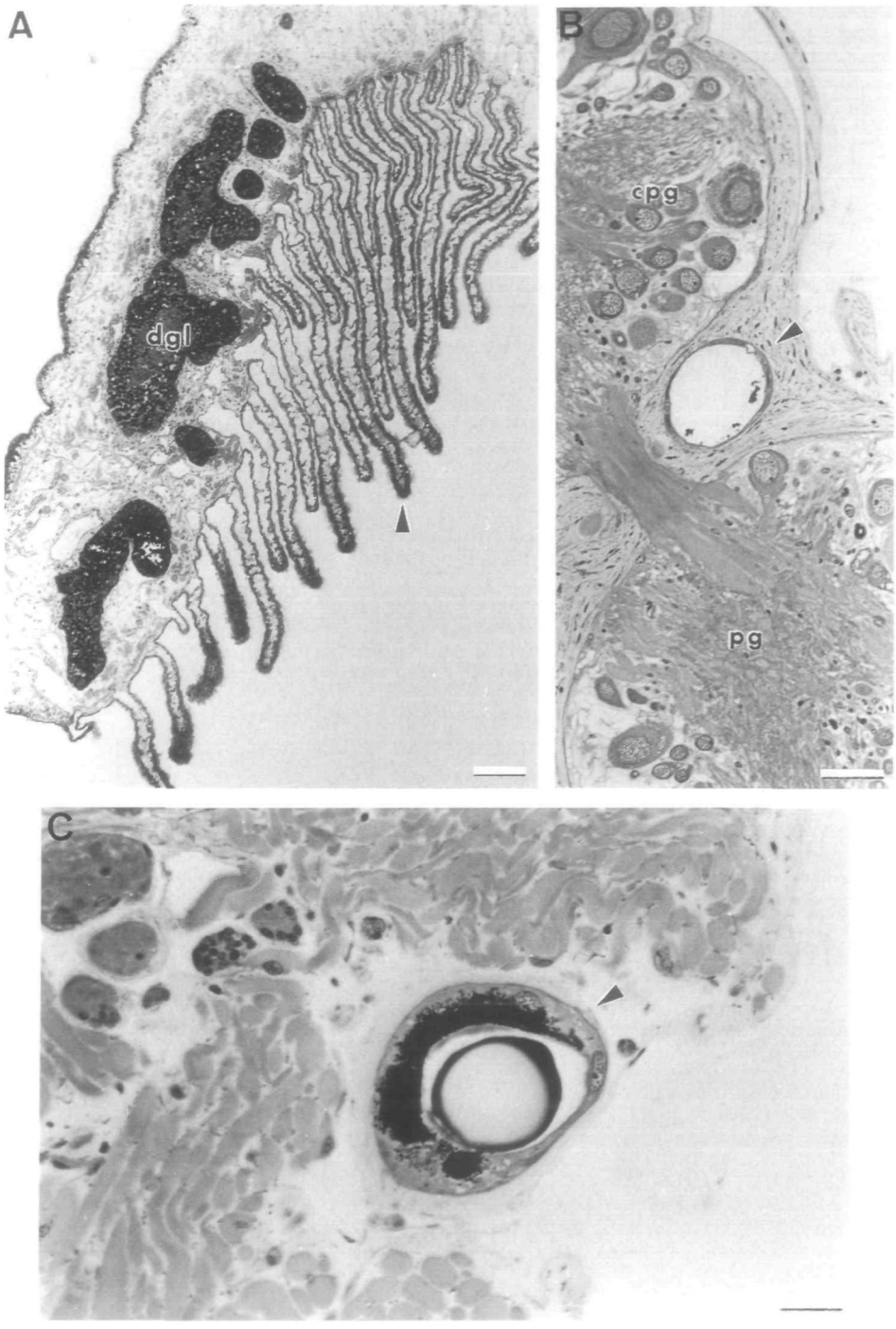


Figure 5. *Armina neapolitana*—Histology. **A.** Cross section of lateral notum with branchial lamellae (arrow); scale bar = 200 μm. **B.** Cross section of ganglia with statocyst (arrow); scale bar = 50 μm; **C.** Cross section of eye (arrow); scale bar = 25 μm. Abbreviations: cpg, cerebropleural ganglion; dgl, digestive gland; pg, pedal ganglion.

Bergh's (1866) original description, nor to the results of this study. Here the caruncle is always found in front of the rhinophores. But one must take into account that the shape of the caruncle may be influenced by fixation and therefore is not valuable for distinguishing species.

Anatomy: The shape of the radula teeth is species-specific in *Armina*. Schmekel & Portmann (1982) mention 0–7 small denticles on the outside of the marginal tooth of *A. neapolitana*, but the marginal teeth of the animal studied here are smooth. Pruvot-Fol (1937) presents a detailed description of the digestive system of *Armina neapolitana*. The author mentions a conically shaped glandular region of the posterior central canal of the digestive gland. This could not be verified by anatomical nor by histological means. Histologically, the epithelium of the posterior section of the central canal differs from the rest only in terms of shape and ciliation of the cells. No glandular cells were found. According to Pruvot-Fol (1937), the arrangement of the branches of the digestive gland is very dense. This is not consistent with the findings of this study, either. Especially in comparison to *A. maculata* and *A. tigrina*, the follicles of the digestive gland of *A. neapolitana* seem packed rather loosely. The author also mentions branches of the digestive gland reaching into the branchial lamellae. As Hoffmann (1940) has already discussed, this cannot be confirmed. The results of this study confirm Bergh's findings regarding the presence of one pair of salivary glands and one pair of oral glands, the latter not detected by Pruvot-Fol (1937).

Pruvot-Fol (1937) assigned *Pleurophyllidia undulata* Bergh, 1866 to *Armina neapolitana*. But according to Bergh's drawings, especially of the radula teeth, this synonymy must be re-evaluated for the shape of the radula teeth indicates a synonymy with *Armina tigrina*.

In the following species only differences in the morphology, anatomy and histology from *Armina neapolitana* are described.

Armina maculata Rafinesque, 1814

Synonymy:

Diphyllidia ocellata Deshayes, 1830
Diphyllidia verrucosa Cantraine, 1835
Diphyllidia pustulosa Schultz in Philippi, 1836
Pleurophyllidia verrucosa Bergh, 1866 (non Cantraine)
Pleurophyllidia ocellata Bergh, 1866 (non Deshayes).
Pleurophyllidia pustulosa Bergh, 1866 (non Schultz-Philippi)

External morphology:

In contrast to *Armina neapolitana*, no notal ridges but irregularly arranged pustules occur on the notum. The living animals have white pustules; the ground colour of the mantle is orange; the foot white. Size and shape of the pustules vary in the preserved specimens according to their position on the notum. Spherical to conical pustules of different sizes are found in the frontal part (Fig. 8A); the number of these pustules decreases towards the medial part of the body and increases again in the posterior part. Large and conical pustules are missing in the posterior part (Fig. 8B). The oral veil is not distinct, but appears as a slender bulge. The rhinophores (only visible in specimen No. 1) are similar in shape to *A. neapolitana*, short, club-shaped, but the tip of the rhinophore is more rounded than in *A. neapolitana*. The left rhinophore bears ten vertical lamellae, the right nine ones. At the lateral sides a few spots of black pigment are present. The caruncle is indistinct and not easily distinguishable from the oral veil in the preserved specimens.

The position of the genital opening, the nephroporus and the anus are the same as described in *A. neapolitana* (Fig. 8E). The number and arrangement of the branchial lamellae are different from the former species: the number of the primary and secondary lamellae is up to 61–80 on each side; the number of the transversely arranged hyponotal lamellae is also higher; 25–34 lamellae were counted in a juvenile animal, up to over 100 hyponotal lamellae per side in an adult specimen from the MNH Paris (Fig. 8E).

The foot narrows towards the posterior end where a white pedal gland is visible (Fig. 8C). On the histological sections a few marginal sacs were found in the lateral notal margin. Some of them represent prestages (Fig. 12A). Mature sacs are lined by pavement cells containing large nuclei. The lumen is filled with large masses of violet-staining (toluidine blue) secretions. The sacs do not have a connection to the digestive gland. Some ejected sacs are visible. These have a collapsed and contracted epithelium. Here no secretions are visible (Fig. 12B).

The epidermis of *Armina maculata* is composed of 'Spezialvakuolenzellen' (Schmekel 1982), but more glandular cells occur around the oral tube and along the foot than in *A. neapolitana*. Around the oral tube and the oral veil the epidermal cells are heavily ciliated.

Anatomy

Digestive system: The oral tube is shorter than in *Armina neapolitana*. Two types of subepidermal glandular cells can be distinguished around the oral tube: a) large cells containing light pink-staining cytoplasm and a dark, centrally lying nucleus; b) smaller cells with blue to violet cytoplasm, containing light granules. The position and shape of the oral glands (Fig. 9B) is the same as described for *A. neapolitana*. The pharynx is 14 mm in length and 9 mm in breadth (9 mm × 8 mm in smaller individual). The jaws are very large and strong, with a hook-like masticatory process bearing few rows of denticulations (Fig. 9E, 13C, D). The masticatory borders are also denticulated. The radula formula are 46 × 39.1.1.1.39 (63 mm specimen) and 38 × 34.1.1.1.34 (35 mm

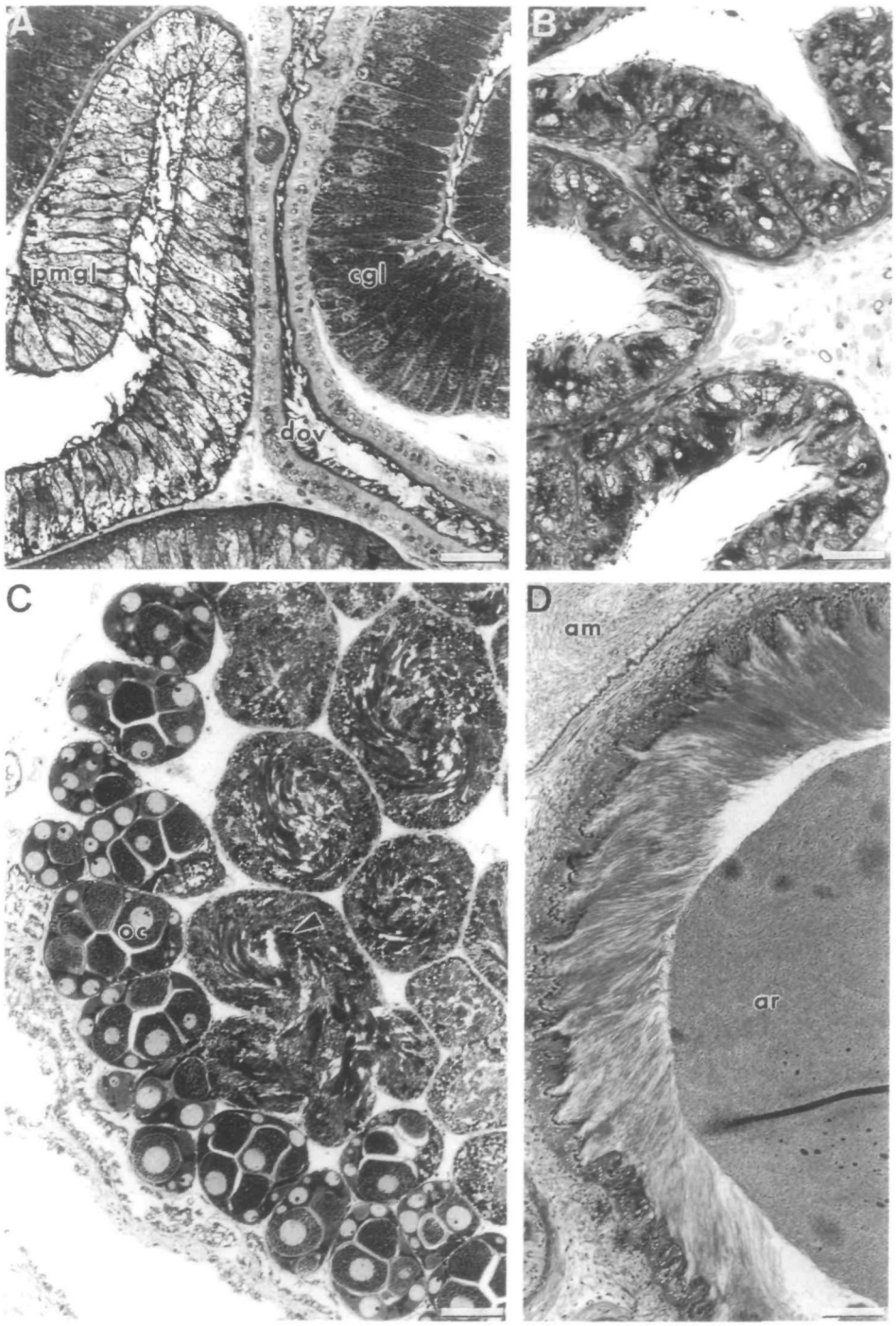


Figure 6. *Armina neapolitana*—Histology of genital complex. **A.** Cross section through female glands; scale bar = 50 μm. **B.** Prostate; scale bar = 50 μm. **C.** Gonad; scale bar = 100 μm. **D.** Allosperm receptacle; scale bar = 100 μm. Abbreviations: am, ampulla; ar, allosperm receptacle; cgl, capsule gland; dov, distal oviduct; oc, oocytes; pmgl, proximal mucus gland.

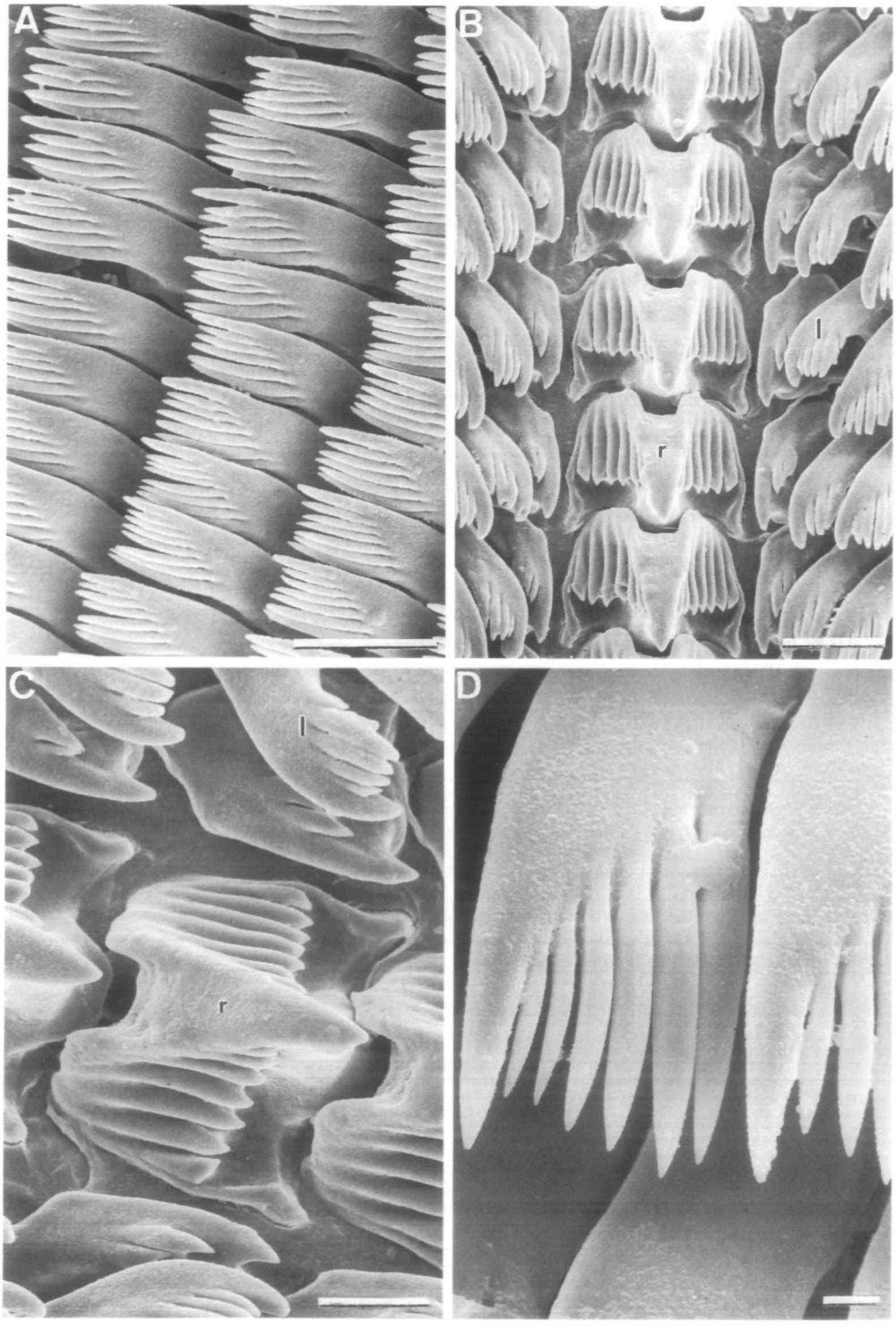


Figure 7. *Armina neapolitana*—SEM-photographs. **A.** Lateral teeth; scale bar = 50 μm. **B.** Rachidian teeth and few laterals; scale bar = 50 μm. **C.** Rachidian tooth; scale bar = 25 μm. **D.** Lateral tooth; scale bar = 5 μm. Abbreviations: l, lateral tooth; r, rachidian tooth.

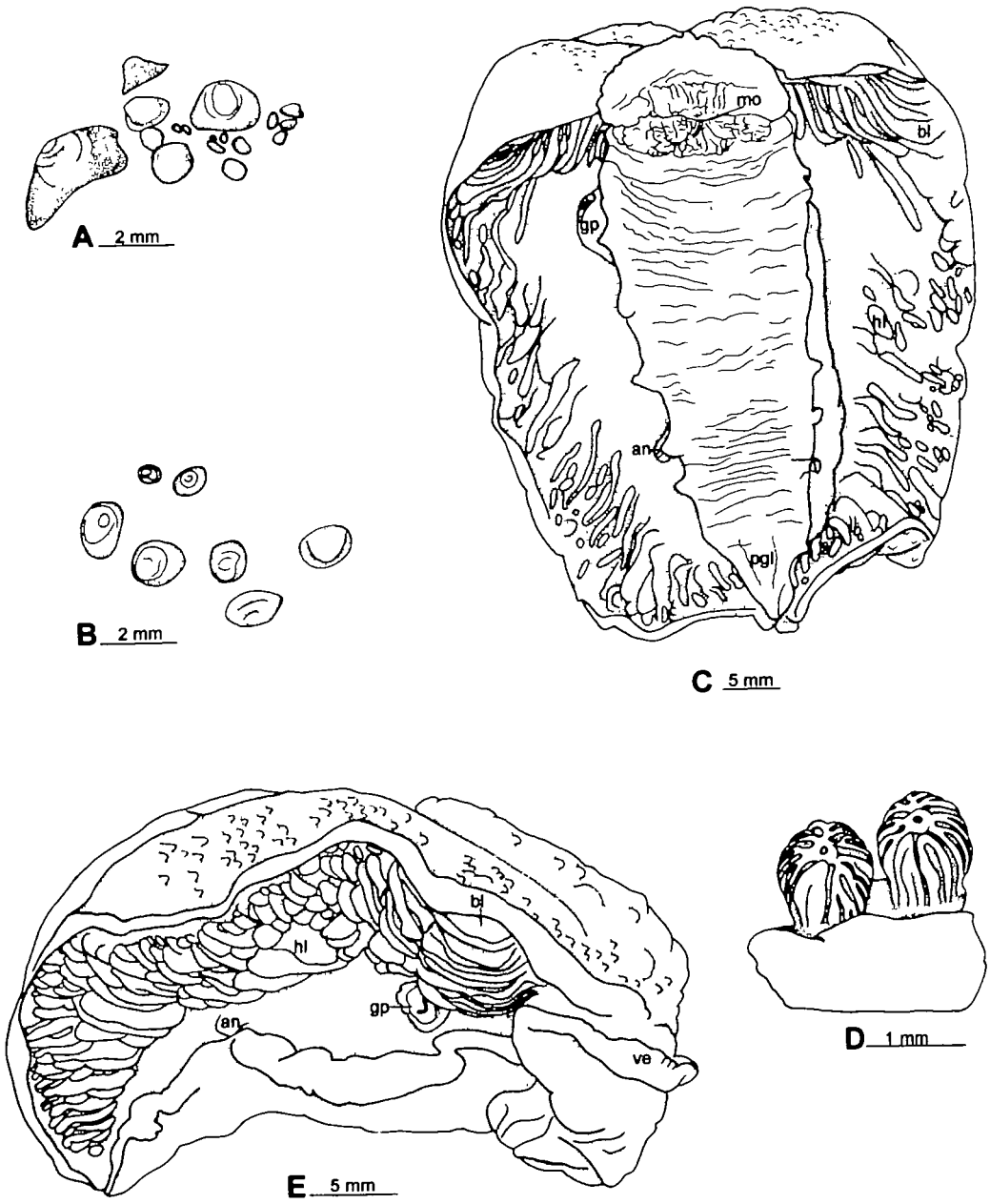


Figure 8. *Armina maculata*. **A.** Notal pustules from anterior body region. **B.** Notal pustules from lateral body region. **C.** Ventral view (No. 4). **D.** Rhinophores. **E.** Lateral view (No. 3). Abbreviations: an, anus; bl, branchial lamellae; gp, genital papilla; hl, hyponotal lamellae; mo, mouth opening; pgl, pedal gland; ve, oral veil.

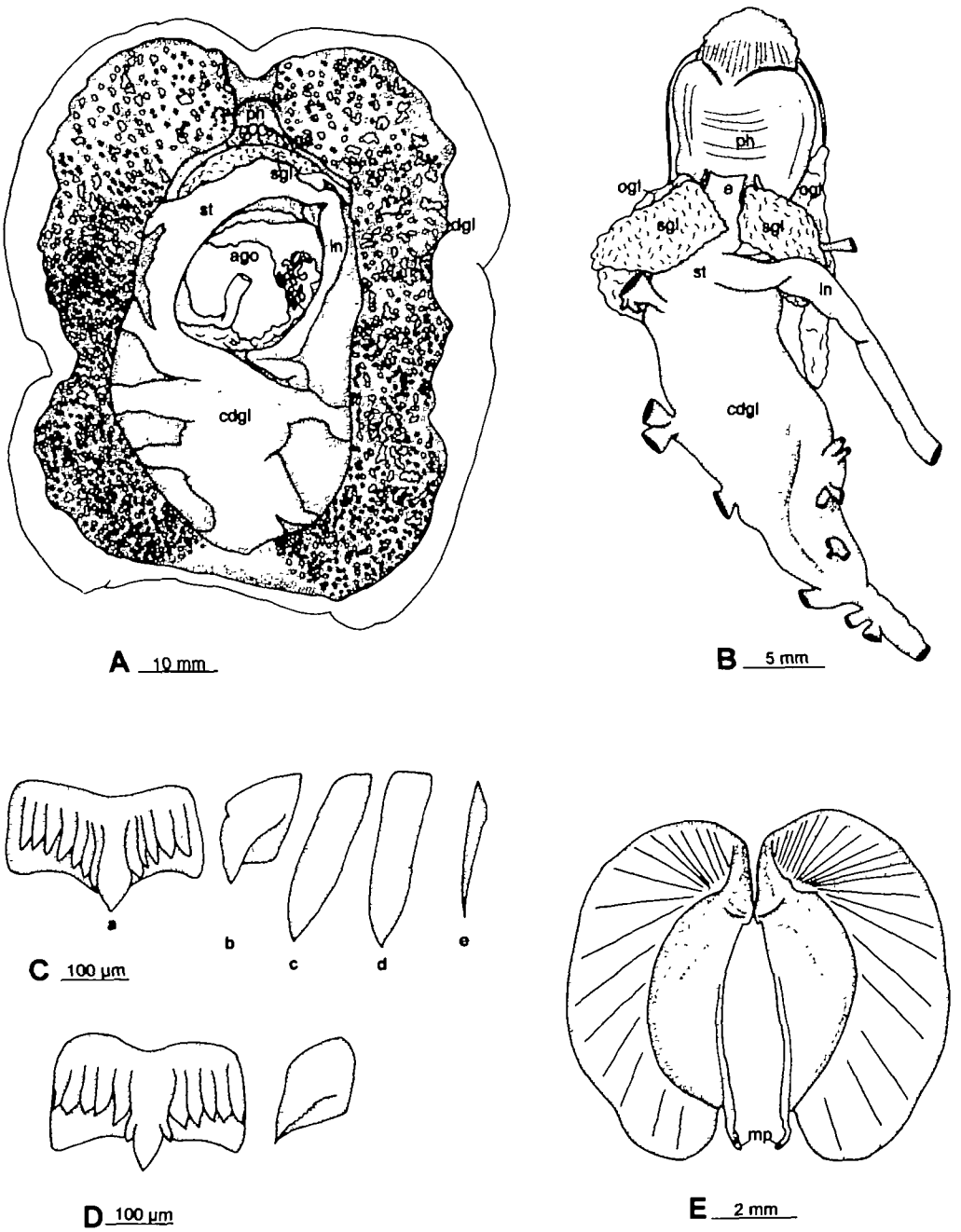


Figure 9. *Armina maculata*—Anatomy. **A.** Dorsal view after removal of notum, pericardium, heart and gonad. **b.** Digestive system without glandular part of digestive gland. **C.** Radula teeth (63 mm specimen), **a.** Rachidian tooth, **b–d.** laterals 1–3, **e.** marginal tooth. **D.** Rachidian tooth and first lateral (35 mm specimen). **E.** Jaws. Abbreviations: ago, anterior genital complex; cdgl, central canal of digestive gland; dgl, digestive gland; e, oesophagus; ga, ganglia; in, intestine; mp, masticatory process; ogl, oral gland; ph, pharynx; sgl, salivary gland; st, stomach.

specimen). The rachidian tooth is very broad (280 μm) and bears 5–6 denticles on both sides of median cusp (Fig. 9Ca, 13A), the latter being shorter than that of *Armina neapolitana*. The first lateral tooth has a broad base and is denticulated on the outer side (Fig. 9D) in the 35 mm individual. The remaining laterals are smooth (Fig. 13B). The outermost lateral (Fig. 9Ce) is very slender in contrast to the others. The salivary glands are similar to those described for *Armina neapolitana* (Fig. 9B). They are very large, covering the whole posterior part of the pharynx and oesophagus (Fig. 9B). The oesophagus originates at the posterior part of the pharynx, enlarging soon to a small stomach. The epithelium of the stomach is thinner than that of the oesophagus and contains more vacuoles. Some of these are filled with blue secretions. The digestive gland and the intestine have the same position as in *Armina neapolitana* (Fig. 9A). The intestine shows a typhlosole on the dorsal side on 2/3 of its length. The central canal of the posterior left digestive gland is broad at its origin, narrowing towards the caudal part of the animal (Fig. 9B). Four lateral tubes originate at the right side of the central canal, six at the left. The glandular part of the digestive gland is restricted to the lateral parts of the body. The follicles of the digestive gland appear very compact in contrast to the former species. In addition to the two cell types described in *Armina neapolitana*, a third cell type was identified in the follicles: small cells with small dark vacuoles.

Nervous system: The position of the ganglia on the dorsal side of the pharynx is the same as in *Armina neapolitana*. The cerebropleural ganglia are 1.5 mm in length and 0.9 mm in breadth and lie close together (Fig. 10A). The pedal ganglia lie at the sides of the cerebropleural ganglia. They are smaller and almost spherical (1.2 × 0.9 mm). A long pedal commissure is present. The parapedal commissure is probably hidden in the connective tissue. The buccal ganglia are ovoid shaped and have approximately the same size as the pedal ganglia. Two small gastro-oesophageal ganglia lie next to the buccal ganglia. No visceral loop could be identified. But in the larger animal one small ganglion was found directly underneath nerve Pl1 (originating from the right pleural ganglion), and this ganglion is assumed to be the visceral ganglion (not shown in Fig. 10A). The rhinophoral ganglia, the eye and the statocyst have the same position as in *A. neapolitana*. Fig. 10A shows a general outline of the nervous system with the identified nerves and their innervation: R: rhinophores; C1 + C2: running along the pharynx towards the oral tube and veil; C3: oral veil; Pl1: lateral margin of mantle; P1: frontal part of foot; P2: lateral part of foot; P3: foot; B1: pharynx; B3*: radula sheath.

Reproductive system: The position and shape of the genital complex are similar to *Armina neapolitana* (Fig. 10B). One juvenile and one adult specimen were examined by histological means. The differentiation of all male and female parts of the genital system is not yet complete in the juvenile; only the ampulla, the oviduct, the vas deferens and the female glands are distinguishable. No differentiation of the female gland mass in capsule gland and mucus gland is visible. The gonad is very small. Only small follicles are present, containing oogonia, spermatogonia and immature gametes. A schematic outline of the genital system of the adult specimen is shown in Fig. 10C. The gonad is very large (length: 30 mm), compact, and consists of numerous follicles. It is located on top of the central canal of the digestive gland. The hermaphrodite duct widens to a long, heavily coiled, tube-like ampulla. The latter runs far into the mass of the female glands. It contains only few sperm. Its lining epithelium is composed of cylindrical cells with large nuclei. The cells are always ciliated. Each cell contains few vacuoles, their number decreasing in the distal part of the ampulla in proximity to the postampullary duct. The latter is hardly distinguishable from the ampulla for the ampulla narrows only a little at this point. The duct divides into the proximal oviduct and the vas deferens at the inner anterior edge of the capsule gland. The vas deferens is long, and leads into the prostate close to the genital opening. The prostate can be distinguished from the vas deferens only by histological means. Its epithelium is folded (Fig. 11C) and dark violet-staining. Within the penial sheath lies the distal vas deferens, which is lined by light blue-staining, cylindrical cells bearing long cilia.

The proximal oviduct is very short, leading into the tissue of the capsule gland. The latter is smaller than the mucus gland and has narrow coils. The epithelial cells of the latter are slender, cylindrical, dark blue in colour and contain numerous granules of the same colour (Fig. 11B). Nuclei are hardly visible. The capsule gland does not show a direct connection to the mucus gland, but discharges into the distal oviduct, which connects to the proximal and distal mucus gland. In the proximal part of the mucus gland the cells do not contain granules or vesicles and appear white (Fig. 11A). The cells bear long cilia. More distally, the cells are similar in shape and colour to those described for the distal mucus gland in *Armina neapolitana*. The distal mucus gland is large and cream white in colour. The distal oviduct is very long and embedded in coils of the mucus gland. It is lined by cuboidal cells with long cilia. The allosperm receptacle is located on the right side of the genital complex. In the adult specimen it does not contain any sperm, so the animal had probably not yet copulated before collection. The lining epithelium of the allosperm receptacle is folded. The cuboidal cells do not bear any cilia and do not contain vacuoles. The vagina originates as a broad tube at the posterior end of the receptacle. It is lined by a heavily folded epithelium consisting of cuboidal cells bearing long cilia. It is also surrounded by a muscle layer and connective tissue.

Heart and circulatory system: The position of the ventricle and the atrium are the same as in the

*: nerve is not symmetrical

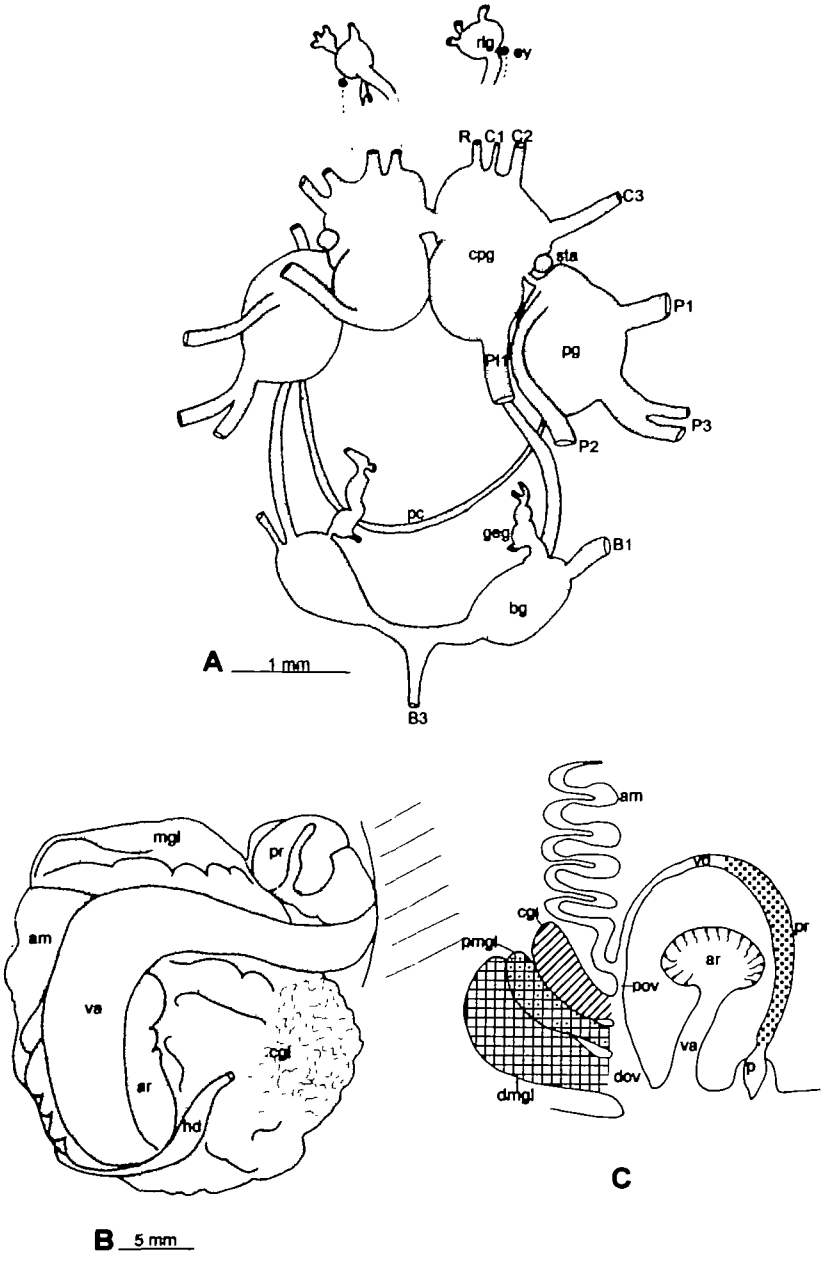


Figure 10. *Armina maculata*—Anatomy. **A.** Nervous system. **B.** Anterior genital complex *in situ*. **C.** Schematic outline of genital complex. Abbreviations: am, ampulla; ar, allosperm receptacle; bg, buccal ganglion; cgl, capsule gland; cpg, cerebropleural ganglion; dmgl, distal mucus gland; dov, distal oviduct; ey, eye; geg, gastroeosophageal ganglion; hd, hermaphrodite duct; mgl, mucus gland; p, penis; pc, pedal commissure; pg, pedal ganglion; pmgl, proximal mucus gland; pov, proximal oviduct; pr, prostate; rig, rhinophoral ganglion; sta, statocyst; va, vagina; vd, vas deferens.

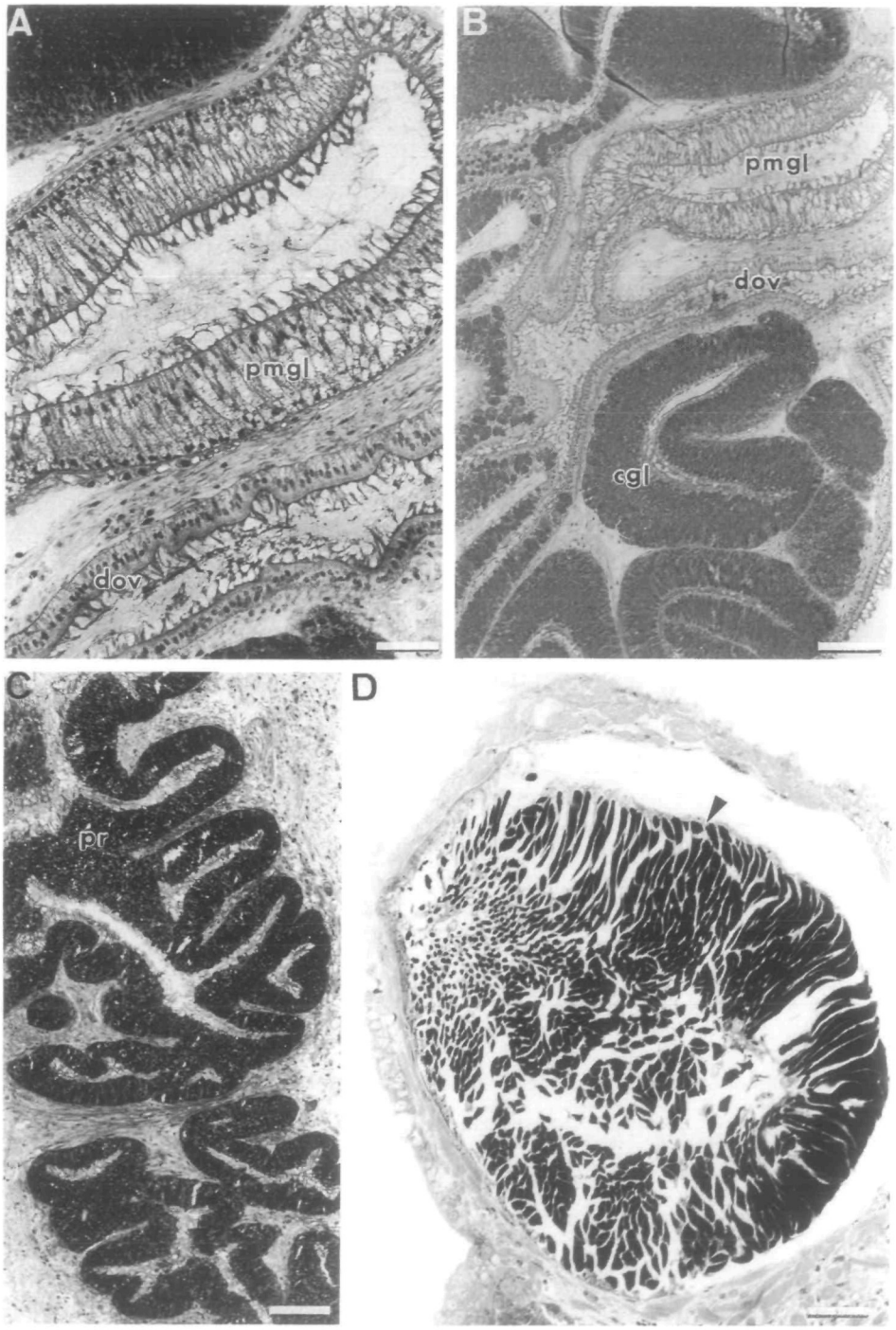


Figure 11. **A.–C.** *Armina maculata*. **D.** *Armina loveni*. **A.** Cross section of female glands; scale bar = 50 μm. **B.** Cross section of female glands; scale bar = 100 μm. **C.** Cross section of prostate; scale bar = 100 μm. **D.** Cross section of marginal sac (arrow: secretions); scale bar = 100 μm. Abbreviations: cgl, capsule gland; dov, distal oviduct; pmgl, proximal mucus gland; pr, prostate.

former species. The ventricle is partly covered by the atrium. The main aorta originates from the ventricle and runs ventrally in between the gonad and the intestine in direction of the anterior genital complex. The posterior artery lies on top of the gonad, sending branches to each side of the gonad and to the surrounding kidney tissue.

Excretory system: The syrinx consists of strong and folded tissue. The kidney covers the large gonad like a sac as described in *Armina neapolitana*. Further caudally, the ureter leads from the lateral kidney tissue through the follicles of the digestive gland to the right side of the body.

DISCUSSION

Anatomy: The digestive system, illustrated by Ballesteros (1983) is generally consistent with the one described in this study. But it is remarkable that the author does not mention any oral glands, since these are very voluminous organs in *A. maculata*. Garcia & Garcia-Gomez (1990a) have described oral glands in *A. maculata* previously. Their anatomical findings about the postion of the glands and the efferent ducts can be confirmed. The description of the radula of *A. maculata* is very uniform. Nevertheless, the aberrant smooth first lateral tooth, described for one specimen here, differs from other descriptions by Bergh (1866–67) and Thompson *et al.* (1990). According to those authors, the first lateral tooth is always slightly denticulated. Pruvot-Fol (1937), in contrast, writes that the laterals can also be smooth, but it is unclear whether this also applies to the first lateral.

Bergh's (1866–67) anatomical figures of *A. maculata* correspond to a great extent with the results of my examinations. Differences occur in the description of the circulatory system. The author interprets a blood vessel, described here in accordance with Garcia & Garcia-Gomez (1990b) as the posterior artery on the dorsal side of the gonad as an 'Urinkammeret' (bladder). Bergh misinterprets the vessels, originating from the sides of the artery, as openings into the kidney tissue ('. . . Aabninger ud i Nyrevaevet'). According to Garcia & Garcia-Gomez (1990b) the kidney ('renal chamber') of *A. maculata* is a continuous canal running on the ventral side of the gonad and being connected to the posterior artery by lateral vessels. This contradicts the results presented here. Garcia & Garcia-Gomez (1988) thoroughly investigated the nervous system of *A. maculata*. A few differences of their findings from my studies need to be mentioned: I have not found a nerve rising from the rhinophoral nerve and innervating the caruncle. The genital ganglion, mentioned by the authors, has not been found in the present examination, either. Moreover, the position of the

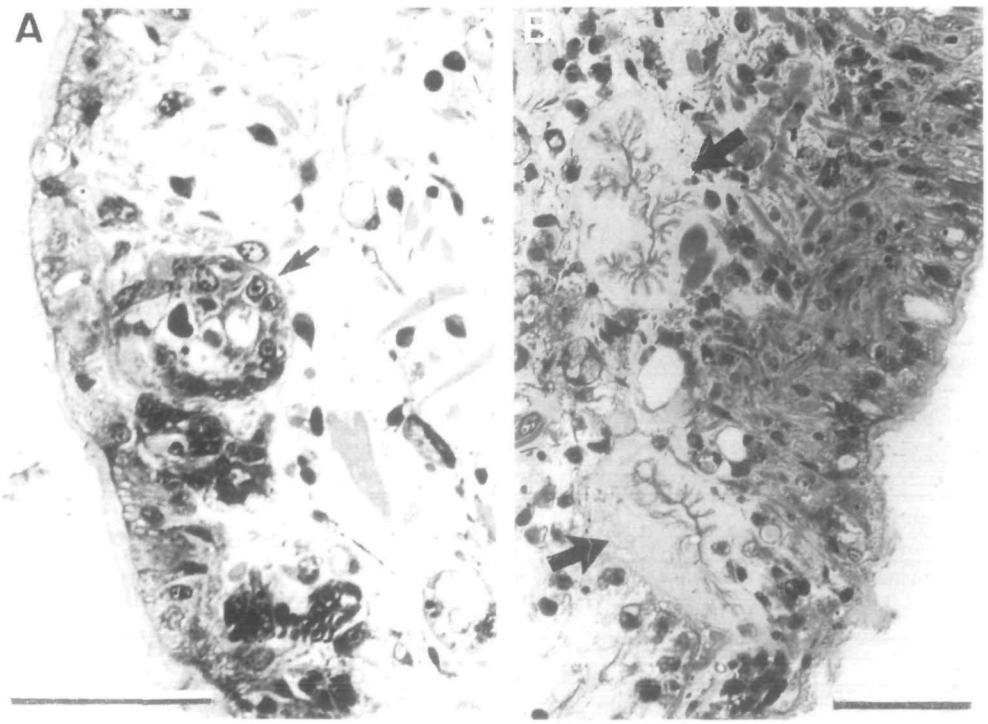


Figure 12. *Armina maculata*. **A.** Prestage of marginal sac (arrow); scale bar = 100 μm. **B.** Empty marginal sacs, collapsed (arrow); scale bar = 100 μm.

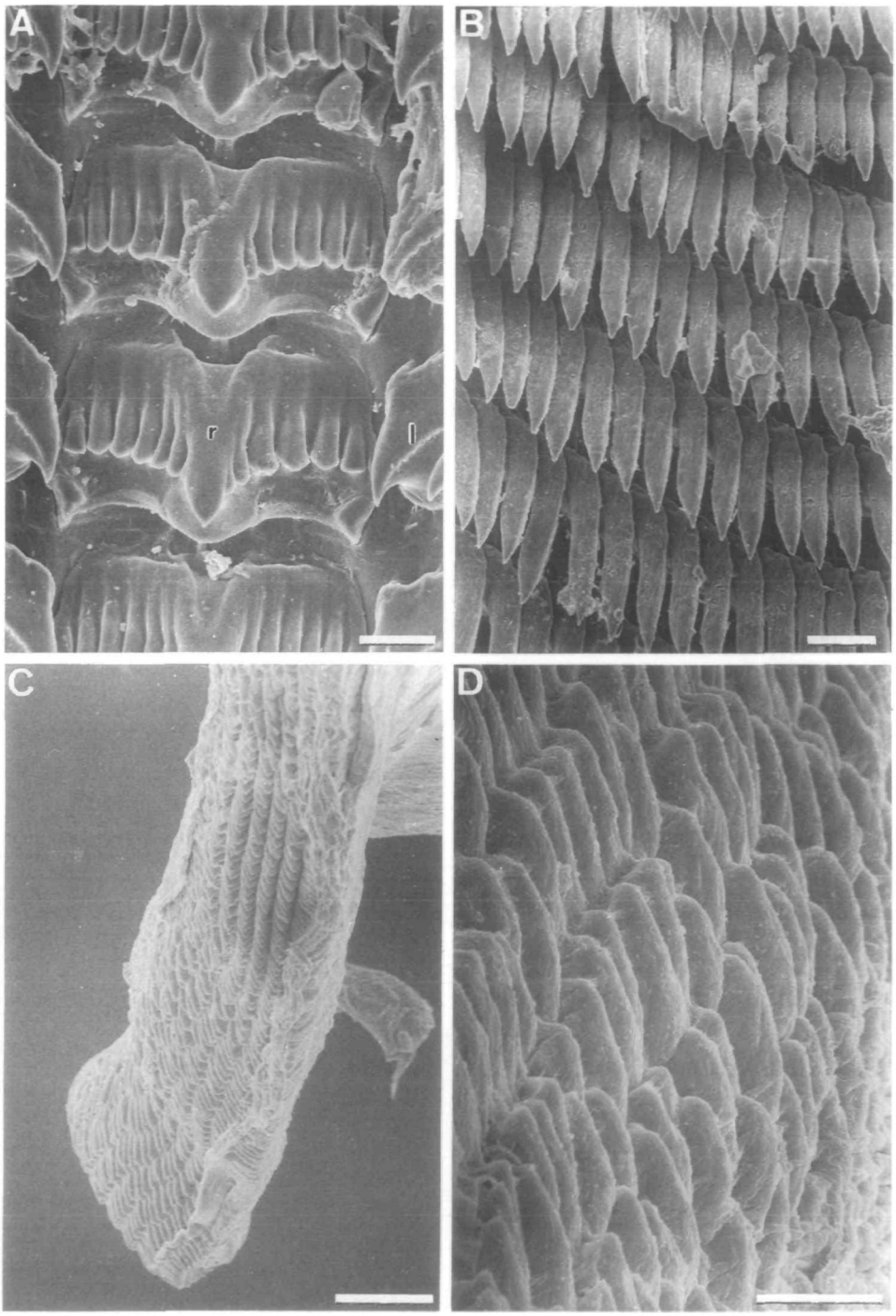


Figure 13. *Armina maculata*—SEM-photographs. **A.** Rachidian teeth and first laterals; scale bar = 50 μm. **B.** Lateral teeth; scale bar = 100 μm. **C.** Masticatory process; scale bar = 100 μm. **D.** Denticles on masticatory process; scale bar = 25 μm. Abbreviations: l, lateral tooth; r, rachidian tooth.

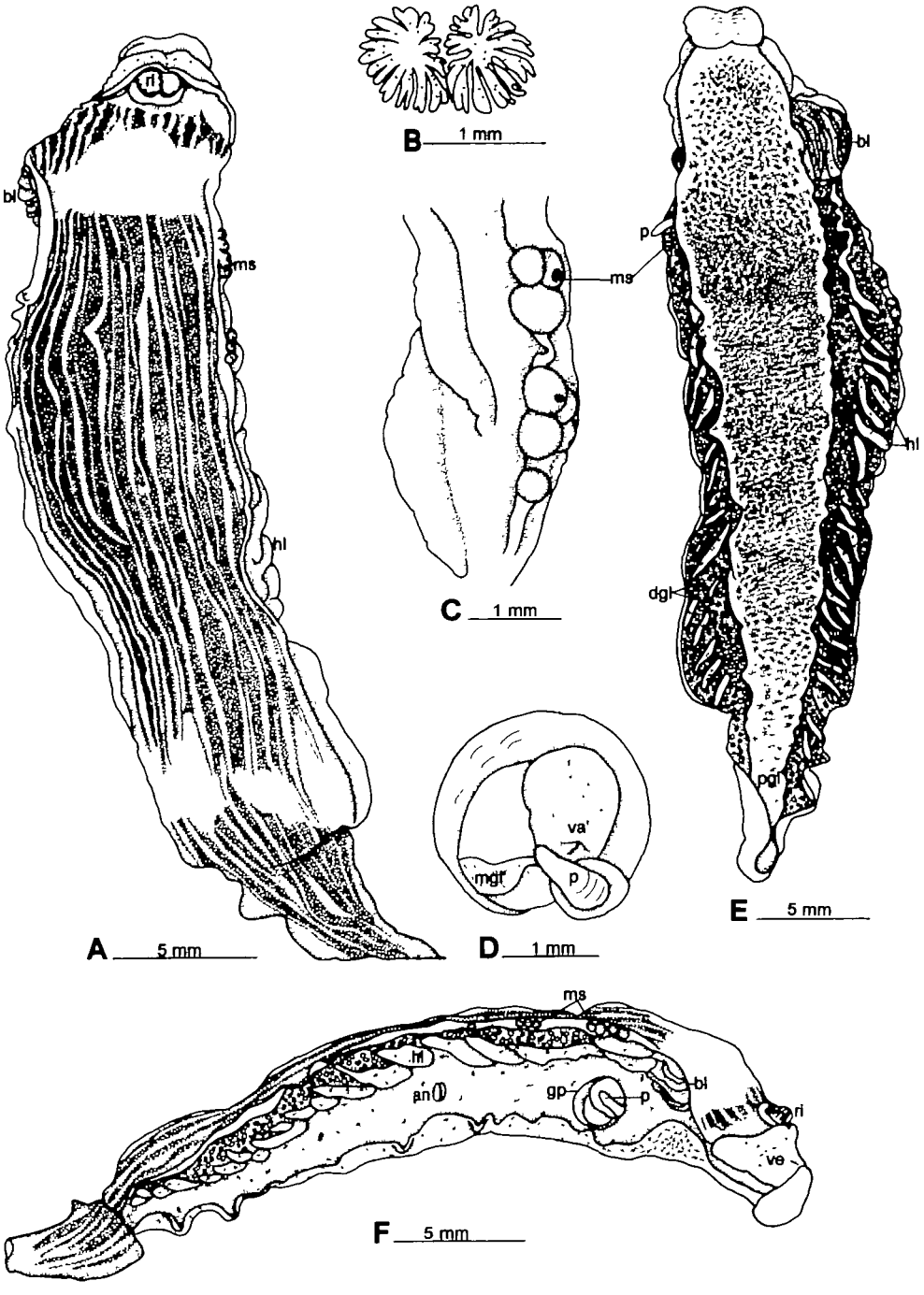


Figure 14. *Armina loveni*. **A.** Dorsal view. **B.** Dorsal view of rhinophores. **C.** Mantle brim with marginal sacs. **D.** Genital papilla. **E.** Ventral view. **F.** Lateral view. Abbreviations: an, anus; bl, branchial lamellae; dgl, digestive gland; gp, genital papilla; hl, hyponotal lamellae; mgl, mucus gland; ms, marginal sac; p, penis; pgl, pedal gland; ri, rhinophores; va, vaginal; ve, oral veil.

eye is described here next to the rhinophoral ganglion, not on the right anterior corner of the cerebopleural ganglion, giving a sessile impression. The distal genital complex is described by Ballesteros (1983) rather superficially. The *in situ* drawings shown in Fig. 4J and 4K are incomplete and not precise. According to the author the whole posterior part of the female gland belongs to the capsule gland. In the present study the capsule gland is described only as a small section of the female gland, while the mucus gland fills the bulk part of it. The author does not report of a prostate gland either. Histologically, however, this structure could be distinguished from the non glandular part of the vas deferens.

Armina loveni (Bergh, 1866)

Synonymy:

Pleurophyllidia loveni Bergh, 1860, nomen nudum
Pleurophyllidia loveni Bergh, 1866
Pleurophyllidia lineata 'Otto, 1820' Bergh, 1866 (misidentification, not *Diphyllidia lineata* Otto, 1820)
Pleurophyllidia henneguyi Labbé, 1922

External morphology:

The body is elongate and the notum smooth as in *Armina neapolitana*, showing 23 notal ridges, broad and narrow ridges alternate (Fig. 14A). Black pigment is visible between the ridges, as well as all over the body, even on the branchial and hyponotal lamellae (Fig. 14E, F). The oral veil is small. A caruncle is hardly distinguishable. Small bulges in front of the rhinophores were interpreted as a rest of the caruncle. The rhinophores are partly drawn back into the body. Fig. 14B shows the rhinophores looking down from above. The number of the lamellae varies from ten to eleven per rhinophore. The genital papilla is located just behind the gill on the right side of the body. Fig. 14D shows a detail of the papilla with three separated openings (oviduct, vagina and penis). A long, digitiform penis protrudes from the papilla. The anal papilla lies approximately in a median position on the right side. A nephropore is not visible.

The subnotal lamellae are partly damaged in the preserved specimen. 19 branchial lamellae are found on the left side and 26 on the right side. The hyponotal lamellae number 15 on the left and 25 on the right (Fig. 14E). At the lateral mantle margin a distinct white fringe is visible. Here, on both sides, in the anterior part of the body, bubble-like swellings are present (Fig. 14C). These are called marginal sacs here in accordance with Baba (1992). They are lined by a thin epithelium with pavement cells (Fig. 11D). Dorsally, in proximity to the opening of the marginal sac elliptical cells with a central nucleus are visible. In the lumen of the sacs sometimes spherical cells containing a large nucleus and few vesicles can be found. These cells are probably glandular. The sacs contain heterogeneous, thick, irregularly shaped, violet-staining secretions. They do not show a connection to the digestive gland. The foot is narrow and long in comparison to the other redescribed species. A white pedal gland is visible just in front of the caudal end of the foot.

Anatomy

Digestive system: The oral tube is elongate. Two lip-like structures surround the mouth opening at its anterior end. The oral glands are of the same colour as the salivary glands and therefore not as easy to distinguish as in *Armina neapolitana* and *A. maculata*. The position and shape of the oral glands and efferent ducts are the same as in the former two species. The pharynx (Fig. 15A) is muscular and elongate. The extension of pharynx and oral tube together is 6.6 mm in length. The breadth of the pharynx is 2.6 mm. The jaws are strong, with a denticulated masticatory border and a long masticatory process (Fig. 15D), each process bearing 5 to 6 rows of denticles (Fig. 17D). The radula has the formula 35 × 27.1.1.1.27. The rachidian tooth (140 μm broad) has 5 denticles on each side of the median cusp. The innermost denticle (Fig. 15Ea, 17A) lies very close to the median cusp. The first lateral is short, and denticulated on the outer side of the cusp (Fig. 15Eb). The following seven laterals also bear denticles. The eighth lateral has very small denticles, whereas the remaining teeth are smooth (Fig. 15Ee–g, 17B). The marginal tooth is small (Fig. 15Eh).

The salivary glands form compact masses; their position is the same as in *A. neapolitana* and *A. maculata* (Fig. 15B).The transition of the slender oesophagus to the stomach is continuous. The position of the digestive gland does not differ from the former species (Fig. 15B, C). The central canal is indistinct from the stomach. The canal is very long (22.4 mm). Nine lateral tubes emerge from the central canal on the right side, seven on the left. Glandular follicles of the digestive gland are densely packed in the lateral part of the body and within the notal tissue.

Nervous system: The nervous system of *Armina loveni* is very similar to that of *A. neapolitana* and *A. maculata*; an outline is shown in Fig. 16A. But the sizes of the ganglia differ from the former species: The cerebropleural ganglia have a size of 0.8 × 0.7 mm. The visceral loop is long, a visceral nerve emerges on the right side. The pedal ganglia are 0.8 × 0.6 mm and connected by a long pedal commissure. The parapedal commissure is not visible. The statocyst is spherical and lies between each cerebropleural and pedal ganglion. The buccal ganglia are ovoid in shape, lying underneath the oesophagus. Small gastro-oesophageal ganglia are present close to them. The eyes are found next to the rhinophoral ganglia but further towards the anterior end of the rhinophoral ganglion than in *A. neapolitana* and *A. maculata*. Following nerves and their innervation

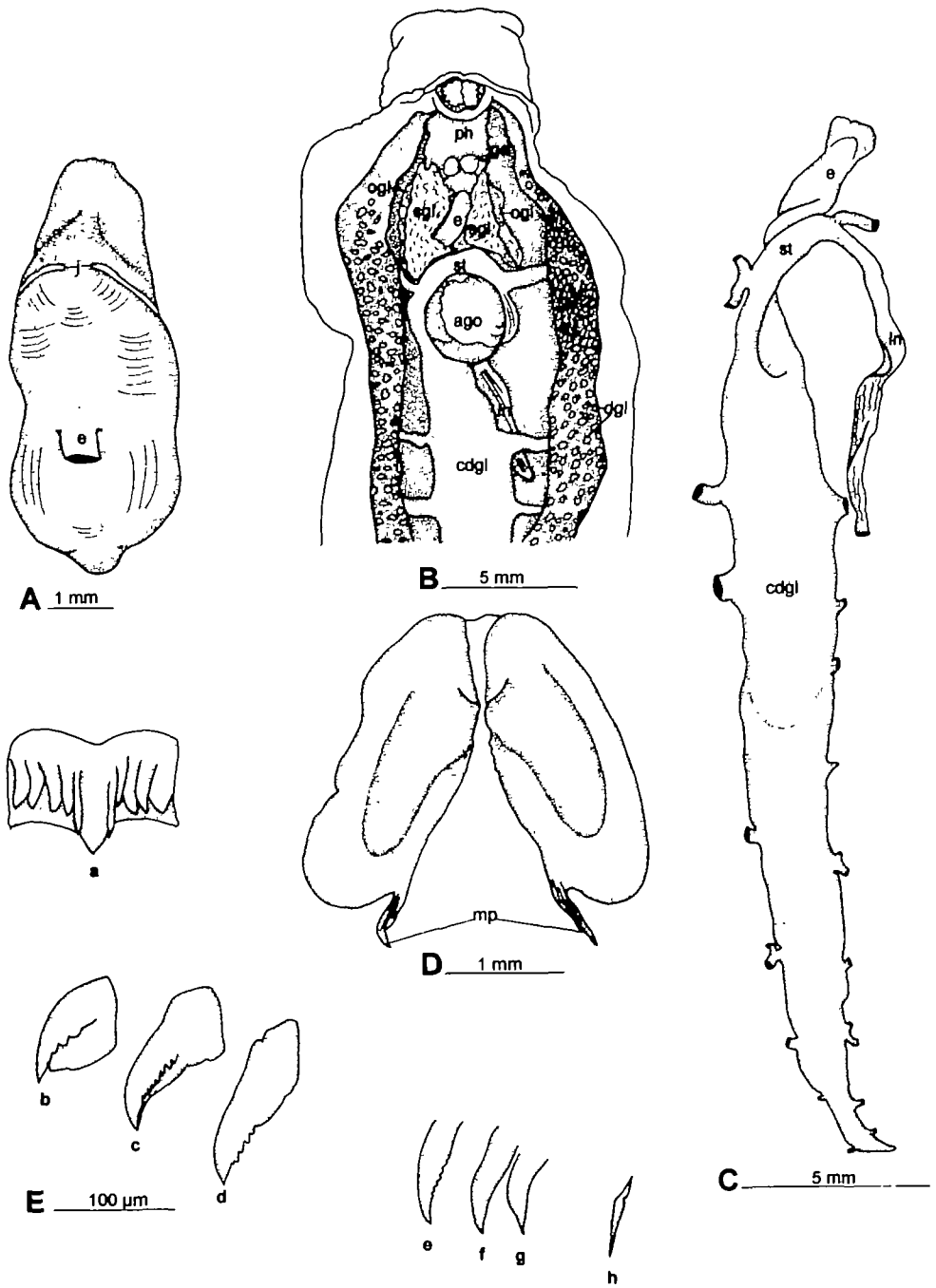


Figure 15. *Armina loveni*—Anatomy. **A.** Pharynx and oral tube. **B.** Dorsal view after removal of notum and gonad. **C.** Digestive system without glandular parts. **D.** Jaws. **E.** Radula teeth, **a.** Rachidian tooth, **b.–d.** Laterals 1–3, **e–g.** Laterals 8–10, **h.** Marginal tooth. Abbreviations: ago, anterior genital complex; cdgl, central canal of digestive gland; dgl, digestive gland; e, oesophagus; ga, ganglia; in, intestine; mp, masticatory process; ogl, oral gland; ph, pharynx; sgl, salivary gland; st, stomach.

areas were identified: R: rhinophores; C1: oral veil; C2: oral tube; C3: oral tube; Pl1: lateral margin of notum; P1: frontal part of foot; P2: lateral part of foot; P3: caudal part of foot; B1: pharynx; B2: pharynx; B3*: radula sheath

Reproductive system: The reproductive system of *Armina loveni* is similar to that of *A. neapolitana* and *A. maculata*. The gonad is compact and 16.5 mm in length. The ampulla is tube-like, coiled, first lying next to the allosperm receptacle, later running in between the female glandular mass (Fig. 16B). The transition of the ampulla to the postampullary duct occurs in proximity of the genital opening. The vas deferens immediately widens to a coiled prostate. The latter enters the penial sheath on its dorsal side. The penis is digitiform.

The short proximal oviduct widens before discharging into the female glands near the genital opening. The female glands are large and with the same, relative positions, as in the former species. Anatomically the proximal and distal parts of the mucus gland could not be distinguished. The distal oviduct leads into the genital papilla on the right side of the capsule gland, just underneath the vagina. The allosperm receptacle is spherical and filled with sperm, the vagina slender and long. A schematic outline of the distal genital complex is shown in Fig. 16C.

Heart and circulatory system: As described in *Armina maculata*.

Excretory system: The position and shape of the kidney are similar to *Armina neapolitana* and *A. maculata*.

DISCUSSION

Morphology: Little is known about the morphology and anatomy of this species. It was first described by Bergh in 1866, but his drawings (Plate II, Figs. 1–24) are restricted to the external morphology, the radula, the jaws and a few anatomical details. The drawing of the ventral side of a specimen shows the arrangement of the hyponotal lamellae rather imprecisely. It seems as if they are running parallel to the lateral mantle edge, which is contradictory to the results of this study where the hyponotal lamellae have a transverse arrangement. According to Pruvot-Fol (1954), the hyponotal lamellae are arranged very close together. This also does not correspond to the results presented in this paper. The marginal sacs, described in this paper, have been previously reported by Bergh (1879). According to Thompson & Brown (1984), numerous 'defensive glands' occur on the lateral mantle edge. In the present study, only a few sacs were found in the anterior part of the mantle edge. Those lying in the posterior part had probably been ejected.

*: nerve is not symetrical.

Anatomy: Bergh's (1866–67) drawings of the radula and the jaws are highly consistent with the findings in this examination, but I found fewer denticles on the rachidian tooth. Pruvot-Fol (1954) described the rachidian tooth as hardly differentiated from the laterals but this does not correspond to my findings.

Bergh (1866–67, 1879) reported a gonad divided into two parts. However, in the specimen examined here, the gonad has a uniform structure.

Armina tigrina Rafinesque, 1814

Synonymy:

Pleurophyllidia undulata Stammer, 1816
Diphyllidia lineata Otto, 1820
Armina undulata (Meckel, 1816)
Pleurophyllidia cuvieri Meckel, 1823
Pleurophyllidia neapolitana <Delle Chiaje> of different authors, non Delle Chiaje, 1824

Only a few organs of this species could be investigated because the specimens were in a poor condition.

External morphology:

On the dorsal side of the notum 23 ridges are visible (Fig. 18A). Along the margin of the ridges, black spots of pigment shimmer through the epidermis. The colour of the preserved specimen is rather dark, probably due to the method of fixation (unknown). A triangular caruncle lies in front of the rhinophores. The latter are partly drawn back into the body. The right rhinophore has eleven vertical lamellae. The body apertures have the same postion as in the former species (Fig. 18C). Numerous branchial lamellae (more than 100 per side) are present. The hyponotal lamellae (Fig. 18B) number 36 on the left side of the body and 40 on the right. A deep groove is present on the ventral side of the foot, probably resulting from fixation. Marginal sacs at the lateral notum edge were found only in histological sections. They are similar to those in *Armina loveni*, but the epithelium and the glandular cells cannot be distinguished because of the poor condition of the specimen. Only masses of secretions are visible in the lumen.

Anatomy:

Digestive system: No detailed examinations of oral tube, oral glands, salivary glands, pharynx, buccal armature, oesophagus and stomach were possible. The central canal of the digestive gland is long, narrow and lies on the left side of the body. The glandular part of the digestive gland is similar to that described for *Armina neapolitana, A. maculata* and *A. loveni*. It has a very compact structure.

Reproductive system: A schematic outline of the distal genital complex is shown in Fig. 19B. The genital system of a specimen from the BMNH was examined anatomically (Fig. 19A). The gonad is

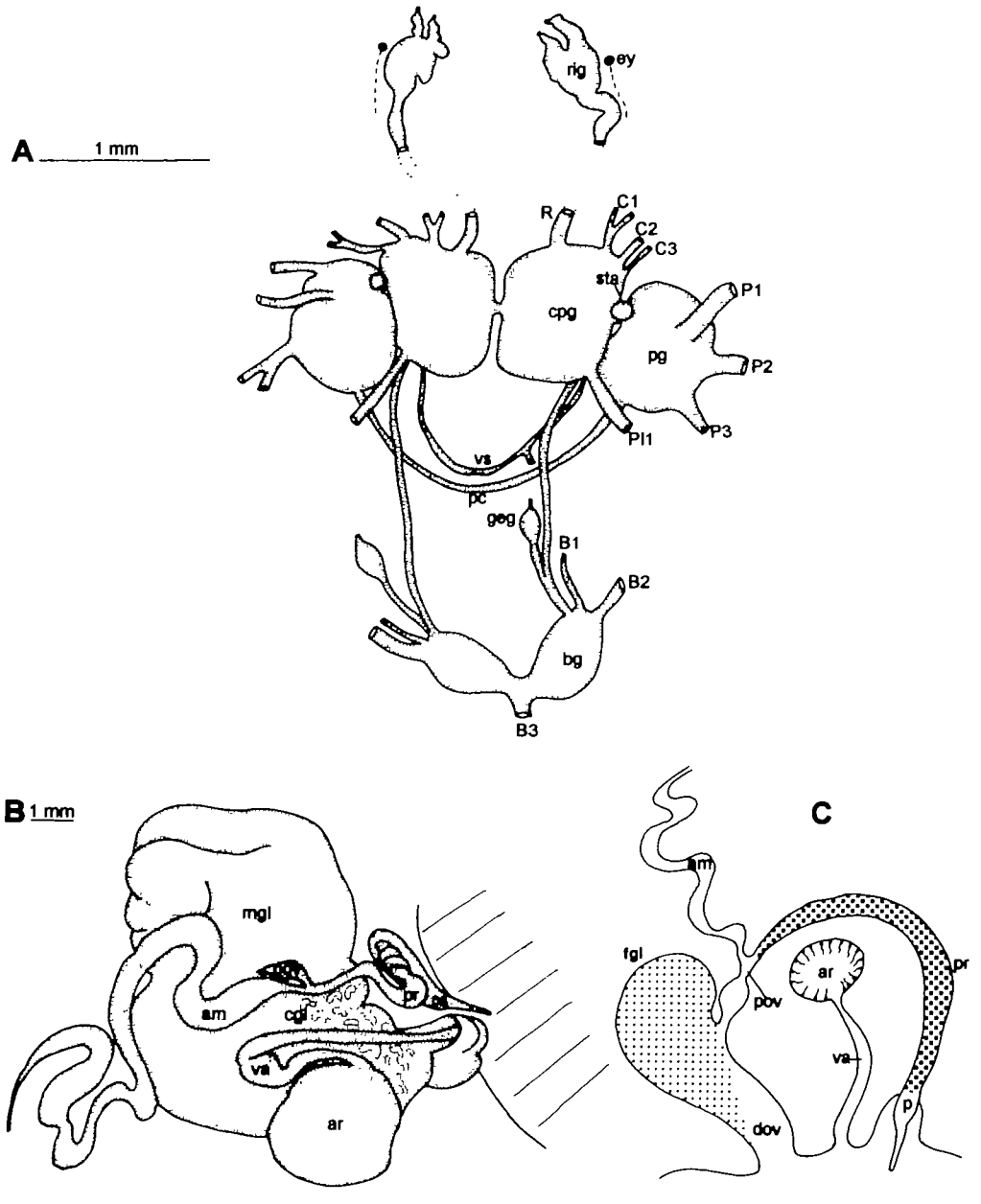
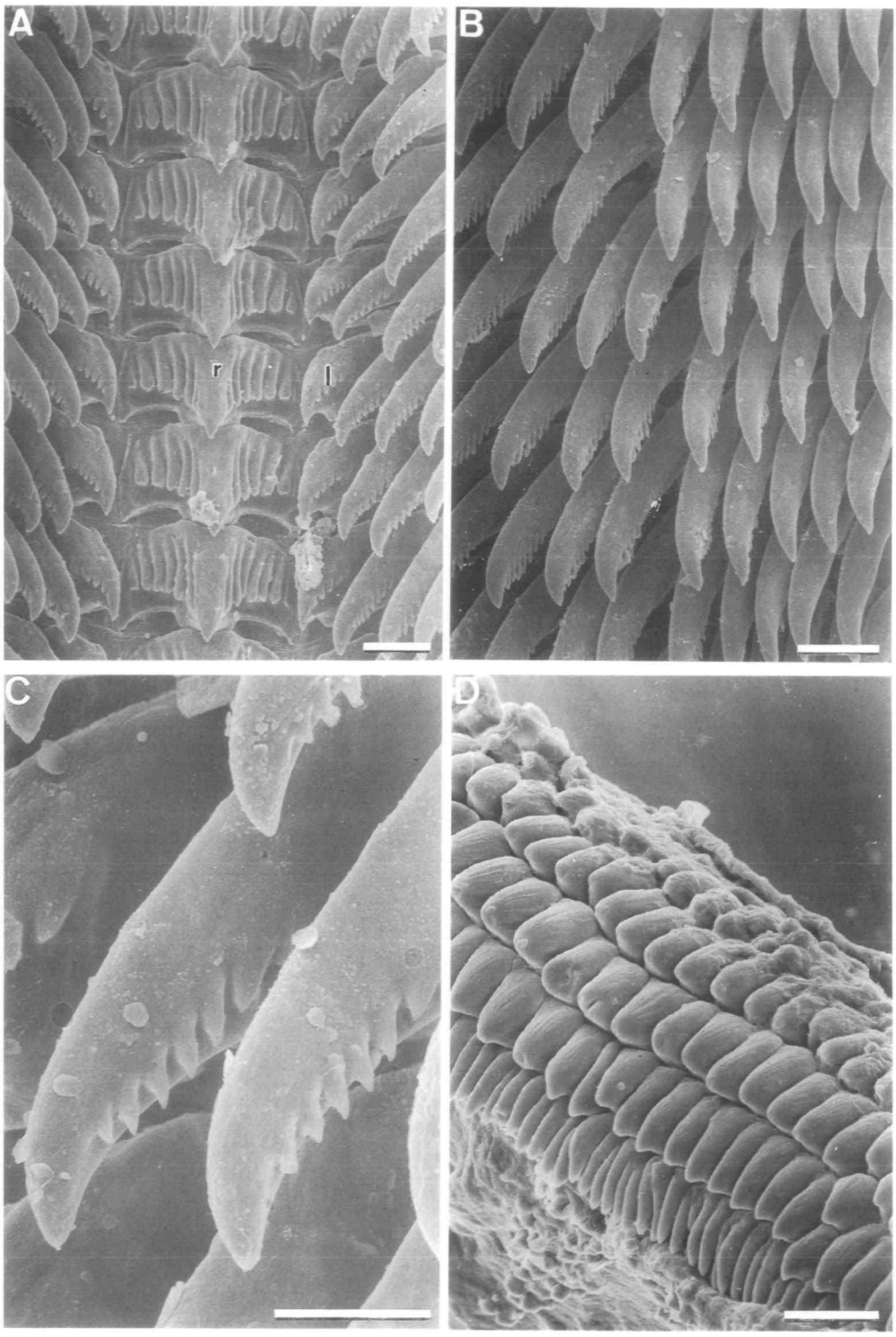


Figure 16. *Armina loveni*—Anatomy. **A.** Nervous system. **B.** Anterior genital complex *in situ* (parts slightly separated). **C.** Schematic outline of genital complex. Abbreviations: am, ampulla; ar, allosperm receptacle; bg, buccal ganglion; cgl, capsule gland; cpg, cerebropleural ganglion; dov, distal oviduct; ey, eye; fgl, female glands; geg, gastro-oesophageal ganglion; mgl, mucus gland; pc, pedal commissure; p, penis; pg, pedal ganglion; pov, proximal oviduct; pr, prostate; rig, rhinophoral ganglion; sta, statocyst; va, vagina; vl, visceral loop.

Figure 17. *Armina loveni*–SEM-photographs. **A.** Rachidian teeth with few laterals; scale bar = 50 μm. **B.** Lateral teeth; scale bar = 50 μm. **C.** Lateral teeth; scale bar = 25 μm. **D.** Rows of denticles on masticatory process; scale bar = 25 μm. Abbreviations: l, lateral tooth; r, rachidian tooth.



very large. Branches of the hermaphrodite duct run in between the follicles of the gonad. The hermaphrodite duct is long and its position is the same as in the former species. The transition from a tube-like, heavily coiled ampulla to the post-ampullary duct is abrupt. The vas deferens coils twice before it enters a small penis. A prostate is not distinguishable macroscopically, but the vas deferens widens at the beginning, and this may be the prostatic section. The penis is elongate, digitiform and protrudes dorsally of the vagina into the genital papilla. The proximal oviduct is more elongate and composed of stronger tissue than in the other species, widening proximally. The female glands are the same as in the former species. But it was not possible to differentiate between proximal and distal mucus gland. The allosperm receptacle is triangular. The vagina is more delicate than in other species.

DISCUSSION

Morphology: A short description of *Armina tigrina* was given by Pruvot-Fol (1937). In her opinion, the number of longitudinal ridges on the notum is greater than in *A. neapolitana* and this is confirmed by the present examination. Ballesteros (1983) counted 30 ridges in *A. tigrina*. The exact number is probably variable within one species and might also depend on the stage of maturity of the animal.

Anatomy: According to Ballesteros (1983), the heart lies at the hind end of the body. The results of the present study do not confirm this peculiar position. In fact the heart lies in the anterior part of the body, similar to all the other species described in this paper. In Ballesteros' (1983) drawings of the distal genital complex, the capsule gland is granted much more space than the mucus gland. The genital complex investigated here shows exactly the opposite relation, as also reported for all other examined *Armina* species.

Comparison of the examined species to each other and to other species of the genus:

The morphology, anatomy and histology of the four species examined in this study is very uniform with only few characters separating the species. Table 2 shows a comparison of 10 different morphological and anatomical characters within the four species. At this point it is relevant to mention that our present knowledge about the anatomy of other *Armina* species is very sparse. Therefore a comparison of certain anatomical characters within the genus must remain incomplete. The best character to distinguish the species is the shape of the radular teeth. While *A. neapolitana* has pectinate lateral teeth, *A. loveni* possesses denticles on the first nine to ten laterals. In *A. tigrina* virtually all the laterals are denticulated. *A. maculata*, on the other hand, has denticles only on the outside of the first lateral tooth and sometimes even this tooth is smooth in some specimens.

Distinctively shaped radular teeth are also found in other species of the genus. All *Armina* species have a multiseriate radula. Lim & Shou (1970) counted 74–80 rows of teeth in a 90 mm specimen of the new described species *A. carneola*. The highest number of teeth per row was reported for *A. major* Baba, 1949. O'Donoghue (1921) found the minimum number of 22 in *A. californica* Cooper, 1862 and Vayssiere (1901) in *A. tigrina* = *A. undulata*. In most species the rachidian tooth bears some denticles on either side of the median cusp. Only Baba (1949) found a smooth rachidian tooth in *A. major*. In *A. tigrina*, the denticles are very small (Thompson *et al.*, 1990). Many species have a distinct first lateral with its typical short, bulky shape. Frequently it bears small denticles on the outside of the cusp. The other lateral teeth mostly have elongated cusps, which can be denticulated or smooth. Marginal teeth seldom bear denticles as for example reported by Narayanan (1969) for *A. cinerea* Farran, 1905. Usually they are more slender and shorter than the other laterals. In some species the denticles of the laterals are elongated, so the teeth have a pectinate shape. This has been described for *A. neapolitana*. Marcus & Marcus (1966) report pectinate teeth also for the new species *A. joia*, O'Donoghue (1921) describes them for *A. californica* and Bergh (1890) for *A. ceylanica* Bergh, 1890. Pectinate teeth are also reported by Marcus (1961) for *A. columbiana* O'Donoghue, 1924.

The jaws are very uniform within the studied species. A denticulated masticatory process is always present. In descriptions of species of the genus *Armina* the number of rows of denticles is often given. But in my opinion this character is not suitable to distinguish the species, since the number is variable even within one species. For example, Ballesteros (1983) mentions six to seven rows for *Armina maculata* while Thompson *et al.* (1990) found about ten rows.

Other specific characters of the four examined *Armina* species are the coloration, the structure of the notum and the size of the body.

A. neapolitana has a light brownish colour with white notal ridges (Schmekel & Portmann 1982). Black pigment occurs between the ridges. The ventral side of the mantle and the foot are also light brownish. Pruvot-Fol (1937) describes the living animal of *A. tigrina* as

Table 2. Comparison of morphological and anatomical characteris in examined species:

Character	*A. neapolitana*	*A. maculata*	*A. loveni*	*A. tigrina*
length	max. 31 mm (fix)	largest species, up to 63 mm (fix)	31 mm (fix)	max. 53 mm (fix)
colour and structure of notum	brownish, white notal ridges, black pigments	orange, white pustules	light red-brown, light notal ridges, alternating broad and narrow ridges, haze of black pigment	black with white notal ridges
number of rhinophoral lamellae	right: 11 left: 9	right: 9 left: 10	right: 10 left: 11	right: 11 left: not visible
caruncle	triangular	insignificant	insignificant	insignificant
number of branchial lamellae	18–32/side	61–80/side	9 and 26/side	>100/side
number of hyponotal lamellae	9–19/side	>100/side	15,25/side	36,40/side
marginal sacs	present	present	present	present
radula	laterals: pectinate	1.lateral: denticulated, remaining laterals: smooth;	laterals: 1–9 slightly denticulated;	all laterals: denticulated;
jaws	large, with masticatory process	large, with process	slender, with process	large, with process (Thompson *et al.* (1990)
digestive gland	loosely packed follicles; branches: right 6, left 5	densely packed branches: right 4 left 6	less compact than *A. maculata* and *A. tigrina*; branches: right 9; left 7	very compact; branches: right 4 left 6 (Ballesteros 1983)
ampulla	few, large coils	heavily coiled	coiled	coiled
prostate	short	folded	very long and short vas deferens present	not distinguish-able
visceral loop	present	not found, one individual with visceral ganglion		no information
cerebral nerves	2 cerebral nerves	3 cerebral nerves	3 cerebral nerves	no information

having a black colour with white notal ridges. The foot is dark rose in colour. *A. maculata* has an orange notum with white pustules (Ballesteros 1981). The foot of this species is white. According to Bergh (1879), *A. loveni* has a red-brownish ground colour with light white-coloured ridges on the notum. Broad and narrow ones alternate. The epithelium is covered by a pale grey sheen which even covers the branchial and hyponotal lamellae. This is not described for any other *Armina* species.

The number of notal ridges also varies. *Armina neapolitana* bears the smallest number of ridges (maximum 16), while *A. loveni* and *A. tigrina* have more. Pruvot-Fol (1937) reported 40 ridges in *A. tigrina*. Because the animal studied by her was larger (and therefore probably older) than the specimen examined here, it could be assumed that the number of longitudinal notal ridges depends on the age or size of the animal. Thus, this character should not be overemphasized in the characterisation of species.

Within the genus *Armina*, the coloration and structure of the notum is variable. Nevertheless a dark ground coloration is typical for most species, while the notal ridges are set off in light colour. Only few species (e.g. *A. major* or *A. taeniolata* Bergh, (1860)) show lighter and brighter colours (Baba 1949, Eliot 1906 respectively). Three-quarters of the *Armina* species have longitudinal notal ridges. Only a few species bear pustules on the notum which may be partly aligned in rows (Baba 1955, Thompson *et al.* 1990, e.g. *A. babai* Tchang, 1936; *A. tricuspidata* Thompson, Cattaneo & Wong, 1990 and *A. variolosa* Bergh, 1904)). *A. bayeri* Marcus & Marcus, 1966 bears notal ridges

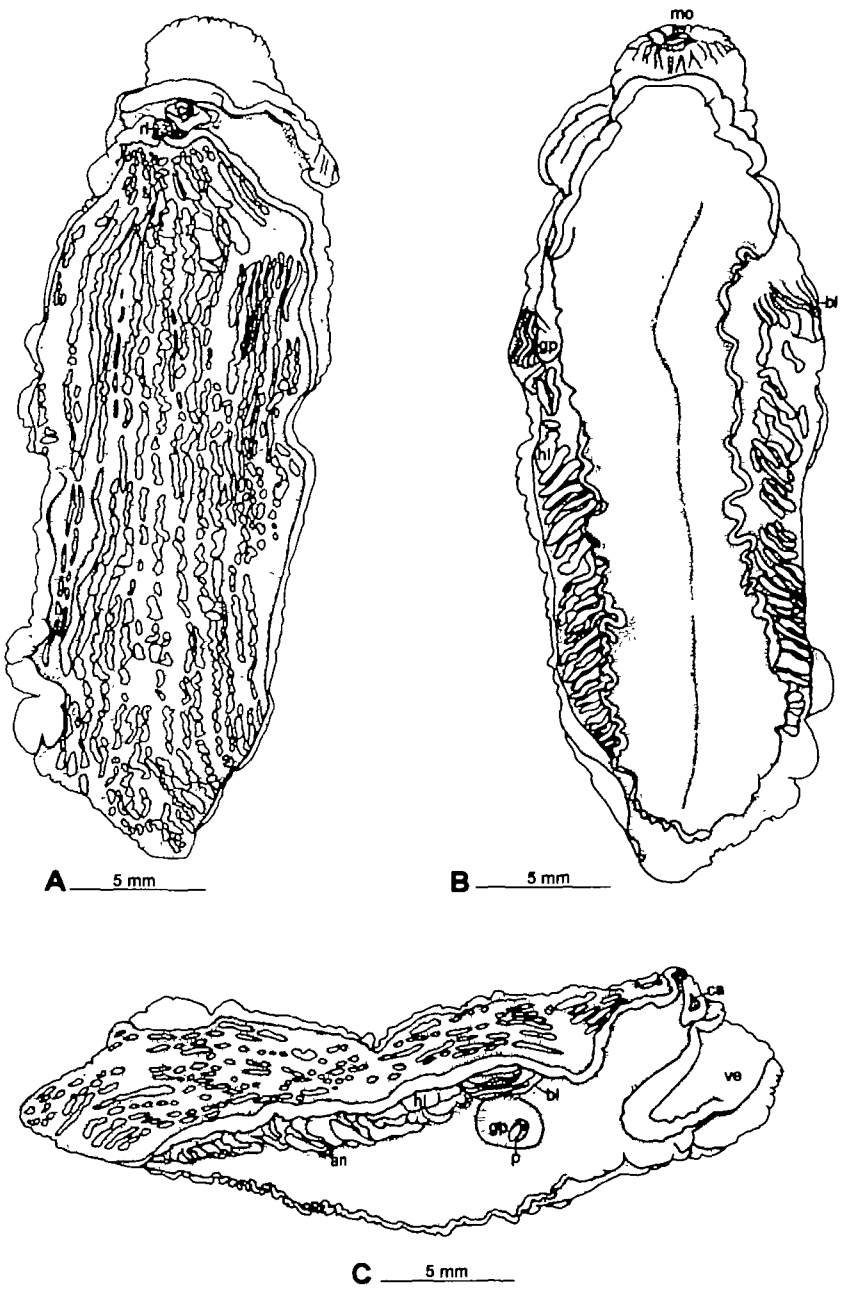


Figure 18. *Armina tigrina*. **A.** Dorsal view. **B.** Ventral view. **C.** Lateral view. Abbreviations: an, anus; bl, branchial lamellae; ca, caruncle; gp, genital papilla; hl, hyponotal lamellae; mo, mouth opening; p, penis; ve, oral veil.

which break down into pustules at the lateral sides of the notum.

A caruncle is found in all the species examined here. It is always situated in front of the rhinophores, but it varies in size and shape. In *Armina maculata* and *A. loveni* the caruncle is indistinct, while it has a distinct triangular shape in the other two species. Within the

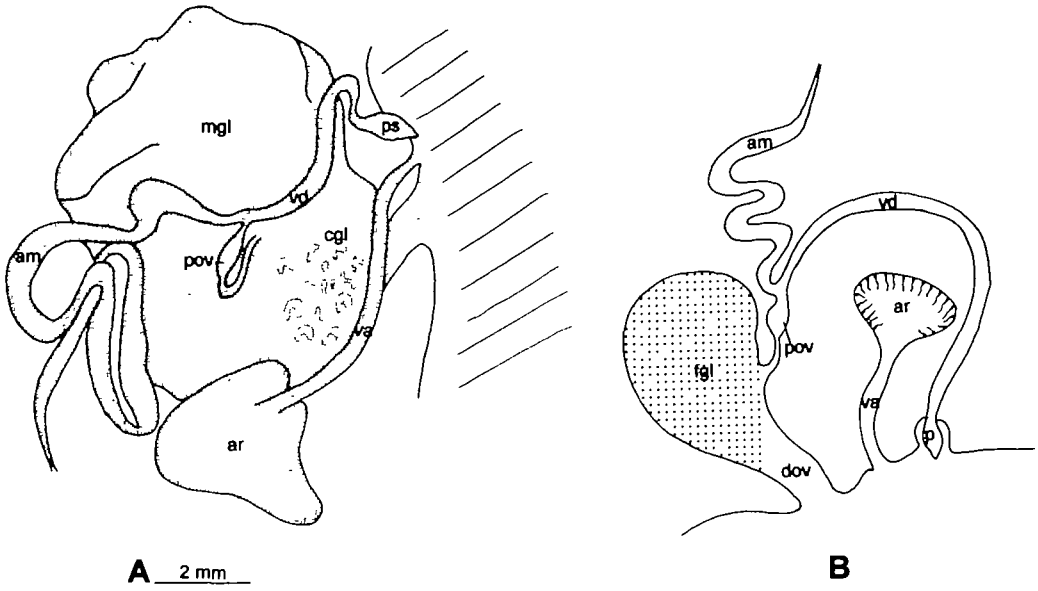


Figure 19. *Armina tigrina*—Anatomy. **A.** Anterior genital complex nearly *in situ* (parts slightly separated). **B.** Schematic outline of genital complex. Abbreviations: am, ampulla; ar, allosperm receptacle; cgl, capsule gland; dov, distal oviduct; fgl, female glands; mgl, mucus gland; p, penis; pov, proximal oviduct; va, vagina; vd, vas deferens.

genus the caruncle is variable in structure. The position is always in front of the rhinophores. In *A. aoteana* Miller & Willan, 1986 and *A. muelleri* v. Ihering, 1886 the caruncle is divided into two parts; one part lies in front of each rhinophore (Marcus & Marcus 1966, Miller & Willan 1986). Pruvot-Fol (1937) has stated that the shape of the caruncle varied within the same animal depending on whether it was alive or dead. Fixation may also have an influence on the shape of the caruncle. Therefore, I agree with Pruvot-Fol (1937) that the use of the caruncle as a character in systematics needs to be treated with caution. Different opinions exist about the function of the caruncle. Otto (1820) believed the caruncle functions as a cover for the rhinophores. Thompson *et al.* (1990) agreed with this, although Thompson & Brown (1984) previously considered it to be a sensory organ. Pruvot-Fol (1937) interpreted the caruncle as a rudiment of the rhinophoral sheaths of the tritoniids. This is impossible, because the rhinophoral sheaths are built by the anterior mantle margin whereas the caruncle according to its position originates from the oral veil. Therefore, the homology of these two structures can be excluded. According to the results of this study it is more probable that the caruncle builds a cover for the rhinophores when they are withdrawn into the body. But this assumption must be confirmed by investigations on living animals.

The number of the subnotal lamellae varies to a great extent amongst the species examined. Even within one specimen, the number of branchial and hyponotal lamellae differs from side to side. *Armina tigrina* possesses the largest number of branchial lamellae—over 100 per side—while *A. maculata* has the largest number of hyponotal lamellae (more than 100 per side in one individual). *A. neapolitana* and *A. loveni* both possess far fewer lamellae than the other two species. The arrangement of the lamellae is similar in all species; the branchial lamellae stand parallel to the notal edge, the hyponotal lamellae are arranged transeversely. The interspecific variability in the number of ventral lamellae is also represented by the other species of the genus. In addition, some *Armina* species show different arrangements of lamellae. The lowest number of branchial lamellae is reported by Hoffman (1940) for *A. pallida* Bergh (6 to 8 per side), the largest number found so far was described for *A. tigrina* in this study. Normally large lamellae alternate with smaller ones. Besides these primary and

secondary lamellae, respectively, also tertiary lamellae can occur as described by Miller & Willan (1986) for *A. aoteana*. Few species (e.g. *A. comta* Bergh, (1880) and *A. pallida*) only have a single hyponotal lamella on either side of the body (Hoffmann, 1940). If an animal has only few hyponotal lamellae they are usually arranged parallel to the edge of the foot. With an increasing number of lamellae, the arrangement is transeverse. Hyponotal lamellae are thicker than branchial lamellae. They contain branches of the digestive gland. This is also confirmed for other *Armina* species (MacFarland, 1966 for *A. californica*; Thompson *et al.*, 1990).

In the present paper marginal sacs are described for *Armina loveni, A. maculata* and *A. tigrina*. In *A. neapolitana* only structures which were interpreted as immature stages of marginal sacs could be observed. Mature marginal sacs were missing in this species. It is possible, that the marginal sacs disappear after the contents have been completely emptied out. Hoffmann (1939) cites Cuenot, who thinks that the lumen of an empty marginal sac is gradually replaced by connective tissue. Cuenot's assumption can be confirmed here, because the same situation was found in histological sections of *A. maculata*.

Marginal sacs are also described for other species of the genus. Usually they appear in large numbers, as e.g. in *A. californica, A. columbiana, A. joia* and *A. wattla* Marcus, 1967 (Marcus, 1961; Marcus & Marcus, 1967). The position of the marginal sacs along the lateral mantle brim is very uniform in all species. The marginal sacs do not show any connection to the digestive gland (MacFarland, 1966; Bergh, 1879; Hoffmann, 1939), as also confirmed here. At present we can only speculate about the function of the marginal sacs. They might be used for defence, as Hoffmann (1939) suggests.

The glandular part of the digestive system is of limited use for the discrimination of species. The structure of the digestive gland and the number of its branches arising from the central canal seem to be species-specific. Most branches were found in *Armina loveni*, fewest in *A. maculata*. However, all four species have two branches originating from the stomach, representing the right anterior and left anterior digestive glandular branch. Other species of the genus also have a cladohepatic digestive gland. The glandular part is found only in the lateral parts of the body. The largest number of branches of the digestive gland is reported for *A. muelleri*—, ten on the left side and eleven to twelve on the right (Marcus & Marcus, 1960). The lowest number is found in *A. maculata* present study; Ballesteros, 1983)—three to four on the right side and five to six on the left. *A. maculata* and *A. tigrina*, in comparison to *A. neapolitana*, show a compact digestive gland. The follicles of the digestive gland are densely packed in the lateral sides of the body in *A. maculata* and *A. tigrina*. *A. neapolitana*, on the other hand, has loosely arranged digestive gland follicles. *A. loveni* is intermediate. But the transitions are continuous from species to species.

The distal genital complex is very similar in all species examined. It is always diaulic. This also applies to the other species of the genus. Nevertheless, the shape and pathway of the ampulla seem to be specific. While the ampulla of *Armina neapolitana* has few, large, wide coils it is heavily coiled in *A. maculata*, whilst *A. loveni* and *A. tigrina* have an ampulla which has several coils prior to the transition to the postampullary duct. The position of the hermaphrodite duct between two parallel coils of the ampulla may be a specific character for *A. neapolitana*. At least it seems not to be variable within the species, because in all three investigated specimens the hermaphrodite duct had the same position.

Descriptions of the ampulla of other *Armina* species are very consistent with the results presented here. An exception is *A. wattla*, which according to Marcus & Marcus (1967) has a bladder-shaped ampulla.

The thorough re-investigation of four *Armina* species and the comparison with other *Armina* species has shown, that there are few characters which can be used to characterize the species. The histological investigation did not reveal new characters which would help to elucidate the phylogenetic relationship of the genus *Armina* within the Arminidae. Further research on the other genera of the Arminidae is needed to clarify their relationships.

ACKNOWLEDGEMENTS

I would like to thank Heike Wägele (Bielefeld) for her critical and helpful comments on the phylogeny of the arminids. I am thankful to Heike Wägele and Richard Willan (Darwin) for reading the manuscript. Material was kindly provided by Heike Wägele, Philippe Bouchet and Philippe Maestrati (Paris), David Reid and Kathie Way (London) and Christine Morrow (Dublin). Moreover, I would like to thank an unknown referee and John Taylor for helpful comments on the manuscript.

REFERENCES

BABA, K. 1949. *Opisthobranchia of Sagami Bay*. Iwanami Shoten, Tokyo.

BABA, K. 1955. Opisthobranch fauna in the vicinity of the Marine Biological station, Sado Island, Japan Seaside. *Collecting & Breeding*, **17**: 165-168.

BABA, K. 1992. Critical Review of *Dermatobranchus striatus* van Hasselt, 1824 (Nudibranchia: Arminidae) with the Description of a New Species. *Venus*, **59**: 239-248.

BALLESTEROS, M. 1981. Sobre un raro arminaceo (Mollusca: Opisthobranchia) de la costa mediterranea espanola: *Armina maculata* Rafinesque, 1814. *Publicaciones del Departamento de Zoologica Barcelona*, **6**: 27-31.

BALLESTEROS, M. 1983. Primera cita de *Armina tigrina* (Mollusca: Opisthobranchia) para las costas espanolas. *Publicaciones del Departamento de Zoologica Barcelona*, **9**: 53-62.

BERGH, R. 1866-67. Bidrag til en Monographi af Pleurophyllidierne, en Familie af de gasteropode Mollusker. *Naturhistorisk Tidskrift*, **4**: 1-81 & 207-380.

BERGH, R. 1869. Anatomische Untersuchungen der *Pleurophyllidia formosa*. *Verhandlungen der Zoologisch-Botanischen Gesellschaft in Wien*, **19**: 225-244.

BERGH, R. 1879. Notizen über *Pleurophyllidia loveni*. *Malakozoologische Blätter*, **26**: 77-87.

BERGH, R. 1890. Weitere Beiträge zur Kenntnis der Pleurophyllidien. *Verhandlungen der Zoologisch-Botanischen Gesellschaft in Wien*, **40**: 1-14.

BERGH, R. 1891. Die cladohepatischen Nudibranchien. *Zoologisches Jahrbuch der Systematik*, **5**: 1-75.

ELIOT, CH. 1906. On the nudibranches of Southern India and Ceylon with special reference to the Drawings of Kelaart, and the collections belonging to Alder & Hancock preserved in the Hancock Museum at Newcastle-on-Tyne. *Proceedings of the Zoological Society of London*: 636-691.

GARCIA, F.J. & GARCIA-GOMEZ, J.C. 1988. Estudio anatomico del sistema nervioso de *Armina maculata* Rafinesque, 1814 (Gastropoda, Opisthobranchia, Arminacea). *Iberus*, **8**: 75-87.

GARCIA, F.J. & GARCIA-GOMEZ, J.C. 1990a. The functional anatomy of the feeding apparatus of the nudibranch gastropod *Armina maculata* Rafinesque 1814, with a comparison with some other opisthobranchs. *Journal of Molluscan Studies*, **56**: 83-95.

GARCIA, F.J. & GARCIA-GOMEZ, J.C. 1990b. The Anatomy of the Circulatory System in the Arminid Nudibranch *Armina maculata* Rafinesque, 1814 (Gastropoda: Opisthobranchia). *Acta Zoologica (Stockholm)*, **71**: 33-35.

HOFFMANN 1939. *Opisthobranchiata*, Buch 3. In: Bronn's Klassen und Ordnungen des Tierreichs. Akademische Verlagsgesellschaft Leipzig.

HOFFMANN 1940. *Opisthobranchiata*, Teil 2. In: Bronn's Klassen und Ordnungen des Tierreichs. Akademische Verlagsgesellschaft Leipzig.

KOLB, A. & WÄGELE, H. (in press): On the phylogeny of the Arminidae (Gastropoda, Opisthobranchia, Nudibranchia) with considerations of biogeography. *Journal of Zoological Systematics and Evolutionary Research*,

LANCE, J.R. 1962. A New Species of *Armina* (Gastropoda: Nudibranchia) from the Gulf of California. *Veliger*, **5**: 51-54.

LIM, C.F. & CHOU, L.M. 1970. The nudibranchs of Singapore, excluding the families Dendrodoridae and Doridae. *Malayan Nature Journal*, **23**: 131-142.

MACFARLAND 1966. Opisthobranchiate Mollusks. *Memoirs of the California Academy of Sciences*, **6**: 546 pp.

MARCUS, E. 1961. Opisthobranch Molluscs from California. *Veliger*, **3**: (Supplement): 43-44.

MARCUS, DU B.-R., E. 1971. On some euthyneuran Gastropods from the Indian and Pacific Oceans. *Proceedings of the Malacological Society of London*, **39**: 355-369.

MARCUS, E. & MARCUS, E. 1960. Opisthobranchs from American Atlantic warm waters. *Bulletin of Marine Science*, **10**: 129-203.

MARCUS, E. & MARCUS, E. 1966. The R/V Pilsbury Deep-Sea biological expedition to the Gulf of Guinea, 1964-65. Opisthobranchs from tropical West Africa. *Studies of Tropical Oceanography Miami*, **4**: 152-208.

MARCUS, E. & MARCUS, E. 1967. Some opisthobranchs from Sapelo Island, Georgia, U.S.A. *Malacologia*, **6**: 199-222.

MILLER, M.C. & WILLAN, R.C. 1986. A review of the New Zealand arminacean nudibranchs (Opisthobranchia: Arminacea). *New Zealand Journal of Zoology*, **13**: 377-408.

NARAYANAN, K.R. 1969. On the opisthobranchiate fauna of the Gulf of Kutch. *Proceedings of the Symposium Mollusca* **1**: 188-213. Marine Biological Association of India, Ernakulam.

ODHNER, N. 1939. Opisthobranchiate Mollusca from the western and northern coasts of Norway. *Det Kgl Norske Videnskabers Selskabs Skrifter*, **1**: *1-93.*

O'DONOGHUE, C.H. 1921. Nudibranchiate Mollusca from the Vancouver Island region. *Transactions of the Canadian Institute*, **13**: 147-209.

OTTO, A.W. 1820. Über eine neue Roche und eine gleichfalls neue Molluske. *Nova Acta Physicomedica Naturae Curiosorum*, **9**: 120-126.

PRUVOT-FOL, A. 1937. Etudes des Opisthobranches des cotes Nord de la Mediteranee. *Archives du Museum d'Histoire Naturelle de Paris*, **14**: 35-74.

PRUVOT-FOL, A. 1954. Mollusques Opisthobranches. In: *Faune de France*, **58**. Paul Lechevalier, Paris.

PRUVOT-FOL, A. 1955. Les Arminidae (Pleurophyllidae ou Diphyllidiadae des anciens auteurs) *Bulletin du Museum National d'Histoire Naturelle de Paris*, **27**: 462-468.

SCHMEKEL, L. 1971. Histologie und Feinstruktur der Genitalorgane von Nudibranchiern (Gastropoda, Euthyneura). *Zeitschrift für Morphologie der Tiere*, **69**: 115-183.

SCHMEKEL, L. 1982. Vorkommen und Feinstruktur

der Vakuolenepidermis von Nudibranchiern (Gastropoda Opisthobranchia). *Malacologia*, **22**: 631-635.

SCHMEKEL, L. & PORTMANN 1982. *Opisthobranchia des Mittelmeeres, Nudibranchia und Sacoglossa*:. Springer-Verlag Berlin, Heidelberg, New York.

THOMPSON, T.E. & BROWN, G.H. 1984. *Biology of opisthobranch molluscs*. Vol. **2**: 94-102 The Ray. Society, London.

THOMPSON, T.E.; CATTANEO, R. & WONG, Y.M. 1990. Eastern Mediterranean Opisthobranchia: Dotidae (Dendronotoidea), Arminidae and Madrellidae (Arminoidea). *Journal of Molluscan Studies*, **56**: 393-413.

VAYSSIERE 1901. Recherches Zoologiques et Anatomiques sur les Mollusques Opisthobranches du Golf de Marseille. *Annales de Musée d'Histoire Naturelle de Marseille*, **2**: 1-130.