

THE "PROTOCHORDATES"

The term "protochordates" today includes the Phylum Hemichordata, and two subphyla of the Chordata, namely:

- A. Subphylum Cephalochordata, with Amphioxus and its kin, not represented locally.
- B. Subphylum Urochordata (= Tunicata) which includes:
 1. Class Ascidiacea, the sessile tunicates, well represented in the Woods Hole fauna and much used in experimental work.
 2. Classes Thaliacea and Larvacea, which are pelagic or planktonic, and are not covered in these keys.

PHYLUM HEMICHORDATA

The only common hemichordate of Woods Hole is the well known Saccoglossus kowalewskyi (A. Agassiz, 1873). This occurs intertidally or in shallow water in fine muddy sand where its presence is marked by little piles of fine, stringlike castings. The wormlike body is exceedingly fragile and difficult to collect undamaged. It is easily recognized by the characteristic whitish proboscis, orange collar, and brownish body (plate 28, fig. 1).

The development of S. kowalewskyi is direct, without drastic metamorphosis, from a ciliated larva. Some confusion has resulted from the claim of Agassiz that S. kowalewskyi arose from the metamorphosis of a planktonic "tornaria" larva. The fact that tornaria larvae do appear in local waters indicates that some other species may exist in not too distant, but probably deeper, waters.

Agassiz described our common species as Balanoglossus kowalewskyi; this was re-assigned to Dolichoglossus by Spengel in 1893, in his well known work on the Enteropneusta of the Gulf of Naples. However, Van der Horst, in his revision of 1939 (Bronn's "Tierreich", IV:4:2:2) adopted the prior name of Saccoglossus (published by Schimkewitsch in 1892), but long overlooked as a consequence of the paper being in Russian). Since there is no question as to the species kowalewskyi (or the variant spelling kowalevskii), experimentalists should use the specific name and not merely the generic name in reporting work done on Saccoglossus kowalewskyi at Woods Hole, in order to avoid possible confusion to foreign readers.

PHYLUM CHORDATA

SUBPHYLUM UROCHORDATA (= TUNICATA)

Class Ascidiacea

This section has been compiled from material written and contributed by Drs. Donald J. Zinn and Donald P. Abbott, whose assistance is much appreciated. The key is intended only for identification of ascidians in the Woods Hole region. Information about the anatomy and ecology of these animals may be found in Berrill (1950) and further material on their taxonomy in Van Name (1945). Embryological details are discussed at some length by Berrill (1936). Figure references on ascidians are to plate 28.

Ascidians show various degrees of colony formation: "simple" ascidians develop singly from eggs and do not bud; individuals in this group are relatively large. Colonial ascidians can reproduce by budding, and form aggregations of several to many small individuals. In "social" ascidians, such as Perophora, the individuals are distinct from each other and are connected only by stolons at their bases (figs. 2, 3). "Compound" ascidians, such as Botryllus, have minute individuals embedded in a common test or tunic which often has a characteristic growth form (figs. 7, 8).

It is often very difficult to see the anatomical features of fresh, unrelaxed ascidians, since they contract strongly when dissected. For purposes of laboratory study of anatomy, or if necessary for identification, relaxed and properly preserved specimens should be used. Place animals in pans of sea water to which is added Epsom salts ($MgSO_4$), about one heaping teaspoon per liter. After 12-24 hours of anaesthetization, fix for at least an hour before study in 5-10% formalin made up with sea water. This treatment permits easy observation of features nearly impossible to discern in delicate, contractile living forms or in contracted preserved material. Specimens that are kept for more than a couple of weeks should be transferred through 2 changes of 70% alcohol. Large specimens should have slits made in them to facilitate penetration. Fresh specimens will usually be easier to study if hardened a little by previous transfer to strong alcohol for a few days.

In using the following key, note that the branchial or inhalent siphon (morphologically the mouth) is considered anterior. Dorsal is defined by the position of the brain and subneural gland, which lie close to the surface between the siphons. The gonads in solitary ascidians are typically conspicuous structures, each consisting of a sac-like ovary on the surface of which may be located the lobes of the testes.

KEY TO COMMON ASCIDIANS

1. Colonial ascidians (individuals are small and connected by stolons or embedded in common test) 2
1. Simple ascidians (individuals usually of good size, and although they may occur in clusters are not organically connected to each other) 7
2. Colony thin, flat, encrusting; zooids colorful, and arranged in stellate or subcircular clusters or "systems" (figs. 7, 8); zooids of each system with branchial apertures opening independently, atrial apertures directed medially and opening into a common cloacal cavity in the tunic Botryllus schlosseri
2. Colony not as above 3
3. Colony consisting of numerous tiny (2-4 mm diameter) greenish individuals, connected to one another only by basal stolons (social ascidians) (figs. 2,3) Perophora viridis
3. Colony not as above; individual zooids completely embedded in matrix of the common test (compound ascidians) 4
4. Forming flat encrusting colonies 2-4 mm thick and up to more than 10 cm in diameter; test smooth on surface, opaque because of large numbers of minute stellate calcareous spicules which are white, yellowish, or reddish in color; no postabdomen present Didemnum candidum
4. Form of colony otherwise; test not containing stellate calcareous spicules, though sometimes with encrusting or embedded sand grains; postabdomen present, containing the gonads 5
5. Colony in the form of massive "meaty" plates, lumps, or lobes up to several inches long and attached by one edge; tunic very firm; stomachs of zooids bearing about 12 longitudinal ridges (fig. 5) Amaroucium stellatum
5. Colony not as above; stomachs of zooids bearing 20 or more longitudinal ridges (fig. 4) 6
6. Colonies low, globular, oval or flat topped, attached by a narrow base Amaroucium constellatum
6. Colonies consisting of globular masses built up of radially arranged, transparent, finger-like lobes, which may contain encrusting and embedded sand Amaroucium pellucidum

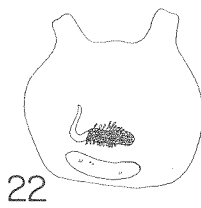
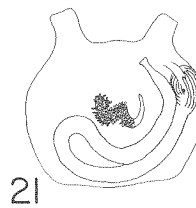
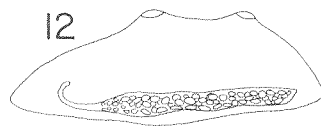
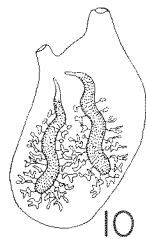
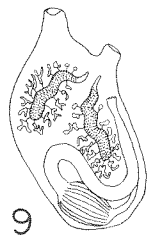
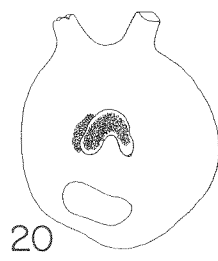
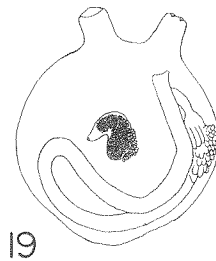
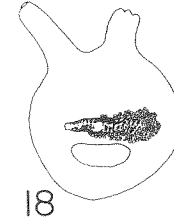
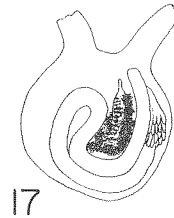
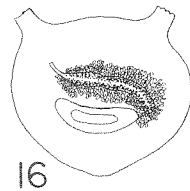
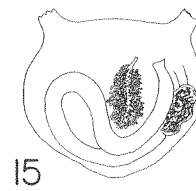
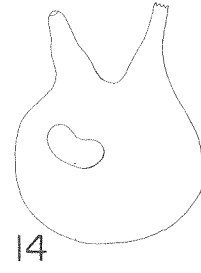
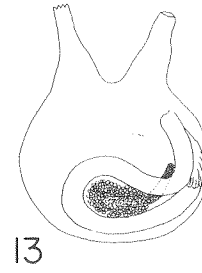
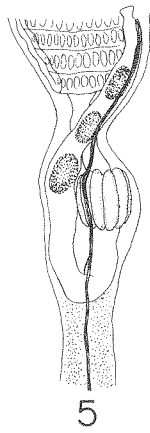
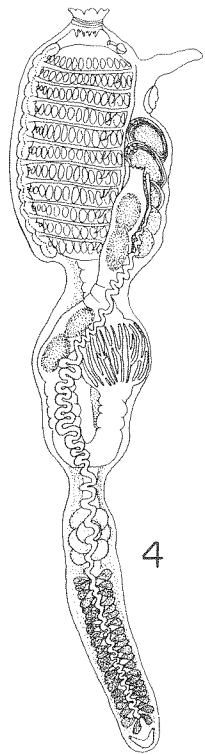
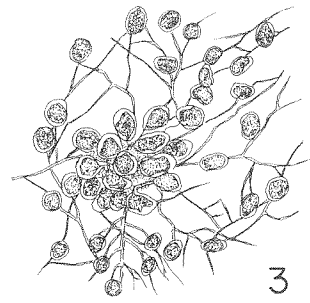
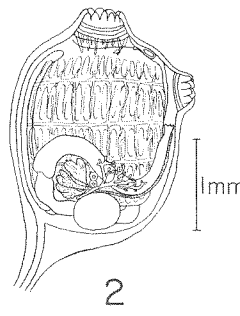
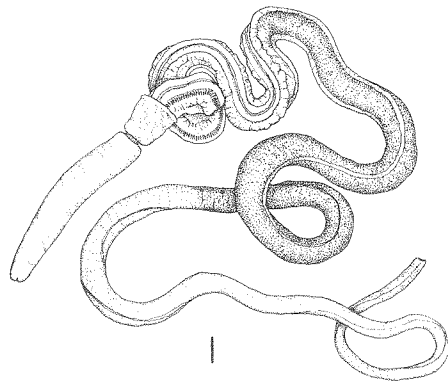
- 7. Branchial siphon with 8 lobes, atrial siphon with 6; body elongate, up to several inches long; test grayish, gelatinous, transparent enough when expanded to reveal the pharynx with 5-7 longitudinal muscle bands on each side and the main loop of the gut, posterior to the pharynx, basally (fig. 6). Ciona intestinalis
- 7. Branchial siphon with 4 or 6 lobes; body and test not as above 8
- 8. Branchial aperture (also atrial aperture) square or 4-lobed; dissection shows pharyngeal stigmata straight, elongate, and not arranged in spiral or circular fields; no kidney present STYELIDAE 9
- 8. Branchial aperture usually 6-lobed, atrial aperture usually 4-lobed; dissection shows pharyngeal stigmata more or less curved and arranged in circular or spiral fields; kidney present on the right side MOLGULIDAE 10
- 9. Test leathery, tough, irregular, yellowish or brownish in color; when collected often feels like a rough lump of gristly consistency; 2 gonads on each side of the body (figs. 9, 10); pharynx with 4 longitudinal folds on each side Styela partita
- 9. Test thin, tough, membranous, pinkish to bright red in life, attached by a relatively large area basally; body depressed with long axis usually parallel to the substrate; 1 gonad on the right side (fig. 12), none on the left (fig. 11); pharynx with one longitudinal fold on the right side, none on the left Dendrodoa carnea
- 10. Adults live in soft sand or mud substrates, unattached to larger objects 11
- 10. Adults do not live free in soft sand or mud, but occur attached to larger objects such as gravel, rocks, floats, pilings, marine plants, shells 12
- 11. Siphons arise close together; body covered with minute fibrous processes which hold a layer of mud about the body; dissection shows only one gonad, this present on the left side (figs. 13, 14); pharynx lacking longitudinal folds but bearing 7 large internal longitudinal vessels on each side Bostrichobranthus pilularis
- 11. Siphons separated at bases by a space greater than 1/5 of body circumference (figs. 15, 16); dissection shows 2 gonads, one on each side of the body; pharynx bearing longitudinal folds on each side, each fold marked by a small group of internal longitudinal vessels Molgula arenata
- 12. Very common, attached to eel grass, floats, pilings, etc., in bays; long axes of both gonads straight (figs. 17, 18) Molgula manhattensis
- 12. Less common; long axes of one or both gonads conspicuously curved 13
- 13. Gonads on both sides an inverted U-shape, leaving open ends of oviducts pointed away from the base of the atrial siphon (figs. 19, 20) Molgula complanata
- 13. Left gonad with an S-shaped or W-shaped curve, right gonad with a straight axis; both gonads with open ends of oviducts curved to point toward the base of the atrial siphon (figs. 21, 22) Molgula citrina

Plate 28

PROTOCHORDATES

Figures 1, 3, and 9 from life; rest redrawn
from Van Name (1945); all by Bruce Shearer.

- Fig. 1. Saccoglossus kowalewskii, whole animal, from life.
2. Perophora viridis, individual, seen from left side.
 3. Same, colony from life, to show "social" growth habit.
 4. Amaroucium constellatum, zooid with larvae in atrial cavity. Note the numerous stomach ridges.
 5. Amaroucium stellatum, region of stomach, to show the fewer and simpler ridges.
 6. Ciona intestinalis, diagram of body seen from left, to show position of gut and gonads; branchial sac omitted.
 7. Botryllus schlosseri, single zooid seen from left side.
 8. Same, small colony from life, showing investment in common tunic.
 9. Styela partita, diagram of body as seen from left and as if wall transparent, branchial sac omitted, showing gut and gonads.
 10. Same, seen from right side, showing gonads.
 11. Dendrodoa carnea, view of left side showing gut.
 12. Same, view of right side, showing gonad.
 13. Bostrichobranchus pilularis, diagram of body seen from left, showing gut and gonad.
 14. Same, from right, showing renal sac.
 15. Molgula arenata, body from left, showing gut and gonad.
 16. Same, body from right, showing renal sac and gonad.
 17. Molgula manhattensis, body from left.
 18. Same, from right.
 19. Molgula complanata, body from left.
 20. Same, from right.
 21. Molgula citrina, body from left.
 22. Same, from right.



ANNOTATED LIST OF ASCIDIANS

Order Aplousobranchia

Family Synoicidae

Amaroucium stellatum Verrill, 1871, Massachusetts to Florida. "Sea Pork"; viviparous.
Amaroucium constellatum Verrill, 1871. New Hampshire to Florida. Good embryological material. Viviparous.
Amaroucium pellucidum (Leidy, 1855). Massachusetts to Florida. In 10-20 meters on sand, shells, or gravel in tidal currents. Viviparous.

Family Didemnidae

Didemnum candidum lutarium Van Name, 1910. Van Name (1945) considered the form prevalent at Woods Hole, and extending from southern Maine to the south side of Cape Cod, to be a northern geographical race of D. candidum Savigny, 1816. This may prove erroneous (see Millar, 1962. *Studies Fauna Curaçao ... 8*: pp. 62-66), but the familiar name D. candidum is retained until further investigation shows the necessity of a change.

Order Phlebobranchia

Family Cionidae

Ciona intestinalis (Linnaeus, 1767). One of larger ascidians of the United States, and one of the best known and most widely distributed of all ascidians. Greenland to Rhode Island. Oviparous.

Family Perophoridae

Perophora viridis Verrill, 1871. Although small, its transparent test makes it favorable for observation of heartbeat, etc. Southern Cape Cod to southern Florida. "Sea grape"; viviparous.

Order Stolidobranchia

Family Botryllidae

Botryllus schlosseri (Pallas, 1766). Very common; colonies most often found encrusting in variety of colors on hydroid stems and eel grass leaves. Portland, Maine to West Coast of Florida; viviparous.

Family Styelidae

Styela partita (Stimpson, 1852). Formerly well known as Cynthia partita; the eggs are classical embryological material. Common on wharf pilings, often with other ascidians, from Massachusetts Bay to West Coast of Florida. "Sea peach" (note that the name "sea peach" is also used for the long-stalked ascidian Boltenia, often cast up on beaches north of Cape Cod, and the large solitary Halocynthia aurantium of more northerly waters).
Dendrodoa (Styelopsis) carnea (Agassiz, 1850). Pinkish to bright red body spreads out on surfaces of stones, dead mussel shells, etc., to which animal is attached. Newfoundland to Long Island Sound. Viviparous.

Family Molgulidae

- Bostrichobranchus pilularis (Verrill, 1871). Size and transparency of test depend on environment. Found on or in mud or sand from one meter to greater depths. Tadpole larvae not formed. St. Lawrence River to Florida.
- Molgula arenata Stimpson, 1852. Commonly unattached; buried; regular, symmetrically circular in profile (except between siphons), disk-like when viewed from above and generally covered with coat of sand grains. South Shore of Cape Cod to Cape May, New Jersey. Tadpole larvae not formed. Oviparous.
- Molgula citrina Alder and Hancock, 1848. Usually attached to hard objects or pilings; vary greatly in appearance and shape; are translucent dull olive. Gulf of St. Lawrence to Narragansett Bay, Rhode Island. Viviparous.
- Molgula complanata Alder and Hancock, 1870. Unusually long synonymy (see Van Name); sand and shell encrusted and more or less globular body attached by slender fibrous processes to stones, shells or hard sand from low water to about 300 fathoms. Gulf of St. Lawrence to Martha's Vineyard. Viviparous.
- Molgula manhattensis (DeKay, 1843). Commonest ascidian of this area. Nearly globular body with test irregularly hirsute and usually papillated near siphons; attached to others of same species in large groups or to wharves, pilings, ship bottoms, eel-grass in shallow, polluted, brackish or ordinary sea water. Portland, Me. to N. E. Texas (southern Florida?) Oviparous with larvae developing outside of body of parent.
- Molgula robusta (Van Name, 1912). Not in key. Differs from M. manhattensis in a) lacking free-swimming larval stage, b) larger average size, c) living at 17-25 meters on compacted mud-sand bottoms in areas of tidal currents, unattached and usually on left side, d) long pointed teeth on margin of posterior part of dorsal lamina, and e) reddish purple eggs. Woods Hole Harbor and Vineyard Sound. Oviparous.

REFERENCES

- Berrill, N. J. 1936. Studies in tunicate development. Part V. The evolution and classification of ascidians. Phil. Trans. Roy. Soc., London Ser. B., Vol. 226: 43.
- Berrill, N. J. 1950. The Tunicata. London, Ray Society, 354 pp.
- Grassé, P. P., Ed. 1948. Traité de Zoologie. Tome XI. Échinoderms, Stomocordés, Pro-cordés. pp. 895-919.
- Van Name, W. G. 1945. The North and South American Ascidiaceans. Bull. Amer. Mus. Nat. Hist. 84: pp. 1-476, pl. 1-31.