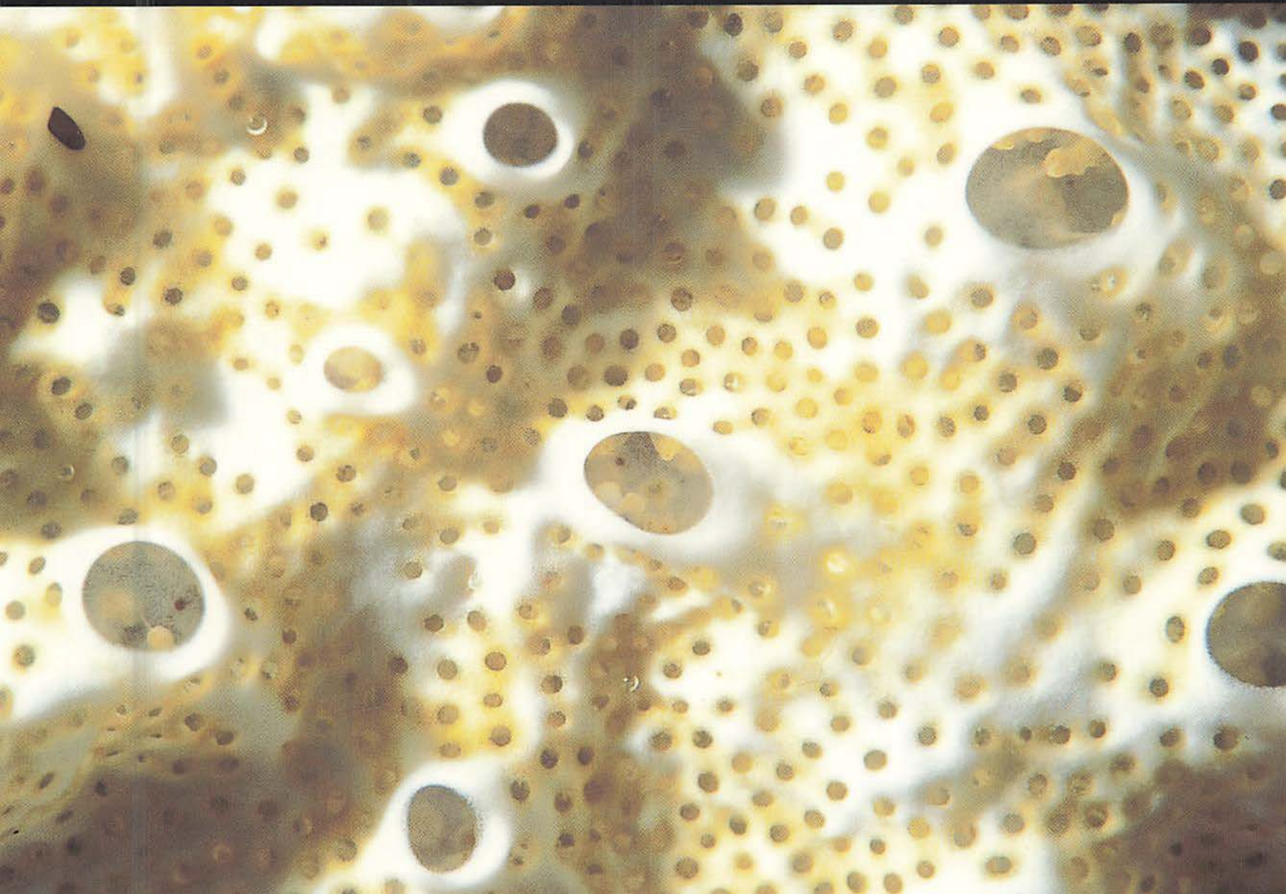
Reef and Shore Fauna of Hawaii

Section 6B: Ascidians (Urochordata)



Donald P. Abbott • A. Todd Newberry • Kendal M. Morris

REEF AND SHORE FAUNA OF HAWAII

Section 6B

Note on the Series

REEF AND SHORE FAUNA OF HAWAII

INTEREST IN THIS SERIES, begun with Charles Howard Edmondson's single-volume publication in 1933, has remained strong for more than half a century. A revised edition was published in 1946, and in 1965 a decision was made to generate an expanded series based on the original work.

The series was originally planned for six volumes:

Section 1: Protozoa, Porifera, Cnidaria, and Ctenophora
Section 2: Platyhelminthes, Nemertina, Aschelminthes, Entoprocta, Bryozoa (Ectoprocta), Brachiopoda, and Phoronida
Section 3: Sipuncula, Echiura, and Annelida
Section 4: Mollusca
Section 5: Arthropoda
Section 6: Echinodermata, Chaetognatha, Hemichordata, and Chordata (excluding vertebrates)

Section 1: Protozoa through Ctenophora was published in 1977. This was followed in 1979 by *Section 4: Mollusca* (*Hawaiian Marine Shells*), with Dr. E. Alison Kay as sole author. In 1987, the combined *Section 2: Platyhelminthes through Phoronida* and *Section 3: Sipuncula, Echiura, and Annelida* was published with eight chapters and multiple authors. This present volume, *Section 6B: Ascidians (Urochordata)*, is the second part of Section 6. The remaining parts are forthcoming.

This volume could not be realized without the efforts of the three authors (D. P. Abbott, A. T. Newberry, and K. M. Morris) and Gretchen Lambert, who edited the final manuscript. Further acknowledgments are extended to Dr. W. Donald Duckworth, President and Director of Bishop Museum.

LUCIUS G. ELDREDGE
Series Editor

REEF AND SHORE FAUNA OF HAWAII

Section 6B: Ascidians (Urochordata)

DONALD P. ABBOTT

A. TODD NEWBERRY
Institute of Marine Sciences, University of California, Santa Cruz

KENDAL M. MORRIS
Science Communication Program, University of California, Santa Cruz

Edited by
GRETCHEN LAMBERT
Department of Biological Science, California State University, Fullerton

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Cover photograph by John Hoover.
A didemnid colony from Pupukea, northern O‘ahu. Symbionts color the white surface. Tadpole larvae lurk in the large, common cloacal orifices.

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DON ABBOTT TAKING A BREAK FROM SNORKELING FOR ASCIDIANS IN KĀNE‘OHE BAY, HAWAI‘I, SUMMER 1985.

PREFACE BY THE SECOND AUTHOR

DONALD ABBOTT wrote his first account of Hawaiian ascidians in 1941, as an undergraduate at the University of Hawaii. The essay won him the Dean Memorial Prize there. That essay has grown into this section of the *Reef and Shore Fauna of Hawaii.* As a professor at Stanford University, Dr. Abbott largely put aside Hawaiian studies in favor of collaborative research with his colleagues and students. When he retired to Honolulu in 1983, he eagerly anticipated completing his survey of Hawaiian ascidians. By the time of his death in 1986, Dr. Abbott had largely completed the first draft, except for an introductory section on methods, commentaries on the Ascidiidae and Pyuridae, and the decisions on the identity of several puzzling species of *Perophora*, *Ascidia*, and *Polyandrocarpa.*

Dr. Abbott left some taxa identified only to genus: *Botryllus* (=*Botrylloides*) sp./spp., *Cnemidocarpa* sp., *Ascidia* "A" and "B." I believe he felt that candid admissions of uncertainty were appropriate for this naturalist's survey and preliminary assessment of Hawai'i's ascidian fauna, so I have left intact some of his generic identifications and retained the tentativeness of some of our species identifications. I hope this candor will provoke new efforts toward clarifying the Hawaiian ascidian fauna.

Besides adding and enlarging introductory sections, I have revised and enlarged the species accounts in most families. Reexamination of specimens and reference to recent monographs have resulted in some important revisions: renaming two familiar species (*Diplosoma macdonaldi* to *D. listerianum; Styela partita* to *S. canopus*), changing two generic designations (*Archidistoma* to *Eudistoma; Botrylloides* to *Botryllus*), and correcting the identification of three species (*Diplosoma virens* to *D. similis; Perophora japonica* to *P. annectens; Corella japonica* to *C. minuta*). Reasons for these changes are noted in the appropriate text sections.

Dr. Abbott had not begun the drawings. Kendal Morris has drawn directly from specimens of all taxa except the didemnids and botryllinids, which are redrawn from published sources. Some drawings combine details of different specimens in order to best depict traits described in the text. This practice reflects our intention to provide nonspecialists with a guide to the identification of the currently known Hawaiian taxa, and to give specialists an acquaintance with which to undertake still more ambitious investigations of the region's ascidian fauna.

The University of California, Santa Cruz, Institute of Marine Sciences and Faculty Research Committee supported parts of this project. Keyt Fischer provided very helpful editorial suggestions. John Hoover's photograph of a living didemnid ascidian colony graces the cover of this volume. I especially thank Charles and Gretchen Lambert, of California State University, Fullerton, for so generously sharing their knowledge, observations, findings, and insights about Hawaiian ascidians and for attending to this project over its many years.

It is only at her request that Gretchen Lambert is not a listed author of this account, as she should be. She has given hundreds of hours to editing the manuscript through a decade of revisions, ensuring that it is as complete and accurate as it can be and as readable as it is. She put the whole project in motion anew when I had given it up for lost a few years ago. Since then, she has patiently and deftly guided it to this fruition, a paragon of skill and a fast friend.

PHYLUM CHORDATA
SUBPHYLUM UROCHORDATA (TUNICATA)
CLASS ASCIDIACEA

THE BEST-KNOWN MEMBERS OF THE PHYLUM CHORDATA are, of course, the vertebrates. But this phylum also includes two "invertebrate" subphyla, Urochordata (or Tunicata) and Cephalochordata. A notochord, hollow dorsal nerve cord, and rudimentary pharyngeal gill slits are found in the larvae of all chordates. Cephalochordates retain all three traits throughout their lives, but among the urochordates only the pelagic larvaceans, which resemble persistent larvae, retain these traits through adulthood.

Urochordata comprises the long-established classes Thaliacea, Larvacea, and Ascidiacea and the class Sorberacea (C. Monniot et al. 1975), whose validity is still debated (Kott 1989a; C. and F. Monniot 1990). Pharyngeal filter-feeding prevails in the three better-known classes, but the wholly abyssal sorberaceans use a prehensile oral lobe to feed macrophagously. Thaliaceans (salps and their relatives) and larvaceans are holoplanktonic and are not dealt with in this account of Hawai'i's benthic reef and shore fauna.

The name Urochordata refers to the notochord, a support-rod of specialized cells that lies largely or wholly in the larval tail (and in the tail of larvacean adults). As the widely used alternative name Tunicata implies, the body is encased in an external tunic, or test. The tunic, an extracellular matrix composed of specialized cellulose, other carbohydrates, and fibrous glycoproteins, contains numerous and diverse living cells.

Members of the class Ascidiacea (the ascidians, or sea squirts) lose their notochord and dorsal nerve cord when they metamorphose from non-feeding, briefly swimming larvae to benthic adults. The "tadpole" larvae of most ascidians, though nearly microscopic in size, superficially resemble frog tadpoles, hence the term applied to them. Metamorphosis includes the swift destruction of the tail and resorption of its musculature and of the notochord and dorsal nerve cord, loss or transformation of the rest of the larval central nervous system, development of numerous pharyngeal gill slits (stigmata) from larval protostigmata (if this did not already occur during the development of the larva itself), postural adjustment of the newly attached larva so that it will be an effective benthic filter-feeder, opening of the anus into a cloacal atrium, opening of the oral and atrial siphons, and commencement of a cilia-driven flow of water through the pharyngeal stigmata.

Some ascidians grow to solitary adulthood. In other species, starting with the oozooid that emerges from metamorphosis of the larva, replicative modular growth ("asexual reproduction") produces colonies of budded or strobilated, genetically identical "blastozooids." According to species, solitary ascidians may be merely a few millimeters in diameter even as adults, or they may grow to several centimeters high or across. The zooids of colonial ascidians are usually much smaller than the bodies of solitary ascidians, and colonies may be diminutive, too. But in some species the colonies themselves may become encrusting or lobate masses several or even many centimeters across or extensive formations of zooids connected by stolons.

For fuller accounts of ascidian physiology, anatomy, and ecology, we recommend the classic treatises by Huus (1937), Brien (1948), and Berrill (1950); major topical reviews by Millar (1971), Godeaux (1974), and Goodbody (1974); and recent works by Kott (1985, 1990, 1992a, in prep.) and C. Monniot et al. (1991). Here, we emphasize those diagnostic characters that help identify Hawaiian ascidians for the naturalist.

TERMINOLOGY

For many people, ascidians present an unfamiliar anatomy described by a specialized vocabulary. Figures 1a and 1b illustrate most of the relevant terms. Those used here are widely accepted in the United States. To associate this vocabulary with what one may encounter elsewhere, especially in Australian and European research, we provide the following parallel terminology:

atrial (aperture, siphon, languet) = cloacal (aperture, etc.)
atrium = cloacal cavity or peribranchial space (or cavity)
dorsal slit = ciliated pit or slit, aperture of dorsal tubercle, opening of ciliated funnel, vibratile organ
endocarps (hernias of the atrium wall) = parietal vesicles
mantle = body wall
pharyngeal (wall, sac, folds) = branchial (wall, etc.)
stigmata = gill slits
tunic = test

SOLITARY ASCIDIANS

The body of an adult solitary ascidian (Fig. 1a) lies within its tunic as a sac bounded by a thin and delicate or thick and muscular mantle. The body has two apertures in its mantle, borne on tubular siphons. The anterior, oral aperture opens into the pharynx through a passage that is screened by a ring of oral tentacles. A dorsal tubercle with a ciliated slit bulges from the roof of this prepharyngeal passage. The ciliated slit is the opening of the duct to the neural gland; the duct and gland may regulate blood volume (Ruppert 1990). A large ganglion close by constitutes the "brain."

Driven by cilia on the pharyngeal wall's stigmata, a water current enters the pharynx through the oral aperture and passes through the stigmata, into a surrounding atrium. This filtered water leaves the body via the dorsal, atrial aperture. The atrium acts as a large cloacal cavity, for it receives water from the pharynx, feces from the gut, and gametes from the gonoducts. It may act, as well, as a brood chamber for larvae. The atrial aperture provides the outlet to the sea for all these products.

The pharyngeal wall is the site of respiratory exchange between the ascidian and its environment. The stigmata of most ascidians are small, simple longitudinal slits arranged in transverse rows (i.e., rows perpendicular to the endostyle), although in a few major families they form elaborate spirals (cf. Fig. 9b). The rows of stigmata and the stigmata within each row are separated by blood sinuses. The pharynx may have two to more than fifty rows of stigmata per side, and the number of stigmata per row also varies from few to many, according to species. The transverse blood sinuses of the pharyngeal wall may bear papillae that project into the lumen of the pharynx, and these papillae may bifurcate distally and join to form internal longitudinal pharyngeal blood vessels. The resultant vascular grid can become still more complicated as subsidiary transverse blood sinuses and secondary papillae proliferate. Spiral complexes of stigmata are accompanied by even more intricate patterns of sinuses and vessels. In some solitary ascidians, the pharyngeal wall is pleated into prominent longitudinal folds that further enhance the surface available for feeding and respiration and increase the complexity of pharyngeal blood circulation.

An elaborate glandular groove, the endostyle, marks the ventral midline of the pharynx. The endostyle produces sheets of mucus that traverse the right and left pharyngeal walls, carried along by coordinated beating of stigmatal cilia as the mucus passes over the stigmata. This mucus is collected dorsally by a dorsal lamina that is membranous or subdivided into a line of languets ("little tongues"). The extremely fine pored mucous sheets intercept and trap food particles (even smaller than 0.5 µm; see Flood and Fiala-Medioni 1981) in the water that passes from the pharynx through the mucus and then through the stigmata. Collected dorsally, the mucous sheets form a food-laden rope that moves along the lamina or languets to the esophagus.

The digestive tract (esophagus, stomach, intestine, and accessory structures) begins near the dorso-posterior end of the pharynx and recurves on itself to form a U-shaped (Fig. 2) or S-shaped (Fig. 13) loop of gut, with the anus opening into the atrium near the atrial aperture. As Figure 1a shows, in solitary ascidians most of the gut loop usually lies to the left of the pharynx. The family Corellidae is exceptional in having its gut loop largely to the right of the pharynx. In a few solitary forms—the Cionidae (Fig. 6) and the solitary forms of Diazonidae—all of the gut except the rectum lies below the pharynx.

Most solitary and colonial ascidians are simultaneous hermaphrodites; their testes and ovaries mature roughly synchronously. Exceptions to this rule are some abyssal solitary forms with wholly separate sexes and the zooids of some colonial botryllinids that are sequentially hermaphroditic. In some solitary ascidians, such as the ascidiids (Figs. 12, 13), the gonads lie in the atrium within the first, or primary, intestinal loop, in what is called the "enterogonid" condition. In others, such as styelids (Figs. 17, 18, 19) and pyurids (Figs. 20, 21), the gonads are "pleurogonid": they lie in the mantle wall of the atrium, mostly without strict relationship to the gut loop.

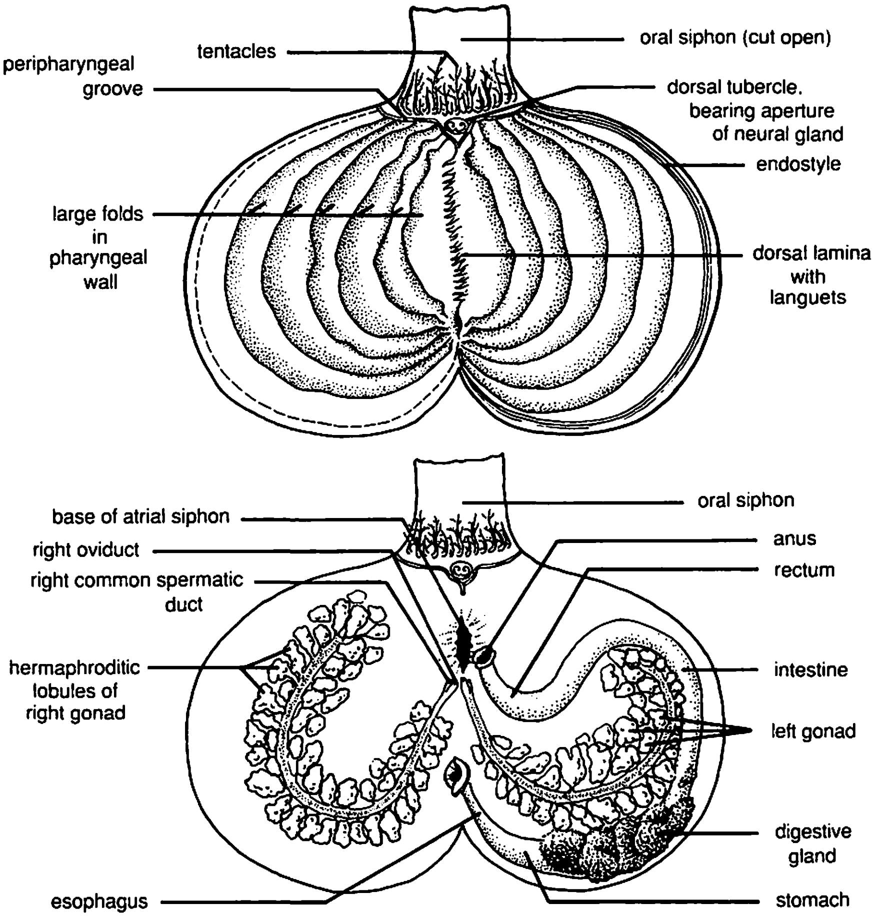

Figure 1a.—Solitary ascidian opened (dashed line) along mid-ventral line to show internal structures; body lies "on its back." These two drawings depict a pyurid stolidobranch of the genus *Pyura*. Various solitary and colonial stolidobranchs and phlebobranchs have different patterns of pharyngeal folds, stomach, gonads, and other structures, as shown in figures of particular species. Upper drawing shows pharynx in place; lower one shows structures on the atrium wall revealed by removal of pharynx. This animal shows the pleurogonid gonadal condition; for the enterogonid condition, see Figs. 12 and 13. (redrawn from Abbott 1975)

COLONIAL ASCIDIANS

The zooids of almost all colonial species are much smaller than the bodies of solitary ascidians. Some colonials—the Polyclinidae (Fig. 2), Polycitoridae (Fig. 3), and Didemnidae (Fig. 5), for example—differ considerably from solitary ones in the structure of their zooids. In others, such as Perophoridae (Figs. 7, 8) and some Styelidae (Figs. 15, 16), the zooids closely resemble small solitary ascidians. Colonial species also vary in the density and orderliness with which zooids are arranged in the colony. In the loosest (so-called social) colonies, such as some perophorids, zooids are well separated from each other and connected only by a

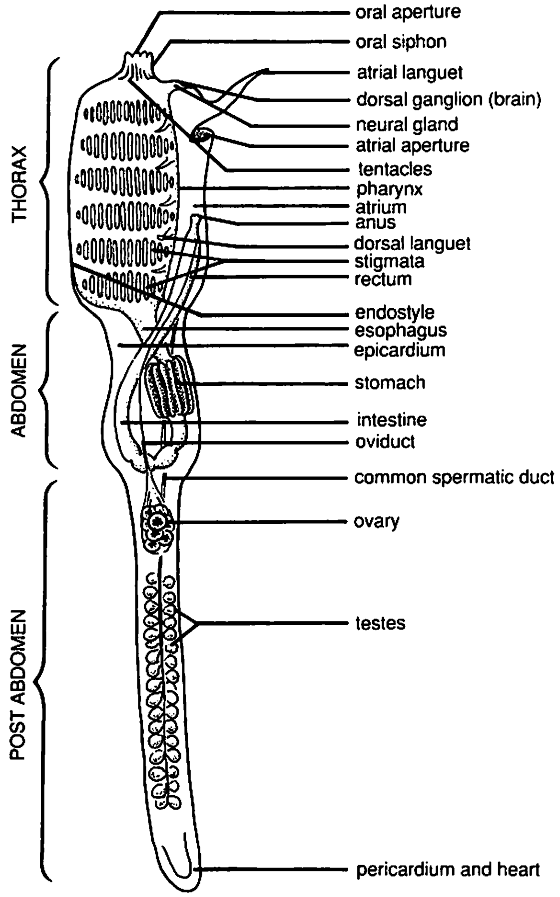

Figure 1b.—Aplousobranch zooid removed intact from its tunic and viewed from left side. This drawing depicts a zooid of the polyclinid genus *Aplidium;* other aplousobranchs differ (e.g., complete atrial siphon, constricted junctions between body regions, lack of post-abdomen), as shown in figures of particular taxa. Lines in this drawing point either to actual structures (e.g., pharynx, enterogonid gonads) or to their location in the zooid (e.g., atrium, epicardium). Atrium surrounds only the pharynx. (redrawn from Abbott 1975)

thin sheet or stolon of tunic that carries a colony-wide system of blood vessels. In the more compact "compound" forms, the zooids may individually occupy lobes that crowd together in bouquet-like colonies, or they may lie more or less completely buried together within the crusts or lobes of a common tunic. The Styelidae, again, are an especially diverse family, with solitary, "social," and "compound" species. In many families and even within some genera, diverse colony forms no doubt reflect the overriding importance of what Jackson (1979) has called the "morphological strategies of sessile animals" even more than they do taxonomic affinities (see also Kott 1981b).

The oral aperture of a colonial zooid always opens atop a siphon that reaches fully to the tunic surface, and the atrial aperture often does, too. Among Hawaiian ascidians, zooids project their atrial apertures directly to the colony surface on obscure atrial siphons (*Eudistoma,* Fig. 3; *Symplegma,* Fig. 15) or prominent ones (*Polyandrocarpa,* Fig. 16). In some compound ascidians, zooids are arranged in little starlike groups, with their still-separate but inconspicuous atrial siphons bunched centrally and their oral siphons on the periphery of the group. Rudimentary systems are shown in Hawaiian polycitorids and some botryllinids. In still more elaborate systems (*Polyclinum,* Fig. 2; didemnids, Fig. 4; *Botryllus* and "*Botrylloides,*" Fig. 14), the zooids' atrial apertures open into spaces (common cloacal cavities) within the tunic which collect the pooled effluent of several to many zooids before passing it to the outside through a common cloacal aperture. The atrial siphons then are usually reduced to canopy-like atrial languets (Fig. 2a, c) or, in many didemnids, have disappeared entirely, leaving each zooid with a gaping dorsal hole (Fig. 5).

The zooids of some colonial ascidians are short and compact, as in the enterogonid perophorids (Figs. 7, 8) and the pleurogonid styelids (Figs. 15, 16), with the intestinal loop lying beside the pharynx. The zooids of other colonials are elongate, with two or three distinct regions: thorax, abdomen, and post-abdomen (Fig. 1b). In these forms, the thorax contains the apertures and the neural complex between them, the pharynx and its surrounding atrium, and the terminal portions of the hindgut and gonoducts. The abdomen contains the loop of the gut—esophagus, stomach, and proximal intestine. In zooids that have only two body regions (polycitorids, didemnids, diazonids), the abdomen also holds the heart and gonads; in forms with three body regions (polyclinids), the heart and gonads lie below the gut loop in a distinct post-abdomen. As in solitary ascidians, the inner pharyngeal wall of some colonial zooids (diazonids, perophorids, styelids) may bear papillae projecting from the transverse blood sinuses and may carry internal longitudinal vessels. But more frequently in compound ascidians, especially those with very small zooids, pharyngeal papillae and internal longitudinal vessels are absent or at least are markedly fewer than in closely related but larger solitary forms.

An actively growing ascidian colony has zooids in all stages of development, from new buds to sexually mature blastozooids. But, beyond a slight difference between the colony's founding oozooid and all its subsequent blastozooids, ascidian colonies—even the highly specialized ones of many didemnids—exhibit no polymorphism among mature zooids. This is in striking contrast to such other colonial invertebrates as hydrozoan cnidarians and gymnolaematid bryozoans.

SYSTEMATICS

Lahille (1886, 1888) proposed that ascidians be arranged by their pharyngeal structure into three orders: Aplousobranchiata (*aplouso* = simple), Phlebobranchiata (*phlebo* = vessel, for the internal longitudinal vessels of the pharynx), and Stolidobranchiata (*stolido* = pleated). Huus (1937), Berrill (1950), and Kott (1969, 1985, 1990, 1992a, in prep.) advocate using gonadal arrangements to erect two ascidian orders, the Enterogona and the Pleurogona. They make Lahille's groupings suborders, putting (as they spell them now) the Aplousobranchia and

Phlebobranchia together into the Enterogona, and the Stolidobranchia alone into the Pleurogona. Either system produces the same result, a tripartite organization of groups of families.

Here, we adopt Kott's (1969) orders and families, except that, despite her persuasive comments (1969, 1990), we follow the wider practice of including the families Cionidae and her Diazonidae among the phlebobranch, not the aplousobranch, enterogonids.

Solitary or colonial, the enterogonid ascidians have gonads lying amidst or on the loop of the gut, or below the gut loop in the post-abdomen if that part of the body is present. Enterogonid gonads have a single testis and ovary, although sometimes these can spread widely over the wall of the gut or over each other and even spill onto the atrium wall. In contrast, in the order Pleurogona the gonads are usually multiple and when mature bulge from the atrium wall, entirely or mostly apart from the gut loop. Pleurogonid gonads may be single or numerous per side; they are often arrayed bilaterally symmetrically. Within the single family Styelidae, testis and ovary may be tightly bound together within a common membrane (*Cnemidocarpa,* Fig. 18) or loosely associated structurally (*Styela,* Fig. 19). And in a few styelid genera (e.g., *Distomus,* not yet found in Hawai'i) the ovaries and testes lie well apart from each other.

All aplousobranchs are colonial. The zooids have a thorax and abdomen and sometimes a post-abdomen. The aplousobranch pharynx lacks internal papillae and longitudinal vessels. The arrangement of the aplousobranchs into families is subject to debate: Kott (1990, 1992a) advocates an especially elaborate array of families, whereas F. Monniot (e.g., C. and F. Monniot 1972; C. Monniot et al. 1991) prefers to divide the group into just three families. Fortunately for our own purposes, currently known Hawaiian aplousobranchs all fall into families—Polyclinidae, Polycitoridae, and Didemnidae—which these two authorities agree on.

Phlebobranchs include solitary and colonial forms. Except for the diazonids, which have a substantial abdomen carrying the gut loop and gonads, the phlebobranch gut twists quickly into a loop that is tucked beneath the pharynx (in Cionidae) or lies in the atrium wall to the right of the pharynx (in Corellidae) or to its left (in all other families, which in Hawai'i are Perophoridae and Ascidiidae). Thus, in most phlebobranchs the thorax dominates the body. The pharynx lacks prominent folds; it has papillae or carries internal longitudinal blood vessels.

The stolidobranchs all belong to one or another of the three pleurogonid families—Styelidae, Pyuridae, and Molgulidae. All pyurids and molgulids are solitary ascidians; styelids may be solitary or colonial. The stolidobranch body is compact, with the gut lying in the atrium wall to the left of the pharynx (Fig. 1a). The pharyngeal walls of almost all solitary stolidobranchs are thrown into longitudinal folds, along which run most of the internal longitudinal vessels. These folds are reduced or absent in a few tiny molgulids and minute solitary styelids and in colonial styelids whose zooids are very small, but even then they are suggested by closely apposed internal longitudinal vessels.

The monogeneric family Cionidae and the probably closely related Diazonidae seem to link aplousobranchs and phlebobranchs, so much so that, although traditional wisdom calls these families phlebobranchs, Kott (1969, 1990) considers them aplousobranchs. For her, *Ciona*'s and the diazonids' regenerative epicardia (sacs derived from the pharynx which often surround various internal organs) and the way colonial diazonids strobilate point the way to the epicardial-based replication of aplousobranch zooids. In the other, more surely phlebobranch families and in all stolidobranchs, on the other hand, if the embryonic epicardia persist at all in adults, they are merely excretory vestiges.

From a cionid source, one can conjecture, the pharynx wall has evolved toward complexity in large solitary phlebobranchs and stolidobranchs but repeatedly has been simplified in the small zooids of colonial phlebobranchs and stolidobranchs, and it has become especially simple in the entirely colonial aplousobranchs. In the stolidobranchs, further, the gonads become pleurogonid; and the ability to fix elemental vanadium, shared by phlebobranchs and aplousobranchs, gives way in stolidobranchs to iron fixation.

Whether *Ciona* is a "stem" aplousobranch or a phlebobranch—or even a "proto-design" from which both suborders eventually arose—the seemingly rudimentary, or at least phylogenetically ambiguous, condition of some of its traits makes it and its diazonid relatives singularly interesting for anyone trying to understand the relationships between these two great ascidian assemblages and their constituent families. Stolidobranchs most likely emerged from early phlebobranchs.

Worldwide, well over 2,500 living species of ascidians have been described. A dozen families have substantial shallow-water representation; nine are known from Hawai'i. A few large and widespread families, such as the Molgulidae and Clavelinidae, have not yet been found here. In part, Hawai'i's isolation from other shallow-water regions probably deprives it of the diversity of ascidians typical of many archipelagoes of the western Pacific. Ascidian tadpole larvae are not, after all, the long-range dispersers that molluscan, echinoderm, and annelidan larvae are.

They may not be able to travel far on their own, but ascidian larvae may hitch rides in the ballast water of ships. C. Monniot et al. (1991), Carlton and Geller (1993), and C. and F. Monniot (1994) address a consequent trend that Carlton and Geller call "the homogenization of the sea." Not only may larvae stow away in ships, to be released in new places, but also adults of many invertebrates, again including ascidians, may foul boat hulls with the same result. Ships and large boats that arrive from far away often tie up in a port for weeks, allowing hull foulers to breed and their larvae to settle; then the vessels move on, probably with new hangers-on. Nonindigenous species undoubtedly have already established themselves this way in Hawaiian ports, affecting the biota in still unknown ways, and more will do so. For example, we can expect more immigrant species of *Polyandrocarpa*, *Microcosmus*, and *Styela* to join port-dwelling ones that have already hitchhiked here.

Although invasions of immigrant taxa are best known in ports, they may account as well for some population fluctuations in other habitats, such as reefs and outer shores. These littoral alterations may not be as profound as Hawai'i's famous terrestrial extinctions, but we know too little about the historical ecology of our coastal waters and of our reefs and shores to distinguish passing shifts there from faunal alterations of longer duration, or sometimes even to tell natural causes from human ones when our marine biodiversity changes.

The rich ascidian fauna (92 species, 39 of them undescribed) that C. and F. Monniot (1987) found in their SCUBA-based survey of French Polynesia, also an isolated mid-Pacific archipelago, suggests that Hawai'i's ascidian fauna, too, may turn out to be far more diverse than it now appears. Indeed, this account draws mostly on intertidal collections and harbor surveys in southern O'ahu, augmented by limited dredging and trawling. But many ascidian species exploit hard vertical substrates, the undersides of rocks and coral blocks, and other subtidal sites that can only be sampled well by SCUBA, which has scarcely been used yet in searches for Hawaiian ascidians. Hawai'i's geographical extent and ecological variety surely hold significant additions to this report.

PREPARATION OF ASCIDIANS FOR IDENTIFICATION

Many ascidians are identifiable alive and in the field, some even most easily that way. But sure identification of most species, especially the colonial forms with their tiny zooids, requires close scrutiny in the lab and dissection of preserved specimens.

Ascidians should be anesthetized before they are fixed for dissection and identification, lest they contract drastically. The distortion of key traits (e.g., rows of stigmata) which accompanies contraction can defeat one's best efforts to identify specimens, especially compound forms. A few taxa, such as species of *Eudistoma*, have strong muscles that contract even in heavily sedated specimens. Relaxing ascidians remains a rough art, particularly now that cocaine hydrochloride, perhaps the best general anesthetic for them, is inconvenient to obtain or keep. Other reagents vary in effectiveness; experience dictates choice. Of them, $MgCl_2$ and MS-222 (tricaine methanosulfonate) are generally useful, menthol is especially effective, and procaine and its relatives are promising.

Somewhat hypotonic (6%) $MgCl_2$ with the pH adjusted to 8–8.2 narcotizes many ascidians that have relaxed of their own accord in bowls of clean seawater. Add this solution slowly to replace the seawater, or gently transfer relaxed specimens into it. Do not dissolve $MgCl_2$ crystals directly into the seawater holding the specimens; animals rarely relax well in the resultant hypertonic solution. MS-222, usually used to narcotize fish, relaxes small ascidians. Tiny amounts suffice. Dissolve a fine forceps' "pinch" of MS-222 powder in about 20 ml of seawater and adjust the pH to 8 (since MS-222 is very acidic); use a drop or two of this freshly prepared solution per several milliliters of seawater containing the specimens. Menthol relaxes many ascidians, too. Scatter menthol crystals onto the water that holds the specimens; or, better, add to the water a few drops of a saturated solution of menthol dissolved in 95% ethanol. Cover the bowl to force the menthol fumes into solution.

Other anesthetics have occasional success with ascidians. These alternatives include simple anoxia, nembutal (usually in combination with $MgCl_2$, MS-222, or propylene phenoxetol), novocaine or tetracaine, chloral hydrate in combination with menthol, nicotine dissolved from shreds of tobacco, and even CO_2 introduced as dry ice. Doubtless, novelties like tiny dosages of some smooth-muscle relaxants now used in gastrointestinal surgery merit consideration, too, especially with recalcitrant taxa like *Eudistoma*, but only when ordinary means do not suffice.

Specimens are ready to be fixed when their apertures do not respond to slight pokes with a needle. An effective relaxant should anesthetize most ascidians thoroughly within an hour; but allowing another hour is sometimes prudent, and some large, tough solitary ascidians may require 4–8 hours' narcotization.

A fixative for taxonomy must preserve anatomy accurately, even on a microscopic scale, but it need not fix cellular structure very well or prepare tissues for histological staining. The standard taxonomic fixative for ascidians is 10% formalin made up with seawater, although Van Name (1945) urged a far weaker solution (2–2.5%). To avoid an excessively hypertonic fixative, G. Lambert (pers. comm.) prepares 10% formalin from 425 ml seawater, 25 ml tapwater, and 50 ml stock solution of formalin (which is 37% formaldehyde). Buffer the stock formalin solution or the 10% working solution with sodium borate or $CaCO_3$, lest its acidity dissolve the calcareous spicules that help distinguish some ascidians, such as didemnids; a tablet of drugstore antacid ($CaCO_3$) conveniently neutralizes more than a pint of formalin. Specimens should be fixed for at least 48 hours before being dissected. One must treat formalin with care; it is used here, after all, to kill and fix. But it is an excellent fixative for ascidian taxonomy—far better than ethanol, which ruins the animals' delicate and complex fresh tissues.

Small colonies, thin ones, and small solitary ascidians, once relaxed, can be fixed whole simply by immersing them in 10% formalin. Gently invert and swirl the jar occasionally to hasten penetration. A massive colony may need to be sliced judiciously to let the formalin enter it quickly. If they must be made, cuts into compound colonies should be wide apart and perpendicular to the surface that carries the zooids' apertures, to lessen damage to the zooids. If need be, one can carefully inject additional fixative through the apertures of large solitary ascidians to flood the pharynx and atrium.

Of course, immersing ascidians in 10% formalin dilutes the fixative even further (with the water in the specimens). To avoid this, P. Kott (pers. comm.) adds buffered stock formalin directly to the jar of seawater that *already* holds the narcotized specimens. To do this, top off the jar's entire contents—the specimens and the water around them—with 10% of their combined volume; this assures a resultant solution that is 10% formalin and 90% specimens-and-seawater. Although the solution is somewhat hypertonic, its convenience in the field (one need bring along just a supply of full-strength stock formalin) and the assurance of easily achieving a roughly 10% working solution may outweigh that disadvantage.

Anesthetization and fixation should begin as promptly as possible after collection, to avoid at least some of the damage to living specimens which is bound to accompany being dislodged, bagged, and transported. This is particularly advisable in tropical heat. Although a toxin like formalin should not be used in the habitat of the animals themselves, one often can anesthetize and fix specimens nearby.

Specimens can be kept in 10% buffered formalin for years; it is not only an excellent fixative but also a fine preservative. To avoid inhaling fumes, examine specimens in tapwater or (better, for its tonicity) seawater, then return them within a few hours to the formalin preservative. Although many museums store ascidians in 70% ethanol, we cannot recommend this. Ethanol is volatile, flammable, and of unpredictable composition when denatured; it makes tissues brittle, bleaches natural pigments, and often imposes constraints and complications if one later stains specimens for close examination. Just leaving specimens in the formalin that fixed them avoids all this.

Alcoholized specimens can be examined in tapwater and returned afterward to 70% ethanol, but the strong convection currents that accompany such transfers can disrupt fine anatomical structures of large or especially delicate specimens. Instead, we advise transferring specimens quickly through a few graded dilutions of ethanol or even, despite the fumes, examining them in 70% ethanol, if the work is brief—or one does not have to drive soon after!

Delicate or tiny ascidian structures, such as stigmata or the ciliated slit of the dorsal tubercle, can be very hard to see. A good dissecting microscope is essential for most work, and only a compound microscope allows examination of some fine structures. Transmitted light reveals many details not visible under reflected light. And, under reflected light, using a background of black or dark red wax may help one see details (e.g., the pyloric gland in polycitorids) that cannot be seen against a lighter background.

Above all, staining helps. A drop or two of 1% methylene blue or toluidine blue in the examining medium often will stain transparent structures adequately for examination even though the color will not last in wholemounts. If this does not work or permanent stains are needed, standard books on microtechnique (e.g., Weesner 1960; Galigher and Kozloff 1964; Humason 1979) provide directions for preparing wholemount stains that work, as well, on dissections. Alcoholized specimens can be colored with Grenacher's alcoholic borax carmine. Formalin-preserved specimens dissected in tapwater stain especially clearly with potassium-hemalum (C. and F. Monniot 1972; C. Monniot et al. 1991). Simply bathe the specimen or tissue with a few drops of the appropriate stain for a few minutes until the tissues gain the desired contrast, then rinse to hold the coloration at roughly that intensity. Vázquez Otero (1993) summarizes the Monniots' hemalum-staining protocol and the subsequent steps they recommend for preparing wholemounts of ascidians embedded in "Araldite."

DISSECTION OF ASCIDIANS FOR IDENTIFICATION

Alive, even relaxed ascidians defy dissections that try to do more than simply remove the body from the tunic or, at most, open the pharynx. The flimsy tissues of most solitary ascidians and all colonial ones quickly tear or turn mushy or sticky, thwarting surgical examination. The hardening of tissues that accompanies fixation is essential for satisfactory dissections.

With the permission of the University of California Press, we reprint here, with revised portions bracketed, Abbott's instructions (1975:640–642) for dissecting ascidians in order to identify them:

> Solitary ascidians are conveniently dissected in the following manner. Insert one point of a scissors into the oral siphon and cut downward along, or very slightly to the right of, the median line. (For most solitary ascidians the median sagittal plane is defined roughly by the positions of the apertures: a plane passing downward through the centers of the two apertures will divide the body bilaterally.) This cut longitudinally splits or closely parallels the endostyle. Continue the cut around the base of the body, cutting through the tissues of the tunic, body wall (mantle), and pharynx, until the two halves can be spread apart like an opened book. Remove the body from the tunic, pin the opened animal down in a wax-bottomed pan or petri dish, and cover it with water. [First removing the supple body from its stiffer tunic will often make the above cut easier. To do this, cut through the tunic near the base and continue around the body and up to each siphon tip; then grasp the body's siphon tips and carefully draw them from their tight attachment to the tunic, freeing the body altogether.] The inner surface of the pharynx . . . shows many features of taxonomic importance. Very dilute dyes applied now will enhance them. To expose the stomach, intestine, and gonads of most simple ascidians, cut the numerous fine tissue strands which attach the pharynx to the mantle and other organs on each side, and fold back the pharynx. In *Ciona* the gut loop and gonads lie posterior to the pharynx and can be seen without this further dissection.
>
> Colonial ascidians with large zooids can be dissected like solitary ascidians. For those with small zooids completely buried in a massive common tunic, usually one need only remove the zooids from the tunic in order to examine them for identification. Select well-expanded individuals. [Removing individual zooids by very careful excavation with fine forceps and microscissors is slower but far less damaging than older methods of just slicing pie wedges from the colony, and it is more likely to provide whole zooids. Use very fine forceps and iridectomy scissors to tease the zooid's whole length free of its bed of tunic and gently ease it free, being careful not to leave its abdomen or post-abdomen behind.]

The aforementioned operations should be done on specimens held in a small bowl of seawater. Transfer opened bodies or selected zooids to a small wax-bottomed petri dish for subsequent examination or dissection. If possible, select sexually mature zooids when examining colonial species. With practice, one can cut open the pharynx even of a small colonial zooid along the endostyle; only this surgery lets one count its oral tentacles and stigmatal rows accurately. Almost every taxon poses its own challenges (e.g., unusually fragile zooids or a glutinous tunic, bulky larvae to be removed from the atrium before opening the pharynx), but skill comes with persistence, especially if a few specimens can be given over to solving surgical problems. Watchmaker's (#5) forceps and very fine iridectomy or neurosurgical scissors are expensive but crucial tools for all dissections. Pin open dissections, even of large specimens, with fine insect pins or even tinier stainless steel "nadeln" ("minutien") needles. After examination, specimens can be preserved in vials or as wholemounts in Canada balsam or (much better) in modified "Araldite" electron microscopy mountant (cf. Vázquez Otero 1993).

KEY TO THE PRINCIPAL SHALLOW-WATER ASCIDIAN FAMILIES OF THE WORLD

We adopt Kott's (1969) taxonomy of families and subfamilies, except that, in agreement with C. Monniot (1965), our key includes in the family Pyuridae several puzzling genera (none yet found in Hawai'i) with spiral or irregular stigmata and even with filiform oral tentacles. Starred (*) families or parts thereof are known from Hawaiian waters.

1	Solitary ascidians	2
	Colonial ascidians; zooids buried together in common tunic ("compound" ascidians) or separate from each other except for basal tunic ("social" ascidians)	10
2(1)	Gut loop, except rectum, located posterior to pharynx	3
	Gut loop located mostly beside pharynx	4
3(2)	Gut loop short, coiled transversely; tunic clear gray to greenish in life	CIONIDAE*
	Gut loop elongate, looped longitudinally; tunic often blue, pink, or purple in life	DIAZONIDAE (part)
4(2)	Pharyngeal stigmata curved, often arranged in spirals	5
	Pharyngeal stigmata straight, not arranged in spirals	7
5(4)	Gut loop mostly on left side of pharynx; pharynx with (or, in a few uncommon molgulid genera, without) large longitudinal folds	6
	Gut loop partly or wholly on right side of pharynx; pharynx without large longitudinal folds	CORELLIDAE (part)*
6(5)	Conspicuous "renal" sac in right body wall; pharyngeal folds absent in a few genera	MOLGULIDAE
	No "renal" sac; pharyngeal folds always present	PYURIDAE (part)
7(4)	Pharynx with 4 or more longitudinal folds on each side	8
	Pharynx without longitudinal folds	9
8(7)	Pharynx with 4 folds on each side; oral tentacles filiform (unbranched)	STYELIDAE (subfamily Styelinae)*
	Pharynx with more than 4 folds on each side; oral tentacles pinnately branched (feathery, fernlike)	PYURIDAE (part)*
9(7)	Gut loop on left side of pharynx	ASCIDIIDAE*
	Gut loop on right side of pharynx	CORELLIDAE (part)
10(1)	Zooid body short and compact, not divided into 2 or 3 distinct regions; pharynx with at least 3 internal longitudinal vessels on each side	11
	Zooid body short to very elongate, divided into 2 or 3 distinct regions; pharynx lacks internal longitudinal vessels (except in Diazonidae)	13
11(10)	Zooids form systems that are oval clusters or elongate double rows; pharynx with 3 internal longitudinal vessels on each side	STYELIDAE (subfamily Botryllinae)*
	Zooids not arranged in systems; pharynx with 4 or more (rarely 3) partial or complete internal longitudinal vessels on each side	12

12(11) Lateral tunics of adjacent zooids usually distinct; gonad only in gut loop .... PEROPHORIDAE*

Tunics of adjacent zooids fused, at least at base; gonads on one or both sides of body, only partly (if at all) in gut loop .... STYELIDAE (subfamily Polyzoinae)*

13(10) Body has 3 parts: post-abdomen present .... POLYCLINIDAE*

Body has 2 parts: no post-abdomen (exc. some Diazonidae) 14

14(13) Transverse pharyngeal sinuses bear internal longitudinal vessels or distinct papillae .... DIAZONIDAE (part)

No internal longitudinal vessels or papillae on transverse pharyngeal sinuses 15

15(14) Colonies compound or social; oral and atrial apertures both open at tunic surface, no common cloacal cavities in tunic; 2 to many rows of pharyngeal stigmata 16

Colonies compound; only oral apertures open at tunic surface, atrial apertures open into common cloacal cavities; never more than 4 rows of stigmata 17

16(15) Colonies compound: zooids entirely or mostly (at least abdomen) embedded in a common tunic; buds arise from abdominal constrictions .... POLYCITORIDAE*

Colonies are clusters of zooids protruding their entire length from a thin basal sheet of tunic; buds arise from stolonic vessels that grow out on substrate from bases of zooids .... CLAVELINIDAE (subfamily Clavelininae)

17(15) Embryos brooded in pouch extending from thorax; tunic lacks calcareous spicules .... CLAVELINIDAE (subfamily Holozoinae)

Embryos brooded in tunic's common cloacal cavities; tunic often contains stellate calcareous spicules .... DIDEMNIDAE*

HAWAIIAN ASCIDIANS

Each of the following accounts of ascidian families describes family-wide traits and comments individually on species that occur regularly in Hawai‘i's shallow-water habitats. Hawaiian representatives of most families are diverse enough to warrant keys. Descriptions of rarely encountered species or ones that have been found only in outlying or deep-water sites are limited to these keys. Unless stated otherwise, colors refer to living animals, since fixed ones often lose or at least change their color.

These accounts slight worldwide variation, because they are meant to help identify known Hawaiian ascidians according to traits they show here. Identifying known species newly found in Hawai‘i and recognizing wholly undescribed species as such are other, larger tasks, beyond the scope of this volume. The technical guidance for those efforts lies in expedition reports, regional surveys, and accounts of major museum collections. For Indo-Pacific forms we recommend Van Name (1918, 1945) and Tokioka (1967) and recent and forthcoming accounts by Kott on the ascidians of Fiji (1981a) and Australia (1985, 1990, 1992a, in prep.), by the Monniots on French Polynesia (1987) and New Caledonia (1987 et seq.), and by

Nishikawa on Micronesia and Melanesia (1984, 1986) and Japan (1990, 1991, 1992). In addition, C. Monniot et al. (1991) have written and illustrated a superb, nontechnical book on the biology of ascidians generally and the ecology and diversity of New Caledonian species in particular.

Order ENTEROGONA, Suborder APLOUSOBRANCHIA

Family **Polyclinidae**

Polyclinid zooids are divided into three parts: thorax, abdomen, and post-abdomen. The post-abdomen contains the gonads and heart. All polyclinids known from Hawai'i have many rows of pharyngeal stigmata. Colonies form by repeated strobilation of their zooids; the zooids undergo transverse constrictions of the post-abdomen and abdomen to form little vesicles which, along with the thoracic remnant, regenerate to form new blastozooids.

The Polyclinidae have two subfamilies. In the Euherdmaniinae, each individual zooid opens its atrial siphon directly to the sea. In some species individual zooids project, each in its own sheath of tunic, above the dense, bouquet-like colony's basal mass of common tunic; in others the zooids are more substantially or even completely embedded together, usually in lobes. In the Polyclininae, the completely embedded zooids form systems in which they open their atrial apertures, beneath atrial languets, into common cloacal cavities. Despite their diversity elsewhere, only four species in two genera of Polyclininae—*Polyclinum* and *Aplidium*—have been found in Hawai'i. *Aplidium*, especially, appears to be peculiarly uncommon here.

1 Post-abdomen short and oval, attached to abdomen by slender neck; outer stomach wall smooth except for a "seam" down one side .... *Polyclinum* 2
Post-abdomen elongate, not separated from the abdomen by a constricted neck; outer stomach wall with longitudinal ridges and grooves .... *Aplidium* 3
2(1) Pharynx usually with 12–13 rows of stigmata .... *Polyclinum vasculosum* Pizon, 1908
Pharynx usually with 14–18 rows of stigmata (Fig. 2a) .... *Polyclinum constellatum* Savigny, 1816
3(1) 15–20 rows of stigmata, 9–14 stomach grooves; colonies soft violet in life, gray-brown after preservation; known only from Gardner Pinnacles, 15 m depth (Fig. 2c) .... (?)*Aplidium crateriferum* (Sluiter, 1909)
13–15 rows of stigmata, 18–23 stomach grooves; dredged between Maui and Lāna'i, 63–120 m depth .... *Aplidium pliciferum* (Redikorzev, 1927)

Polyclinum constellatum (Fig. 2a, b) is the commonest inshore species of this family. Smaller colonies (1–3 cm across) form almost spherical cushions, attached by a restricted undersurface to the substrate (Kott 1992a); larger, massive colonies (5–15 cm across) are hemispherical or flattened. The colony surface may

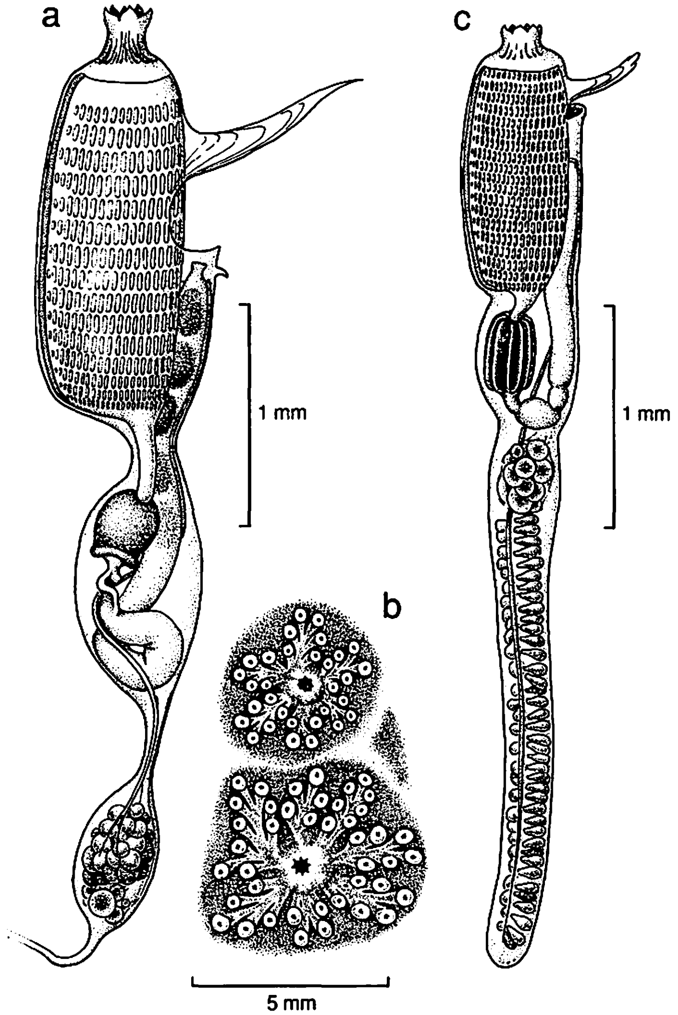

Figure 2.—a, *Polyclinum constellatum* zooid viewed from left side, showing atrial languet, smooth stomach, and constriction between abdomen and post-abdomen; b, two systems of a *P. constellatum* colony, each with many oral apertures, converging common cloacal canals, and single common cloacal aperture; c, *Aplidium crateriferum* zooid viewed from left side, showing atrial languet, stomach grooves, and lack of constriction between abdomen and post-abdomen.

be clean or lightly encrusted with sand or silt. The tunic has a stiff, jelly-like or cartilaginous consistency and a tough, thin outer "skin." Living colonies range from light yellow-brown or reddish through dark brown and maroon to purplish or black, with whitish or golden yellow zooids arranged in prominent systems. The common cloacal apertures of these systems rise like low volcanoes from the colony. The zooids lie close to the colony's upper surface, with their thoraxes "heavily covered with white pigment granules" (G. Lambert, pers. comm.). Deeper in the colony the clear tunic contains long vascular processes extending from the zooids' post-abdomens. This species is sometimes abundant in Kāne'ohe Bay, Pearl Harbor, Honolulu Harbor, and the Ala Moana yacht basin, growing on dock pilings, floats, dead coral, or coral rubble lying on sandflats. In more exposed places, such as Honolulu's Black Point, small colonies grow on the undersurfaces of rocks. The species is distributed virtually worldwide in tropical seas and extends into subtropical waters.

Tokioka (1967) reports *Polyclinum vasculosum* from Pearl Harbor. It closely resembles *P. constellatum*, but its zooidal systems tend to be less prominently visible from the outside; in many colonies the zooids' thoraxes lack a coating of white pigment granules. Colonies of *P. vasculosum* adhere more broadly to the substrate than do the cushions of *P. constellatum* (Kott 1992a). In Hawai'i these two species appear to intergrade; in fact, many Hawaiian specimens deemed "*P. vasculosum*" may be variant colonies of *P. constellatum*. *Polyclinum vasculosum* has been reported widely in the tropical western Pacific: the Malay archipelago, Vietnam, the Philippines, the Marianas, and the Amami Islands of Japan.

Further studies of the genus *Polyclinum* are needed to sort out what Van Name (1945) called its many "unsatisfactorily distinguished species," including several confusingly variable tropical ones. The genus *Aplidium*, neglected in this account because of its meager and entirely subtidal records from Hawai'i, also merits a thorough review.

Family **Polycitoridae**

Kott (1969, 1990) restricts the family Polycitoridae to a half-dozen genera of rather similar design. C. and F. Monniot (1972) and C. Monniot et al. (1991) include many genera that Kott places in other aplousobranch families—for example, *Distaplia*, *Sycozoa*, *Clavelina*, and *Pycnoclavella*. Kott and the Monniots agree that *Polycitor*, *Cystodytes*, and *Eudistoma*, diverse and widespread in the tropics, are all polycitorids, but of these three genera only *Eudistoma* has been found in Hawai'i. The name of this genus has long been disputed. Berrill (1950) proposed that *Eudistoma* be made a subgenus of *Archidistoma*, noting their similar larvae, modes of budding, and (he felt) a continuity between these two genera in the extent to which zooids are embedded in the tunic. Kott (1990), after gradually (1952a, 1957a, 1957b) moving away from Berrill's position, gives both *Archidistoma* and *Eudistoma* full generic status without further reference to Berrill's case. So do Millar (1975, 1977), Tokioka (1967), Nishikawa (1984, 1990), F. Monniot (1969, 1972, 1983b, 1988), C. and F. Monniot (1972, 1987), and C. Monniot et al. (1991)—a consensus that provides, as well, a guide to most of the recent literature about this difficult genus.

The Monniots note (pers. comm.) that, contrary to Berrill's assertion, there is a distinct difference rather than a continuum between *Archidistoma* colonies, whose zooids stand out from each other, and *Eudistoma* colonies, with their much more deeply embedded zooids. They agree with Kott that, even if similarities were to argue for synonymy, sheer practicality should make us pause before renaming the many *Eudistoma* species to make them conform by formal priority with the few now called *Archidistoma*.

In his key to California ascidians, Abbott (1954) called the genus *Eudistoma*, but in the next edition (1975) he assigned all of California's *Eudistoma* species without comment to the genus *Archidistoma*. In this account of Hawaiian ascidians we treat *Eudistoma* as a legitimate genus to which the Hawaiian species belong, and *Archidistoma* as another genus none of whose species have been found in our (or California's) waters.

Eudistoma zooids have only three rows of stigmata and a smooth-walled stomach and are buried more or less completely in the colony's common tunic, into which their strong musculature pulls them even more deeply when they are disturbed. Both oral and atrial apertures open to the colony surface, but individuals usually form rudimentary systems of 6–10 zooids radiating in a circle, with their oral siphons at the periphery and their atrial apertures close together at the center. Budding is by abdominal strobilation; the resultant abdominal vesicles and the thoracic remnant regenerate into daughter blastozooids.

Sluiter (1900) described three species of *Eudistoma* from Laysan Island. None has been found in the main Hawaiian group, nor have they been sought anew even around Laysan Island. A fourth species, *E. angolanum*, was recently found at Gardner Pinnacles, between Laysan and Necker Islands.

1 Preserved colonies with conspicuous dark pigment in tunic or
zooids or both 2
Preserved colonies yellowish brown to transparent; dark
pigment lacking in zooids and tunic 3
2(1) Zooids with darkly pigmented thorax; 6 stigmata in each row
on each side of pharynx; known only from Laysan Is.
.... *Eudistoma austerum* Hartmeyer, 1909-11
[formerly *Distoma fusca* Sluiter, 1900]
Zooids with yellowish thorax, lacking dark pigment; 10 or
more stigmata in each row on each side of pharynx; found
at Gardner Pinnacles; also found in Palau, Gilbert Is.,
Marianas Is., Australia, Vietnam, and West Africa (Fig. 3)
.... *Eudistoma angolanum* (Michaelsen, 1914)
3(1) Colonies form flat-topped lobes about 8 mm across, borne on
short yellowish stalks; 5–6 stigmata per row on each side
of pharynx; known only from Laysan Is. ...... *Eudistoma parva* (Sluiter, 1900)
[not *E. parvum* (Oka, 1927)]
Colonies form encrusting masses from which arise numerous
columnar lobes, each bearing one to many zooids that
open at the lobes' flat tops; 13–15 or more stigmata per
row on each side of pharynx; known from Laysan Is.; also
found in Palau, Philippines, Indonesia, Vietnam, Thailand,
Australia, Society Is .... *Eudistoma laysani* (Sluiter, 1900)

Eudistoma angolanum forms lobate patches a few millimeters to about 5 cm across. Living colonies are light pink, yellow, orange, or green. The siphons of relaxed zooids protrude slightly, giving the colony an odd, prickly look (Fig. 3b). Preserved colonies have a smooth surface (Fig. 3c), since the zooids always contract somewhat, and the upper 1–3 mm of the tunic turns dark purplish brown. The colony surface is free of sand, but sand grains and fecal pellets impregnate the tunic. This species grows abundantly on dead coral at a depth of about 15 m at Gardner Pinnacles.

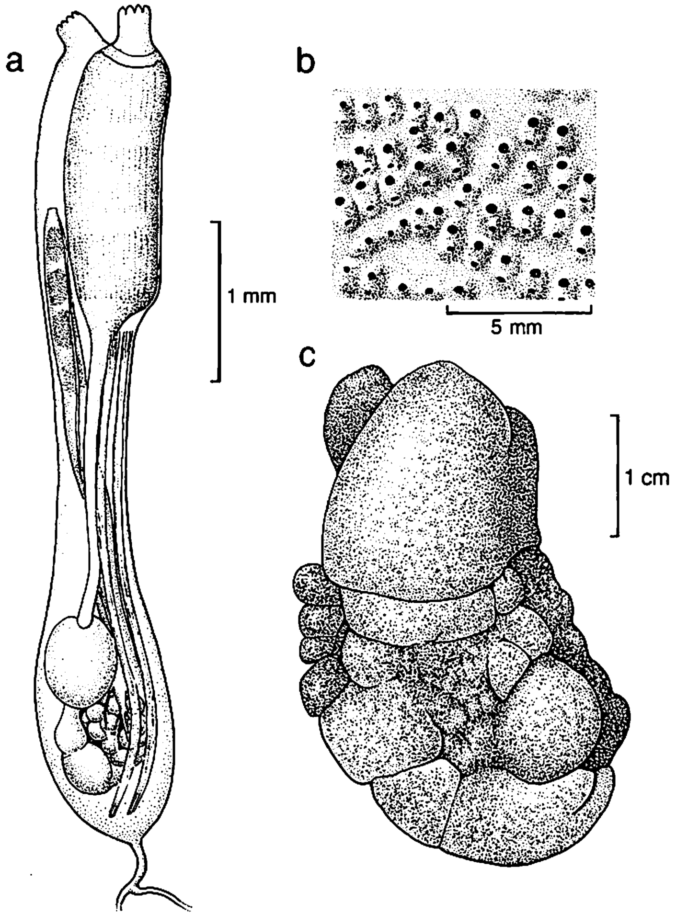

Figure 3.—*Eudistoma angolanum:* a, zooid viewed from right side, with 3 rows of stigmata (characteristic of genus), long esophagus, only 2 body regions, and strong longitudinal mantle muscle bands; b, surface of living colony (drawn from photograph); zooids, arranged somewhat parallel to each other, are entirely embedded in common tunic; both their oral and atrial apertures open individually to the colony surface; c, contracted colony; insufficient relaxation before fixation usually leads to specimens of this appearance, with distorted zooids.

Family **Didemnidae**

Although didemnid colonies may be large, the zooids are the smallest of aplousobranchs, rarely exceeding 1 mm. Colonies are permeated by extensive common cloacal cavities (Fig. 4), where the weak excurrents of individual zooids combine into substantial flows pouring through one or more common cloacal apertures. In relaxed colonies each of these apertures is raised on a delicate, chimney-like protrusion of the tunic. Didemnids usually form patches or mounds; some species divide into sibling colonies after growing to only a few millimeters across, whereas others maintain their colonies intact.

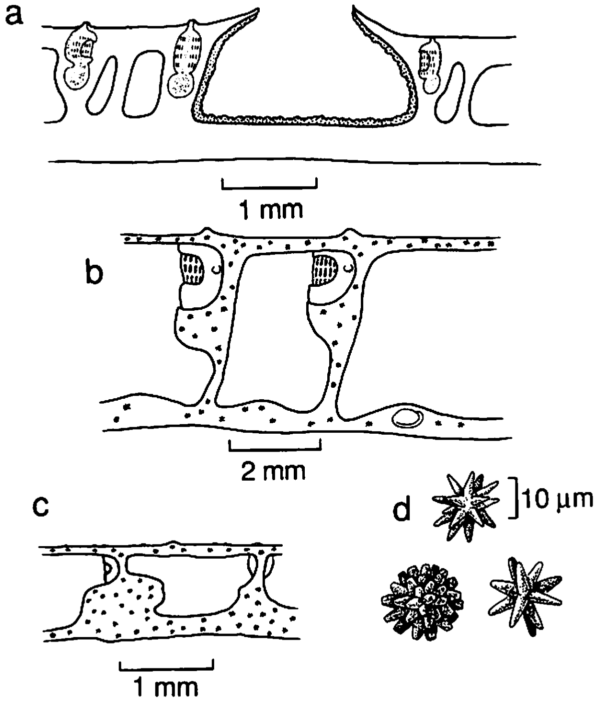

Figure 4.—a, schematic cross section of a didemnid colony (*Diplosoma similis*, modified from Kott 1982); zooids lie in pillar-like strands of tunic around common cloacal cavities; the tunic is permeated by labyrinthine common cloacal canals, shown here in cross section; b, schematic cross section of *Lissoclinum fragile* colony; zooids are suspended in slender strands of tunic surrounded by large common cloacal canals (after Eldredge 1967); c, schematic cross section of *Didemnum moseleyi* colony; thick tunic elements support zooids amid common cloacal canals (after Eldredge 1967); d, calcareous spicules from various didemnid colonies.

Most didemnids have minute, stellate, calcareous spicules (Fig. 4d) in the tunic. Spicule shape, distribution, and density vary greatly and are among the characters used to distinguish species in this family. They make virtual crusts in some *Didemnum* and *Trididemnum* species, but they are entirely absent in *Diplosoma*. When abundant, spicules may render colonies opaque and uniformly or variably blanched, despite the presence of pigments. In contrast, *Diplosoma* colonies are dark or clear and translucent or even transparent. *Prochloron* or unicellular algal symbionts living on the colony surface, in the common cloacal cavities, or in the tunic itself give many didemnids a bright green or pale reddish color.

A didemnid zooid (Fig. 5) consists of a thorax and abdomen linked by a sharply constricted waist. The atrial aperture may be borne on a full siphon, lie beneath an atrial languet, or form merely a gaping hole in the dorsal mantle. In many didemnids a prominent retractor muscle projects posteriorly from the posterior-ventral region of the thorax. The abdomen contains the gut loop and gonads. The sperm duct is either straight (in *Diplosoma*, Fig. 5a, c) or spirally coiled (Fig. 5b, d–f).

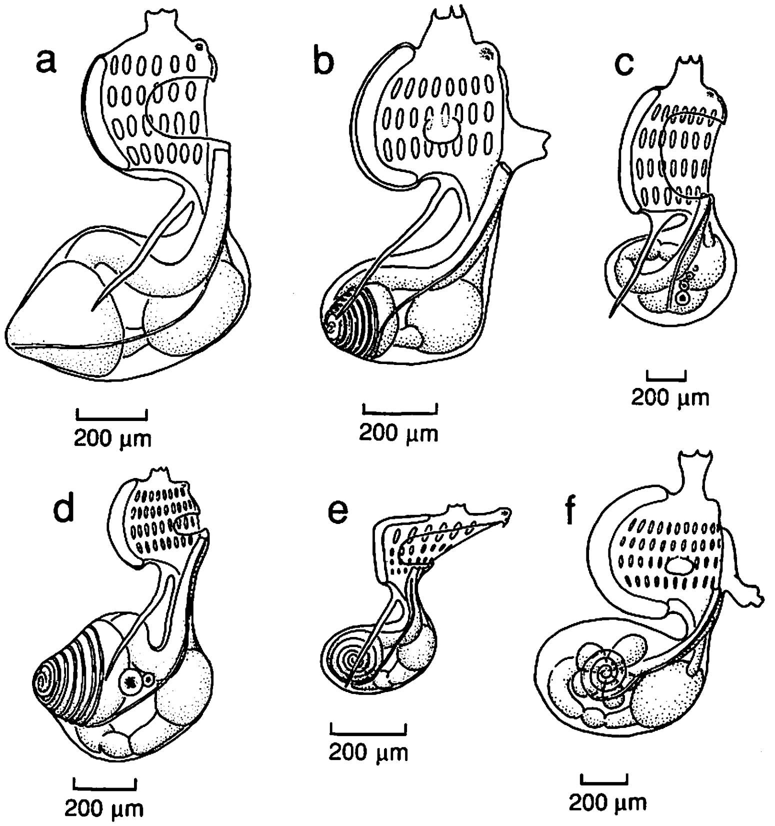

Figure 5.—Didemnid zooids viewed from left side: a, *Diplosoma similis;* note straight sperm duct, gaping atrial aperture; b, *Trididemnum savignii;* note coiled sperm duct, 3 rows of stigmata, and lateral organ on thorax; c, *Diplosoma listerianum;* d, *Didemnum edmondsoni;* e, *Didemnum granulatum;* f, *Leptoclinides madara.* (some drawings modified from Eldredge 1967, after subsequent accounts of these species)

In forming their colonies, didemnids bud in an odd way: a zooid protrudes one bud rudiment (which becomes a new abdomen) from the thoracic end of its esophageal region and simultaneously another (which becomes a new thorax) from the abdominal end of that region. This results in a zooid with two thoraxes and two abdomens which eventually splits into daughter zooids.

Spicules form especially abundantly where a zooid's epidermis adheres tightly to the tunic. Spiculate didemnids have a saccular "lateral organ" on either side of the thorax (Fig. 5b, f). These outpouchings have long been associated with localized spicule formation, but precisely what they do is still debated, as is whether tunic spicules originate intra- or extracellularly. Ballan-Dufrançais et al. (1995) clearly show by TEM the intracellular origin of spicules in the epidermal cells of the lateral thoracic organs in *Trididemnum*. Some species with densely spiculate tunics seem to lack lateral organs, but Lafargue and Kniprath (1978; Kniprath and Lafargue 1980) believe that all spiculate species do have them, even if often very small and inconspicuous ones that may be lost when fixed zooids are removed from the tunic for examination. If this is so, this artifact of handling may account for the disorderly reported taxonomic distribution of lateral organs.

Though often inconspicuous, didemnids are the most common and diverse ascidians in Hawaiian and many other tropical waters. They often live in calm habitats on floats, docks, and pilings. Some species exploit the intricate surfaces of living reefs and coral rubble, and a few even grow inside large sponges and on the backs of dromiid (sponge) crabs. Many species occur most often in shallow water, although *Leptoclinides hawaiiensis* has been taken from 64 m in the ʻAuʻau Channel.

Identifying didemnids can be very difficult. First, one should note the colors of living specimens; preserved colonies soon fade and leave merely the whiteness of the tunic's spicules. In fact, careful attention to field traits narrows most identifications to a few species, even though they may be in different genera. This step can be followed by close examination of colony and zooidal structure, spicule shape and density, and other details, to get the identity to species. It is usually almost impossible to remove the tiny zooids intact from the tunic, but it is sometimes not too difficult to reconstruct "conceptual" zooids and discern their diagnostic traits by assembling zooid fragments, like pieces of a jigsaw puzzle.

The following key to the shallow-water didemnids of Oʻahu has been modified from a key to central Pacific didemnids by Eldredge (1967). We give *Lissoclinum*, once a subgenus of *Diplosoma*, full generic status, and we have changed Eldredge's identification of Hawaiian *Diplosoma* species in light of challenges by Rowe (1966), Kott (1980), and C. and F. Monniot (1987). Our key neglects larval traits, which surely are crucial to deciphering didemnid relationships. But we believe that this gap does not diminish the key's usefulness as a naturalist's guide to the likely identity of didemnids known to occur here. Figure 5 can be used as a glossary: it illustrates for just some species traits that our key uses for many others as well.

1	Sperm duct coiled; 3 or 4 rows of stigmata	2
	Sperm duct straight; always 4 rows of stigmata	9
2(1)	3 rows of stigmata	*Trididemnum* 3
	4 rows of stigmata	4
3(2)	Common cloacal cavities mostly around thoraxes but with extensions below zooids; 6–7 sperm duct coils; no lateral organs; color alive and preserved dark brown; tunic usually aspiculate, but may have tiny scattered spicule clusters only on surface; spicule diameter 10–25 µm	*Trididemnum profundum* (Sluiter, 1909)
	Common cloacal cavities only thoracic; 7–9 sperm duct coils; lateral organs present; color alive and preserved light tan or white; tunic spiculate; spicule diam. 20–50 µm (Fig. 5b)	*Trididemnum savignii* (Herdman, 1886)

4(2) Atrial siphon posteriorly directed; testes have multiple lobes; zooids dark; 5–6 sperm duct coils; no retractor muscle; living colonies tan or gray, with occasional orange streaks; spicule diam. 50–80 µm (Fig. 5f) .... *Leptoclinides madara* Tokioka, 1953 [*Leptoclinides rufus* (Sluiter, 1909) in Eldredge 1967, reassigned by Kott 1981a]

Atrial aperture lacks siphon or languet; usually single testis .... *Didemnum* 5

5(4) Thorax shape "typical": stigmatal rows equal in height 6

Thorax distorted; first stigmatal row at least twice as tall as other rows; living colony light lemon-yellow, with scattered papillae; spicule diam. 20–30 µm (Fig. 5e) .... *Didemnum granulatum* Tokioka, 1954 [*D. pele* Eldredge, 1967 in Eldredge 1967, reassigned by Kott 1981a; see also *D. moseleyi* (8)]

6(5) Atrial aperture a small, oval, narrow slit, or wide but not gaping broadly over sides of pharynx; tunic spiculate 7

Atrial aperture wide, opening broadly over sides of pharynx; living colony black and aspiculate, but white pigment granules are scattered through deeper portions of colony .... *Didemnum elikapekae* Eldredge, 1967

7(6) Oral siphons level with colony surface; living colonies not darkly colored; moderately to densely spiculate tunic 8

Oral siphons raised as distinct little lobes; living colony velvety purple, black, or maroon, with distinct white oral lobes; spicules dense, diam. 20–30 µm (Fig. 5d) .... *Didemnum edmondsoni* Eldredge, 1967

8(7) Atrial aperture wide; colony surface often with papillae near oral siphons; living colonies variably colored pink, yellow, orange; spicule diam. 25–40 µm, plus some tetrahedral spicules 100µm across (colony profile, Fig. 4c) .... *Didemnum moseleyi* (Herdman, 1886) [tentatively assigned to *D. cuculliferum* (Sluiter, 1909) and/or *D. granulatum* Tokioka, 1954 by Kott 1981a]

Atrial aperture a short, narrow slit; surface of colony never papillose; living colony white or off-white; spicule diameter 20µm, with numerous tiny rays .... *Didemnum candidum* Savigny, 1816

9(1) Aspiculate; without lateral organs .... *Diplosoma* 10

Spicules dense especially at colony surface, small (10–20 µm) and burrlike; living colony white (colony profile, Fig. 4b) .... *Lissoclinum fragile* (Van Name, 1902) [*Diplosoma (Lissoclinum) fragile* in Eldredge 1967]

10(9) Algae present in common cloacal cavities; zooids usually unpigmented; living colony dark green with turquoise oral rings (Fig. 5a) .... *Diplosoma similis* (Sluiter, 1909) [*D. virens* (Hartmeyer, 1909–11) in Eldredge 1967, reassigned by Kott 1980]

Colony alga-free; zooids have dark tan or gray-orange
abdomens; living colony tan or gray with white pigment
granules scattered over surface (Fig. 5c)
.... *Diplosoma listerianum* (Milne-Edwards, 1841)
[*D. macdonaldi* Herdman, 1886 in Eldredge 1967,
but species synonymized by Rowe 1966]

The two species of *Diplosoma* in this key are important research animals, so changes in the names by which they have long been known in Hawai'i deserve comment. Kott's (1980) correction of Eldredge's *D. virens* to *D. similis* is an excellent example of how diverse traits, sometimes trivial by themselves, can combine to reveal the identity of puzzling taxa: (1) *Diplosoma virens* forms isolated or massed, roughly ovoid colonies, each only 1–2 cm across and 2–5 mm thick; *D. similis* spreads sheetlike colonies that are less than 2 mm thick. (2) *Diplosoma virens* is bright, if pale, green, often with a darker green margin; *D. similis* ranges through dark or bright shades of green, blue, and turquoise, with a surface iridescence. (3) In *D. virens,* common cloacal canals extend deeper than the zooids throughout the colony; in *D. similis* these canals extend below the zooids only "in the centre of larger colonies." (4) A *D. virens* colony usually has one common cloacal aperture located centrally atop a cone of tunic; *D. similis* colonies have several "inconspicuous and sessile" common cloacal apertures. (5) In *D. virens* the tunic "is very tough and apparently fibrous"; in *D. similis* it "is firm and gelatinous, but is not hard or tough, and is easily torn." (6) *Diplosoma virens* colonies attach to the substrate only in a few restricted places on their undersurface; *D. similis* colonies "adhere closely to the substrate by the whole of their basal surface." (7) *Diplosoma virens* zooids have much longer retractor muscles than do *D. similis* zooids.

What has long been called *Diplosoma virens* in Hawai'i shows all the above traits of *D. similis*, except that it has a somewhat thicker tunic (to 4 mm) and carries its common cloacal apertures on "transparent, somewhat raised" sites (Eldredge 1967). These minor variations are attributable to geographic variation within the suite of traits that distinguishes *D. similis* from *D. virens*. *Diplosoma similis* occurs widely in Polynesia, Micronesia, Melanesia, and Indonesia and as far north as the Tokara Islands of Japan and south through Australia's Great Barrier Reef.

Diplosoma macdonaldi and several other similar purported species of this genus are actually *D. listerianum*. Rowe's (1966) synonymy is accepted by Kott (1981a) and F. Monniot (1983a). C. and F. Monniot (1987) confirmed the synonymy of mid-Pacific material by chromatographic comparison of French Polynesian with European material. *Diplosoma listerianum*, a hardy species, occurs virtually worldwide in tropical and temperate waters, especially as a fouling organism in the quiet waters of ports. It probably is continually carried in ships' ballast water from one harbor to another.

Order ENTEROGONA, Suborder PHLEBOBRANCHIA

Family **Cionidae**

The family Cionidae is an intriguing puzzle, for it seems to present traits from which many other ascidian families, and by implication lineages, can be derived—at least in theory. We have dealt briefly with this possibility in our introductory comments. What genera to include in the family—hence whether the family includes only solitary or also some colonial forms, and even whether the family should be considered in one order (Phlebobranchia) or another (Aplousobranchia)—remain vigorously debated. But Hawai'i's cionid fauna is so restricted—one species, *Ciona intestinalis*—that these comparative questions will not be settled by examining local forms.

The long, columnar thorax of *Ciona* contains an enormous pharynx; posterior to this are the transversely coiled gut loop, heart, and gonads. The pharynx has many internal longitudinal vessels, which bear curved papillae.

Ciona intestinalis (Linnaeus, 1767) (Fig. 6), the cionid commonly found in Hawai'i, often exceeds 8 cm in length. It attaches by its posterior end; its siphons project close to each other at the anterior end. The living animal feels soft and yielding, even gelatinous. Its translucent grayish green tunic reveals strong longi-

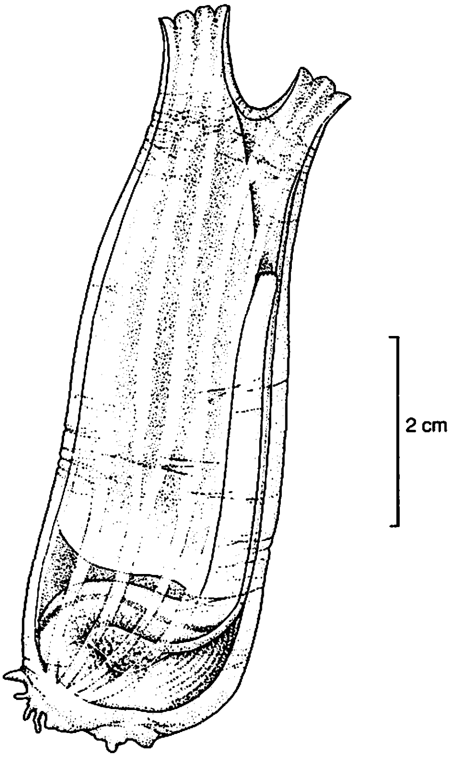

Figure 6.—*Ciona intestinalis* in its transparent tunic, through which longitudinal muscle bands, gut loop, and enterogonid gonad are visible.

tudinal muscle bands and a fine grid of pharyngeal sinuses and stigmata. The species occurs worldwide in temperate waters. It is a prominent fouling animal that exploits artificial surfaces in ports but does not spread much into less disturbed habitats. On O'ahu it is found on boat hulls and docks in such places as Pearl Harbor, Honolulu Harbor, and Kāne'ohe Bay.

Big, hardy, oviparous, and widespread, *Ciona intestinalis* has often been used in laboratory studies, especially in embryological and physiological research.

Family **Perophoridae**

Perophorids are "social" colonial ascidians. The zooids of a colony arise along branching stolons and are fairly clearly separated from each other, rarely buried together in a massive tunic. A perophorid zooid, even if elongated by a large pharynx, is very compact; its gut loop and gonad lie largely beside, not behind, its pharynx (Fig. 7a). The family comprises two common and widespread genera, *Perophora* and *Ecteinascidia*, both of which occur in Hawai'i. The two are distinguished by body size and the number of rows of pharyngeal stigmata. The small, rather spherical *Perophora* zooids have fewer than five complete rows of stigmata; the larger and more elongate zooids of *Ecteinascidia* have more than five stigmatal rows—a dozen or more rows in Hawaiian species.

What we identify tentatively as *Perophora annectens* Ritter, 1893 forms delicately ramifying colonies (Fig. 7b) of pale green to gray, rounded zooids 2–5 mm in diameter. Our uncertainty stems from the Hawaiian species' resemblance to *P. japonica* Oka, 1927 and from the evident confusion that attends the separation of *Perophora* species in the Pacific. In the colder waters of the northwestern North American coast, *P. annectens* forms much denser, more "compound" colonies than our "social" *Perophora* does, but the species' colonies are rather loose, as

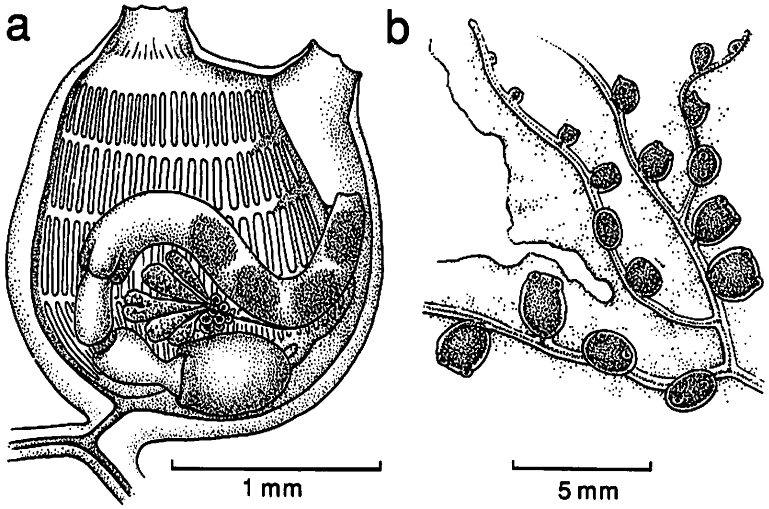

Figure 7.—*Perophora annectens:* a, zooid with left tunic and mantle cut away; b, part of a young colony.

ours are, in the warmer waters of southern California. Nishikawa (1991) believes that *P. annectens* is very closely related to *P. japonica*, if not identical.

Zooids of the Hawaiian *Perophora* (Fig. 7a) have four complete rows of stigmata (occasionally an incomplete fifth row); each row has 18–25 stigmata per side. Transverse blood sinuses bear seven to nine mostly T-shaped papillae. In younger zooids the pharynx lacks internal longitudinal vessels. Though many papillae fail to join their arms to form internal longitudinal vessels even in mature zooids, vessels do form here and there to bridge two or three adjacent rows of stigmata. The presence of even these incomplete internal longitudinal vessels appears to rule out *P. annectens*, which according to most accounts lacks such vessels altogether. But Nishikawa (1991) reports the occasional presence of incomplete internal longitudinal vessels in the species. An incomplete fifth row of stigmata appears, too, from time to time in *P. annectens* from North America. The gonad lies to the left of the pharynx, in the intestinal loop. Four to eight testis lobes radiate around a small central ovary.

Perophora grows amid the calcareous alga *Porolithon japonica* at Waikīkī, on branched bryozoans in the Ala Moana yacht basin, on algae, solitary ascidians, and sponges at Kāne‘ohe Bay, on rocks near O‘ahu’s Hālona Blowhole, and in diverse habitats elsewhere along our shores. *Perophora annectens* is distributed outside Hawai‘i from southern California around the entire northern margin of the Pacific to Japan. *Perophora japonica* has been found in many places around Japan but not elsewhere. A third species closely resembling ours, *P. viridis* Verrill, 1871, occurs widely in warm waters in the Atlantic and has been reported from Tahiti (C. and F. Monniot 1987).

The following key distinguishes three species of the other perophorid genus, *Ecteinascidia*, and allies a fourth species, *E. garstangi*, with one of them. Only *E. imperfecta* is to be expected along our reefs and shores, but the other species present a notable taxonomic puzzle.

1 Pharynx with 11 rows of stigmata; stomach with 4–5 grooves; known only from Laysan Is. (but see remarks below) .... *Ecteinascidia diligens* Sluiter, 1900

Pharynx with 10–17 (usually 13–16) rows of stigmata; stomach smooth-walled 2

2(1) Internal longitudinal pharyngeal vessels mostly incompletely formed even in large, sexually mature zooids; lacks concentration of red pigment near base of oral siphon .... *Ecteinascidia imperfecta* Tokioka, 1950

Internal longitudinal vessels of pharynx largely or wholly complete except in very small or young zooids; with band(s) of red pigment near base of oral siphon; in Hawai‘i known only from Laysan Is. .... *Ecteinascidia rubricollis* Sluiter, 1885 and (see below) *E. garstangi* Sluiter, 1898

Sluiter (1900) reported three species of *Ecteinascidia* from Laysan Island. He described *E. diligens* as new and merely listed *E. rubricollis* and *E. garstangi*. Nishikawa (1986) argues that *E. diligens* is just a variant of *E. garstangi*. Kott

(1964, 1985) and Nishikawa (1986) suggest, as well, that *E. garstangi* and *E. rubricollis* may be synonymous. Thus, all three forms may be one widespread species, *E. rubricollis*. *Ecteinascidia garstangi* (in its current designation) has been reported from Mozambique and from Melanesia as well as from Laysan Island—an extraordinary distribution for an animal which, from its habitat, does not appear to be spread from port to port by ships. *Ecteinascidia rubricollis* has been found at least in Indonesia and northeastern Australia, and of course more widely if the synonymies noted above turn out to be justified.

The O'ahu *Ecteinascidia* resembles *E. rubricollis* in some ways, especially in the size and profile of zooids and their scanty musculature. But our species entirely lacks the ruddy (even vermilion) color of *E. rubricollis*, most of the internal longitudinal vessels are incomplete, and its smooth stomach lacks the four spiral grooves reported (Kott 1985) for *E. rubricollis*. Its attributes suggest, instead, *E. imperfecta* Tokioka, 1950, and it has gone by that name for some years. But we take note here of our species' confusing mix of traits with *E. rubricollis*. It would not surprise us to see its identification change once the genus is thoroughly reviewed and type specimens consulted.

The zooids of *Ecteinascidia imperfecta* (Fig. 8) are well separated from each other; they could be mistaken for solitary ascidians of the genus *Ascidia* if one missed the stolons connecting them (thus, we believe that *Ascidia minuta* Tokioka, 1950 is probably *E. imperfecta*). The zooids attach to the substrate posteroventrally. Large ones can reach a length of 1 cm. The thin tunic is colorless or faintly brownish, and the body is so clear that the zooid's bright orange or yellow stomach and proximal intestine show through. Sexual maturity in this species has not

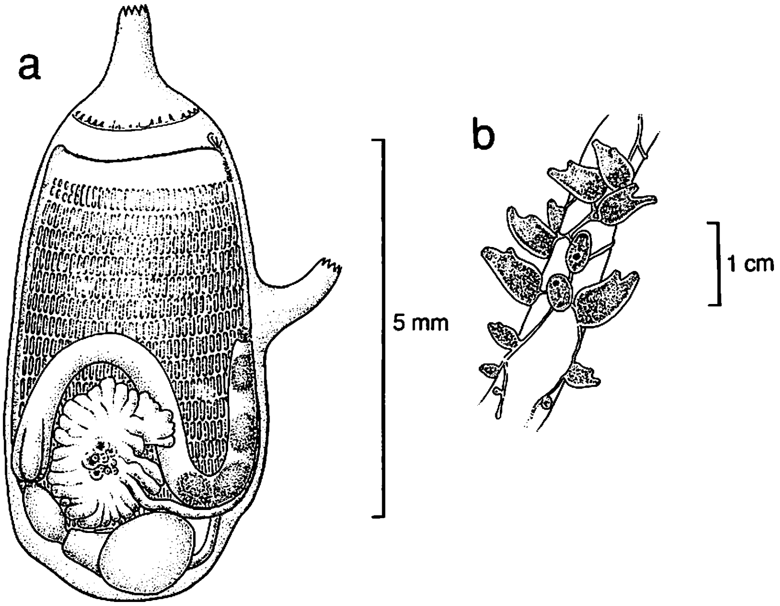

Figure 8.—*Ecteinascidia imperfecta:* a, zooid viewed from left side; b, young colony.

been described before. Hawaiian animals taken during the autumn often have a very large, fanlike testis with rather indistinct lobes surrounding a small ovary in the gut loop (Fig. 8a). This gonadal morphology is widespread in the genus, although the testis lobes are generally better defined in other species. Many autumn zooids have regressed gonads and brood very large embryos in the posterior right region of the atrium. This ascidian grows on worm tubes, dead coral, bryozoans, algae, and solitary ascidians in Kāne‘ohe Bay, Pearl Harbor, and the Ala Moana yacht basin. *Ecteinascidia imperfecta* was described by Tokioka (1950) from Palau; Kott (1985) reports it from northeastern Australia.

Family **Corellidae**

In the family Corellidae, all solitary ascidians, most of the gut loop lies on the *right* side of the pharynx, a very unusual condition in the class. The pharynx has large, pointed dorsal languets, and its internal longitudinal vessels lack papillae. In *Corella*, the only corellid genus known from Hawai‘i, the pharyngeal stigmata are arranged in spirals (Fig. 9b), so that a "row" of stigmata is a row of such elaborate complexes.

The Hawaiian corellid is *Corella minuta* Traustedt, 1882. Adults (Fig. 9a) are 1–2 cm high, with a clear, colorless tunic that reveals many of the body's internal organs. Some animals are prominently marked with white on the mantle and endostyle or with pink or greenish hues near the siphons. A few short, meager, longitudinal muscle bundles lie anteriorly and dorsally in the mantle. There are 18–24 transverse rows of spiral stigmata and 25–26 internal longitudinal vessels on each side of the pharynx.

Until recently, *Corella minuta* was considered a strictly Atlantic species, and *C. japonica* Herdman, 1880 its Pacific relative, so our Hawaiian corellid was called *Corella japonica*. We identify it as *C. minuta* for three reasons:

1. Musculature: *Corella japonica* has robust and extensive, anastomosing muscle bands in its left dorsal mantle. The left dorsal mantle musculature of *C. minuta* is sparse, and its bands do not anastomose. Although this distinction is one of degree, it is clear and consistent. Hawaiian *Corella* has a sparse left dorsal mantle musculature of well-separated strands (Fig. 9a).

2. Pharynx: Herdman (1882) says (and his Plate 26, Fig. 4 shows) that the transverse pharyngeal blood channels of *Corella japonica* are two to three times more numerous than its stigmatal rows, so this array must include one or more "parastigmatic" vessels per row in addition to the transverse blood sinuses that separate rows. *Corella minuta* (as shown by Van Name 1921, Fig. 72) has only a single transverse sinus per stigmatal row, the one that separates one row from the next. Hawaiian *Corella* has a transverse sinus between each stigmatal row and no extra, parastigmatic transverse vessels overriding the spirals (Fig. 9b).

3. Gonads: The ovary of *Corella minuta* from Guadeloupe (C. Monniot 1983a) and French Polynesia (C. and F. Monniot 1987) spreads to cover much of the gut loop, hiding the far less prominent testis. This is the situation in sexually mature Hawaiian *Corella*, too. C. Monniot (1983a:56) remarks that, in contrast, "in all other *Corella* species the ovary lies within the central part of the intestinal loop while the testis spreads onto the surface itself of the gut loop."

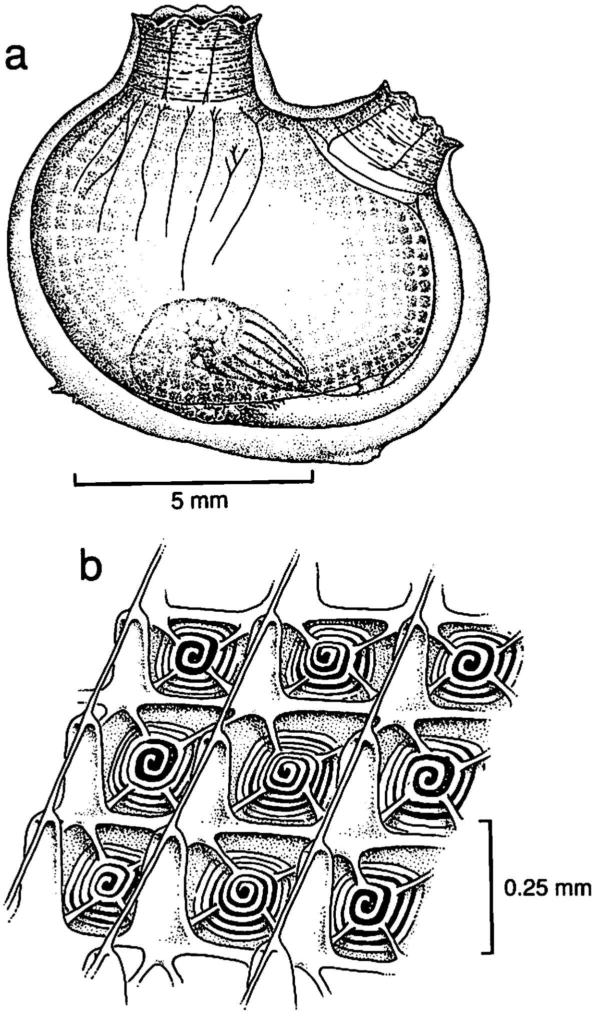

Figure 9.—*Corella minuta:* a, specimen viewed from left side, with tunic cut away; note gut loop on the *right* side of the pharynx and species' sparse mantle musculature near oral siphon; b, detail of pharyngeal wall, showing braced spiral complexes of stigmata, internal longitudinal vessels riding on struts, and transverse sinuses between rows of spirals.

Corella minuta has been found only sporadically in Hawai'i, on floating docks at Coconut Island in Kāne'ohe Bay. It is now known to occur widely in the central and western Pacific (C. and F. Monniot 1987). In the Atlantic, it lives in western Florida and around the eastern islands of the Caribbean Sea. *Corella japonica* appears to be confined to Japanese waters.

Family **Ascidiidae**

The large family Ascidiidae is represented in Hawai'i by four described solitary species and by two others, here called "A" and "B," of still uncertain identity. This family was one of Donald Abbott's specialties; its taxonomy poses some difficult problems, including possible species clusters and species with wide morphological variations. For these reasons, and because ascidiids are often prominent members of our fauna, we describe the Hawaiian ones at some length.

Ascidiids are usually ovoid and attach to the substrate by the left side. Both the mantle musculature and the tunic tend to develop most on the right side, away from the substrate. Most ascidiids have a translucent, firm tunic, often intricately vascularized, sometimes deeply pigmented. The contracted siphons may be hard to find in disturbed specimens, even in species whose relaxed siphons extend prominently. The siphons are well separated, in some species by more than half the length of the body. Siphonal apertures have lobed margins, which may be smooth or saw-toothed. Bright red or orange interlobal pigment spots and other siphonal markings decorate many otherwise drab species.

Numerous filiform tentacles mark the internal base of the oral siphon. Their number, the complexity of the ciliated slit, the length of the neural duct, and (ambiguously) the presence or lack of tiny pores between the neural duct and the atrium help distinguish some ascidiid taxa.

The pharynx has many rows of small, straight stigmata. Its internal longitudinal vessels combine with transverse sinuses to form an impressive grid. Papillae project into the pharynx from the intersections of these blood channels. The pharyngeal wall is finely fluted longitudinally. These plications become tiny pockets delimited by the internal pharyngeal blood channels and pierced by several stigmata per pocket (Fig. 10f). The pharynx's dorsal lamina, often doubled along the anterior third of its length, has transverse ribs on its left side that may extend beyond its free margin as projections that resemble tiny dorsal languets. The gut curves into a C- or S-shaped loop to the left of the pharynx. Gonads lie in the loop of the gut and when ripe may spread over its surface. Their gonoducts follow the intestine virtually to the anus, which opens near the atrial aperture.

1 Right-side tunic translucent but darkly pigmented, obscuring body within, especially around siphons 2

Right-side tunic gray or colorless, translucent to transparent, revealing body within, which may be brightly colored 3

2(1) Black or blue-black to gray, red-purple, or brown; attached to substrate by left posterior region; atrial siphon less than half body length from oral siphon and directed forward roughly parallel to oral siphon; neural complex far from dorsal tubercle (neural duct exceeds four times length of gland/ganglion complex itself); in quiet waters (Fig. 11) .... *Phallusia nigra* Savigny, 1816

Distinctly brown, brown-black, brown-gray, at least around siphons, which may be very dark; typically attached to substrate by bulk of left side; atrial siphon half to two-thirds body length from oral siphon, and directed away from oral siphon so that apertures are far apart; neural complex far from dorsal tubercle (neural duct exceeds four times length of neural gland/ganglion complex); subtidal in quiet or open waters .... *Ascidia melanostoma* Sluiter, 1885 [but see comments in species accounts]

3(1) Right-side musculature restricted to fringe of fibers arranged perpendicular to margin of body; neural complex close to dorsal tubercle; hindgut greatly swollen (Fig. 10) .... *Ascidia sydneiensis* Stimpson, 1855
Right-side musculature not restricted to fringe; hindgut not swollen 4
4(3) Right-side musculature forms weak overall mesh, with fringe like that of *A. sydneiensis;* oral siphon long, atrial siphon somewhat shorter; aperture borders have high triangular, elaborately toothed lobes; both siphons prominently ridged from base to tip; 100–150 long oral tentacles; neural gland/ganglion complex close to dorsal tubercle (Fig. 13a) .... *Ascidia* "A"
Right-side muscular meshwork lacks fringe of short fibers; lobes of aperture borders low, smooth-edged; siphons lack prominent longitudinal ridges; 30–70 oral tentacles 5
5(4) Tunic colorless; body bright orange (in sun) to gray-white and transparent (in deep shade); both siphons short; neural gland/ganglion complex close to dorsal tubercle; lobate or pinnate ovary connects by narrow neck to swollen, sausage-like oviduct; stomach and proximal intestine often heavily coated with vesicular tissue (Fig. 12) .... *Ascidia archaia* Sluiter, 1890
Body colorless to red in siphons, reddish elsewhere; oral region and siphon long, atrial siphon very short; aperture borders are low triangles; neural duct two to three times length of neural gland/ganglion complex; lobate ovary without swollen oviduct (Fig. 13b) .... *Ascidia* "B"

Ascidia sydneiensis is our commonest large ascidian in calm waters. Specimens may exceed 9 cm in length, although most are smaller. The tunic on the right side is only about 0.5 mm thick, more membranous than cartilaginous, finely wrinkled, and somewhat hairy or meagerly papillated (Fig. 10a, b). Translucent, it is clear enough to let the black, dark brown, orange-brown, or salmon red body show through. The siphons stand well apart from each other. One of this species' distinctive traits is the right side's prominent fringe of short, stout muscle fibers set perpendicular to the margin of the body (Fig. 10c), with the central region almost devoid of musculature.

A big *Ascidia sydneiensis* has more than 100 oral tentacles. The large dorsal tubercle's ciliated slit, U-shaped in small specimens, is highly convoluted in larger animals (Fig. 10e). The neural complex of gland and ganglion lies close to the dorsal tubercle. The pharynx has 40 or more internal longitudinal vessels per side. These and the dozens of transverse sinuses form a grid with 8–10 stigmata per mesh (Fig. 10f). The S-shaped gut loop is so deeply recurved that the intestine crowds or even overlaps the esophagus (Fig. 10d). The hindgut is grossly swollen, a bulbous intestinal chamber, usually packed with mud (Fig. 10d). The testis lies mostly on the intestine's outer curves and squeezes between the gut and the mantle; the ovary is more restricted to the intestine's inner curves and when ripe spills onto the intestine's atrial surface (Fig. 10d).

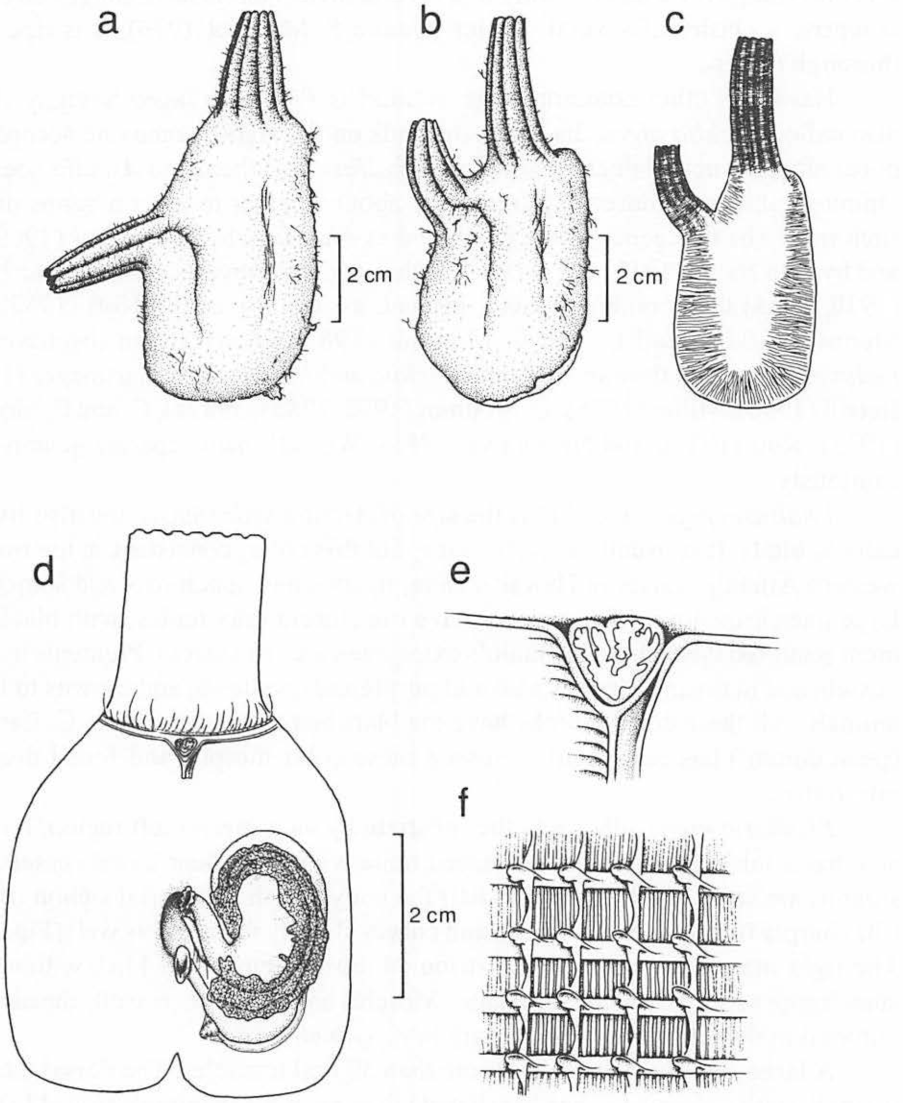

Figure 10.—*Ascidia sydneiensis:* a, b, specimens in their tunics viewed from right side; c, specimen in (b) removed from tunic to show fringe of musculature around otherwise unmuscled right mantle; d, specimen opened along mid-ventral line, pharynx removed, showing oral tentacles, dorsal tubercle, gut loop (note bulbous hindgut), and gonad; e, dorsal tubercle and convoluted ciliated slit of a large specimen; f, detail of pharyngeal wall, showing several stigmata per vascular mesh and papillae projecting from intersections of internal longitudinal vessels and transverse sinuses.

On O‘ahu, *Ascidia sydneiensis* exploits docks, floats, boat hulls, solid debris on mudflats, and other firm substrates in calm habitats such as Kāne‘ohe Bay, Pearl Harbor, and the Ke‘ehi boat harbor. The species occurs virtually worldwide in the tropics and in warm and temperate regions of the Southern Hemisphere.

Over this range it varies so greatly that several investigators have suggested it may comprise a cluster of several species (C. and F. Monniot 1976); it is ripe for a thorough review.

Hawai'i's other common large ascidiid is *Phallusia nigra* Savigny, 1816, also called *Ascidia nigra*. Its genus depends on the significance one accords the pores along its neural duct: in principle, *Phallusia* has them and *Ascidia* does not. Opinions shift with unsettling frequency about whether to erect a genus on one such trait. The two genera were combined as *Phallusia* by Hartmeyer (1909–11) and by Van Name (1912, 1921, 1924), following Hartmeyer's opinion. Van Name (1930, 1945) then combined them, instead, as *Ascidia*, as did Kott (1952b), C. Monniot (1970), and C. and F. Monniot (1987, tentatively in discussing *A. melanostoma*). But they are split into *Ascidia* and *Phallusia* by Hartmeyer (1924), Berrill (1950), Millar (1975), C. Monniot (1972, 1983a, 1987a), C. and F. Monniot (1972), Kott (1985), and Nishikawa (1986). We call them separate genera—but cautiously.

Phallusia nigra (Fig. 11) is the size of *Ascidia sydneiensis* and also lives in calm habitats. It is usually velvety black, but this color, consistent in the tropical western Atlantic, varies in Hawai'i. Here, many small specimens and sometimes large ones growing in shady places have translucent gray tunics, with black pigment scattered throughout the tunic's extensive vascular system. Pigments in these vessels and in the mantle may also add purple-reds, yellows, and browns to living animals. All these color morphs have the blanched eggs of *P. nigra*. C. Lambert (pers. comm.) has reciprocally crossed these color morphs and found them all interfertile.

Phallusia nigra adheres to the substrate by its posterior left region. Its right side has a fairly thick, smooth-surfaced tunic with prominent blood vessels. The siphons are separated by a third to half the body length. The atrial siphon usually tilts sharply toward the oral siphon and curves slightly toward it as well (Fig. 11a). The right mantle has an evenly distributed musculature (Fig. 11a), without any such fringe as *Ascidia sydneiensis* has. Muscles are apparent, as well, ahead of the gut loop in the left mantle (as they are in *A. sydneiensis*).

A large *Phallusia nigra* has more than 50 oral tentacles. The dorsal tubercle is small, with a simple U-shaped ciliated slit even in large animals (Fig. 11c). The neural duct extends from the dorsal tubercle halfway or more toward the base of the atrial siphon; thus, the complex of neural gland and cerebral ganglion lies far from the dorsal tubercle (Fig. 11b, c). The neural duct's atrial wall is pierced by many tiny openings (Fig. 11d) of unknown function and, as we noted above, of debated taxonomic value. The pharynx carries more than 40 internal longitudinal vessels per side, which cross dozens of transverse sinuses to form a grid with six to eight stigmata per mesh. Curved papillae project from the intersections of these pharyngeal blood channels. The S-shaped gut loop is large and tightly recurved. The ripe testis spreads extensively over the more anterior surface of the hindgut; the ovary tends to ramify more posteriorly, within the proximal loop of the hindgut.

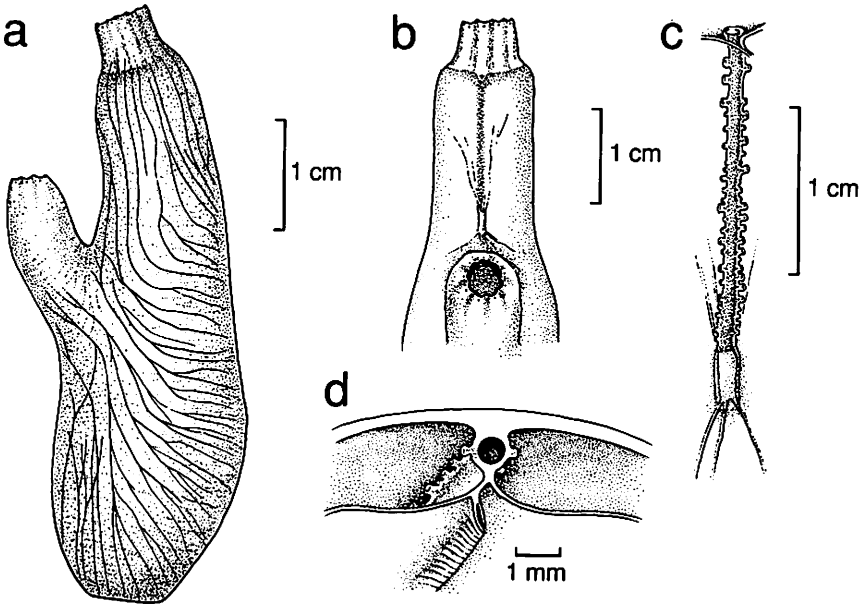

Figure 11.—*Phallusia nigra:* a, specimen viewed from right side, removed from tunic to reveal right-side musculature (cf. *Ascidia sydneiensis*, Fig. 10c); b, dorsal view of intersiphonal region, oral siphon at top, showing long duct from dorsal tubercle to neural gland near base of atrial siphon; c, ventral view of neural duct and neural gland excised from body; ciliated slit of dorsal tubercle is at top; long neural duct shows many pores along its sides; d, schematic sectional view of neural duct, showing pores along its length opening into atrium above pharynx.

On O'ahu, *Phallusia nigra* lives on rocks and dead coral on barely subtidal mudflats in Kāne'ohe Bay and on floats and pilings there and in Pearl Harbor and in the Ke'ehi boat harbor. In all the rest of the Pacific, it has been found only in Micronesia. It is a widespread species in the western Atlantic tropics, and it has also been recorded (in fact, it was described originally) from the Red Sea and the northwest coast of the Indian Ocean.

Long (1974) reports two instances of *Ascidia melanostoma* settling on offshore test panels 15 m deep off the southern coast of O'ahu and 9 m deep in Pearl Harbor. This dark species is usually only 2 or 3 cm long. Its siphons are brownish black, but the rest of the tunic is more often a translucent gray or grayish brown with scattered, deep brown pigment. The body is rather elongated, accentuating the length of the oral siphon; the short atrial siphon usually arises more than halfway down the body's dorsal midline. In contrast to the almost parallel siphons of *Phallusia nigra*, those of *A. melanostoma* point away from each other.

The *Ascidia melanostoma* neural duct extends halfway to the atrial siphon; one specimen from French Polynesia has accessory pores opening from this duct into the atrium, raising anew doubts about the worth of these pores as the trait by which to separate *Phallusia* from *Ascidia* (C. and F. Monniot 1987). Large adults have 60–80 oral tentacles. The pharynx has 30–40 internal longitudinal vessels per side, many transverse sinuses, and as few as three to five stigmata (in French Polynesia) or as many as eight (in Melanesia) per vascular mesh. The gut loop forms a loose S-shape. The ripe ovary covers the intestine's atrial side, even hiding the testis, which is more tightly confined to the proximal intestinal loop. Beyond Hawai'i, *A. melanostoma* has been found in the Philippines, Indonesia, Melanesia, and French Polynesia.

We do not doubt the validity of *Ascidia melanostoma* as a species, but its presence in Hawai'i must be questioned now because of recent work by C. and G. Lambert (pers. comm.). While examining fertilization in *Phallusia nigra*, they found a disconcertingly wide variation in this species' external traits. Even specimens that seemed to key out as *A. melanostoma*—that had, for example, the associated color, disposition of siphons, and manner of attachment to the substrate—interbred with indubitable *P. nigra*. So although Long's records and *A. melanostoma*'s characters should be considered when one finds dark red-brown Hawaiian ascidiids of moderate size, such specimens may be variants of *P. nigra*.

Some years ago, small, clear specimens of *Ascidia archaia* grew on the walls of aquaria in the old Honolulu Aquarium. Recently it has been found growing on other solitary ascidians and on oysters in the seawater run-off trenches at the Kewalo Marine Laboratory, Honolulu. *Ascidia archaia* (Fig. 12a–c) from these artificial sites reaches 2 cm, smaller than this species becomes in natural habitats. Its fairly smooth, firm tunic is transparent and colorless. At the Kewalo laboratory, animals growing in bright sunlight have a distinctly orange mantle and pharynx, whereas others growing nearby in deep shade are much paler. The animal attaches by its left side; shell debris may adhere to its right side. The siphons are separated by about a third of the body length (Fig. 12b); the atrial siphon points sharply away from the oral siphon.

The dorsal tubercle of *Ascidia archaia* has a simple U- or C-shaped ciliated slit (Fig. 12c); a short neural duct connects the dorsal tubercle to the neural complex nearby. About 30 large oral tentacles and somewhat fewer small ones encircle the entrance to the pharynx. The pharynx carries 25–35 internal longitudinal vessels per side, about 30 transverse sinuses, and, in Hawai'i, two to four stigmata per mesh in the resultant grid. Papillae project from the junctions of these pharyngeal blood channels, alternating with smaller intermediate papillae that arise from the junctions of internal longitudinal vessels with parastigmatic blood vessels. The gut loop forms a fairly open S-shape. An odd, vesicular tissue usually envelops the stomach and proximal intestine. The ovary, lying within the curve of the gut loop, may form a tight lobular mass (Fig. 12a), or it may be long and extend many side branches onto the intestine (Fig. 12c). It connects by a narrow neck to a stout, sausage-shaped oviduct. The testis lobes are scattered over the proximal intestine, and the usually inconspicuous sperm duct is prominent when full.

Records for this species, and the decades separating them, suggest that it lives subtidally off Honolulu. *Ascidia archaia* ranges widely over the tropics. By its current name or as *A. rhabdophora* Sluiter, 1904, it has been reported from

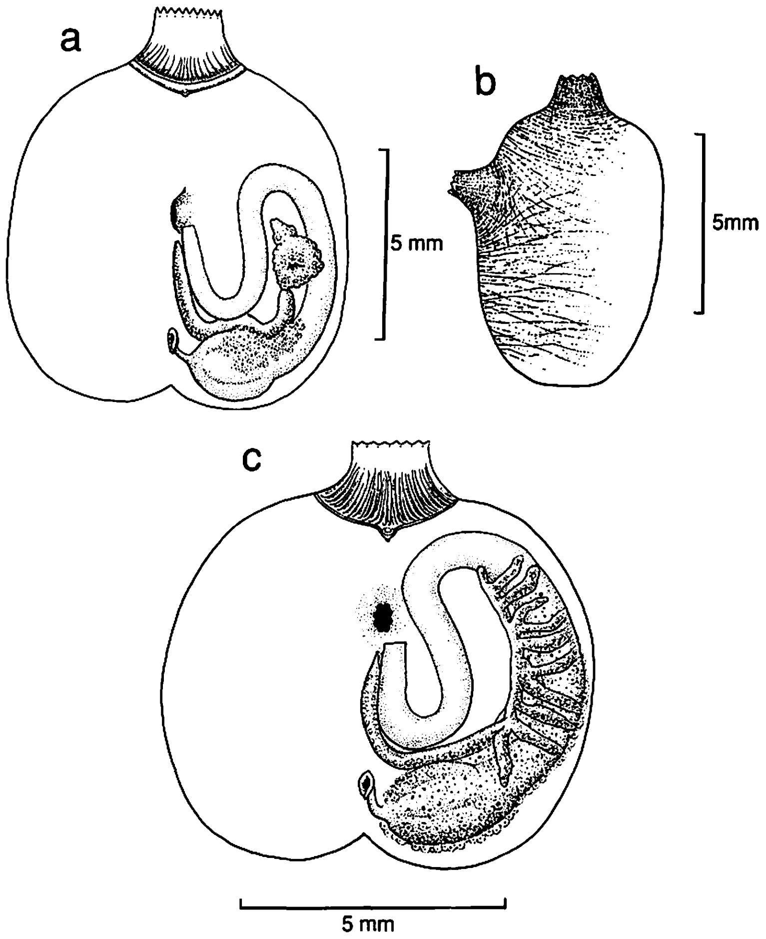

Figure 12.—*Ascidia archaia:* a, small specimen opened along mid-ventral line, pharynx removed; note small oral tentacles, dorsal tubercle, compact ovary in gut loop, inflated oviduct, and some vesicular tissue on stomach and adjacent intestine; b, unopened specimen, divested of tunic, viewed from right side; c, larger specimen opened as in (a), with larger oral tentacles, pinnately ramified ripe ovary, and extensive vesicular tissue on stomach, proximal intestine, and ovary.

Japan, the Philippines, Australia, Indonesia, Micronesia, Melanesia, and French Polynesia. And, by its junior synonym *A. corelloides* (Van Name, 1924), it has been recorded from the Atlantic in the West Indies (C. Monniot 1983a).

Two enigmatic forms of *Ascidia* occur on O‘ahu. We call them simply *Ascidia* “A” and *Ascidia* “B” (Fig. 13a, b). Both are fairly common on floats and subtidal reefs in protected habitats and are found regularly in Kāne‘ohe Bay and Pearl Harbor. Neither form fits well into a currently described *Ascidia* species, but each can be recognized in Hawai‘i for what it is (either “A” or “B”). The following comparison is based on several collections from Kāne‘ohe Bay. We include in this comparison several traits that we have not dealt with in describing other ascidiids, because this report is the first to introduce these forms.

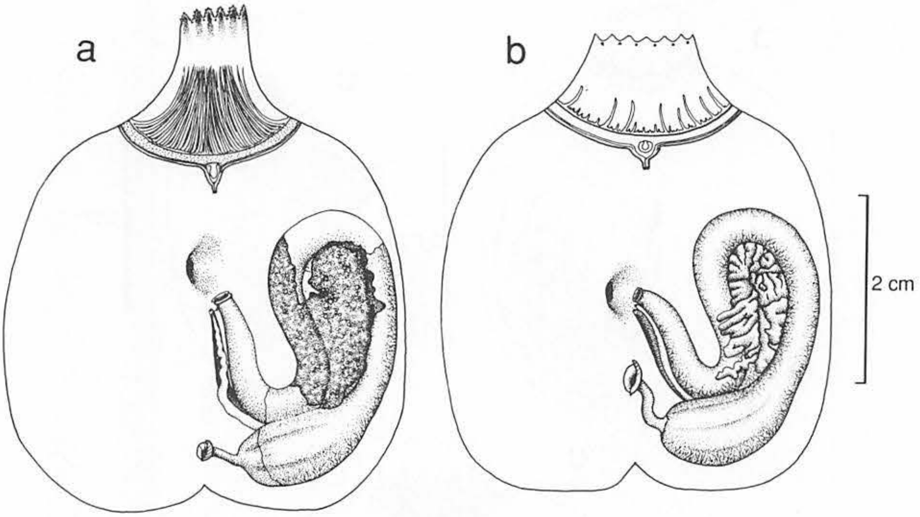

Figure 13.—a, *Ascidia* "A" opened along mid-ventral line, pharynx removed to show various structures referred to in the text; b, *Ascidia* "B" similarly prepared.

	Ascidia "A"	*Ascidia* "B"
length of large relaxed body in tunic, base to tip of oral siphon	5 cm	5 cm
attachment region	posterior left	posterior left
tunic	fairly transparent; fairly smooth, or with criss-crossed wrinkles, sparse hairs, or tiny papillae	fairly transparent; smooth or minutely wrinkled; somewhat gelatinous quality
outer surface of siphons	ridged from base to marginal lobes	smooth
length of siphons	oral about twice length of atrial	oral very long, atrial very short
position of atrial siphon	third to halfway down body	halfway down body
shape of lobes around apertures	high triangular, elaborately toothed	low triangular with smooth borders
number of lobes around apertures	oral, 6–7 atrial, 5–6	oral, 7–10 atrial, 6
red spots between aperture-margin lobes?	yes	yes
mantle pigmentation	faint salmon siphons with red or purplish brown borders; orange-red pigment vaguely dispersed in mantle	colorless to red in siphons, reddish mantle and pharynx

	Ascidia "A"	*Ascidia* "B"
right-side mantle's musculature	weak meshwork augmented by marginal fringe like that of *A. sydneiensis*	uniform meshwork on whole right side
oral tentacles	100–150	30–70
slit of dorsal tubercle	simple U-shape or horseshoe-shaped	U-shaped or somewhat convoluted
length of neural duct	short (less than length of neural gland)	long (two to three times length of neural gland)
papillated surface between oral tentacles and peripharyngeal band?	usually	only occasionally
accessory tentacles on upper ridge of peripharyngeal band?	yes	no
internal longitudinal vessels (per side)	30–40	25–40
intermediate papillae on internal longitudinal vessels?	rare and tiny	no
stigmata per pharyngeal mesh	8–10	about 6
gut loop	tightly recurved S-shaped	tightly recurved S-shaped
hindgut enlarged into an intestinal chamber?	no	no
anal margin	smooth, bilobed	smooth, bilobed
ovary	compact, smooth, non-lobular masses in curves of mid- and hind-intestine	compact, distinctly lobular masses in curves of mid-intestine
testis	spread widely over gut loop	spread widely over gut loop

The fringe of right-side *Ascidia* "A" musculature resembles that of *A. sydneiensis*, but in *Ascidia* "A" the muscles spread, as well, weakly through the rest of the right mantle. *Ascidia* "A" differs from *A. sydneiensis* in other ways, too: in its lack of a swollen hindgut, its simple U-shaped ciliated slit, and the much more elaborately toothed borders of its siphonal apertures. But *Ascidia* "A" traits have been encountered in animals which, all else considered, appear to be variants of *A. sydneiensis*. *Ascidia sydneiensis* may even lack an inflated hindgut—for example, Tokioka's *A. sydneiensis* from Sagami Bay, Japan (1953), Millar's from South Africa (1964), or Vasseur's from Mauritius (1967). And the fringe of right-side mantle musculature blends into a general mesh in some specimens from Hong Kong (Kott and Goodbody 1982). Faced with this variety, one may conclude that *Ascidia* "A," despite its many differences from "typical" *A. sydneiensis*, is still another variant within that species. In that case, why are there no intermediates in

Hawai'i between "typical" *A. sydneiensis* and *Ascidia* "A"? Vasseur (1967) claims that three distinct forms of *A. sydneiensis* occur on Mauritius. Are these intraspecific morphs, somehow maintained distinctively, or are they three real species? Variations and puzzles like these persuade us to leave Hawai'i's *Ascidia* "A" as a taxon of uncertain status.

Ascidia "B" presents a less complicated picture. Its neural duct is fairly long, two to three times the length of the neural gland. Of described Pacific ascidiids with fairly long neural ducts, *A. gemmata* Sluiter, 1895, widespread in the tropical western Pacific, resembles *Ascidia* "B" in many ways. It differs from our ascidian in having a densely papillated surface at the internal base of the oral siphon, usually bluish or purplish eggs instead of our animal's green ones, and other details that one can attribute to geographic variations. We believe that *Ascidia* "B" is close to if not actually *A. gemmata;* but, without examining the type specimen of that species, we must stop short of certain identification.

Order PLEUROGONA, Suborder STOLIDOBRANCHIA

Family **Styelidae**

These solitary or colonial pleurogonid ascidians have filiform oral tentacles, a smooth-edged dorsal lamina, and rows of straight pharyngeal stigmata. The body is compact and ovoid. The siphonal apertures are four-lobed or even-bordered. Pharyngeal folds—four or fewer per side—are present in all but the smallest solitary styelids and the zooids of some colonial species. These folds and the spaces between them carry internal longitudinal vessels, the numbers and groupings of which help identify many taxa.

Solitary styelids have stout bodies and usually reflect this in their outward appearance. Some species, but none yet recorded from Hawai'i, have a pedunculate tunic that raises the body into the water on a narrow stalk. The harbor-dwelling *Styela clava* Herdman, 1881 is stalked. A fouling animal, it is being spread by ships into ports around the temperate world and may turn up here, too. Colonial styelids encrust substrates. The colonies vary in the size of the usually rather small zooids, the distance budded zooids move away from their parents, the thickness of the common tunic relative to the size of the zooids, and colonial patterns imposed by the space available to spread at all.

In styelids as in the other stolidobranchs (pyurids and molgulids), most of the gut loop lies in the body wall to the left of the pharynx. The styelid stomach has many longitudinal grooves, and in some species it has a little caecum. The gonads, usually right-left paired or multiple, lie on or in the outer wall of the large atrium. In many styelids, the atrium wall produces saccular outpouchings called endocarps (Fig. 17a, b), which may be mistaken for gonads. Despite their prominence, their function is unknown.

For convenience, we divide the family Styelidae into three subfamilies—the colonial Botryllinae and Polyzoinae and the solitary Styelinae. But whereas Botryllinae is probably a natural, monophyletic taxon, Polyzoinae is most likely polyphyletic—an artificial combination from several styelid sources—and Styelinae is likely commensurately incomplete (paraphyletic).

In botryllinid colonies the tiny zooids, deeply buried in a soft tunic, form systems around common cloacal cavities that open to the outside by common cloacal apertures. A botryllinid zooid's pharyngeal wall is not folded: it has three internal longitudinal vessels on each side. The group has long been felt to comprise two old and well-known genera, *Botryllus* Gaertner, 1774 and *Botrylloides* Milne Edwards, 1841. A standard rule of thumb for telling them apart in the field has been that the zooids of *Botryllus* colonies arrange themselves into stellate, oval or round systems, each with a central common cloacal aperture (Fig. 14a), whereas *Botrylloides* zooids form linear, often meandering, ladder-like systems that contain several common cloacal apertures (Fig. 14b). In fact, these colony-level patterns of systems vary inconsistently among species that would be assigned by their zooids' internal traits—that is, by criteria normally used to classify ascidians—to one genus or the other. C. and F. Monniot (1987), reviewing the shifting and persistently unsatisfactory zooid-level criteria that have been used to distinguish the genera *Botryllus* and *Botrylloides*, argue that the genera are in fact synonymous, and that all botryllinid species therefore belong to the senior genus, *Botryllus*. We agree with the Monniots' assessment. Since we hesitate in a venue such as this simply to discard a long-standing, familiar generic name like *Botrylloides*, we instead use the formula "*Botryllus* (=*Botrylloides*)" when referring to the only Hawaiian botryllinids that have been found so far, which are of one or more species with ladder-like systems.

In contrast to the entirely "compound" botryllinid styelids, colonies of the subfamily Polyzoinae may be "compound" ones whose zooids are packed into a

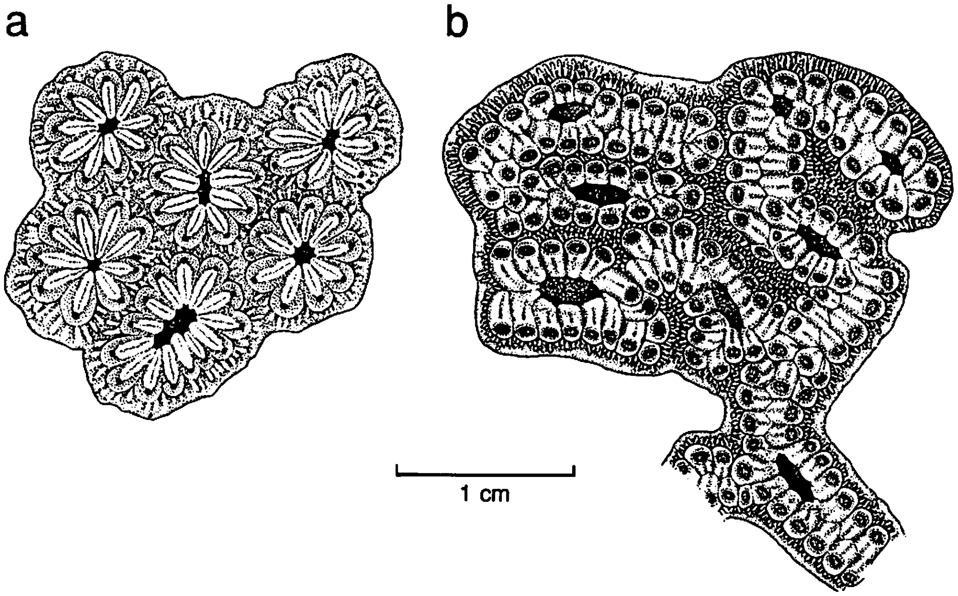

Figure 14.—a, *Botryllus* sp. with stellate systems of zooids; b, *Botryllus* (=*Botrylloides*) sp. with meandering linear systems of zooids.

relatively thick tunic or "social" ones in which well-separated zooids are joined only by a thin, vascularized sheet of common tunic. Clearly, the adjectives "compound" and "social" are of descriptive but not taxonomic value. Polyzoinid zooids are not arranged in systems; their atrial apertures open independently to the outside. The pharyngeal wall may be smooth or folded and always bears four or more internal longitudinal vessels on each side. The genera *Symplegma*, *Eusynstyela*, and *Polyandrocarpa* occur in Hawai'i.

The subfamily Styelinae comprises solitary styelids. In most species the pharyngeal wall has four folds per side, but in a few very small (and non-Hawaiian) styelinids these folds are represented merely by groupings of blood vessels. The styelinid genera known so far from Hawai'i are *Styela*, *Cnemidocarpa*, and *Polycarpa*.

The Hawaiian species of all three styelid subfamilies are treated together in the following key.

1	Colonial ascidians	2
	Simple ascidians	6
2(1)	Zooids in meandering, elongate systems, often appearing as double rows of zooids; atrial apertures open into common cloacal cavities in tunic; pharynx with 3 internal longitudinal vessels per side; gonads and brooded embryos protrude from sides of zooids; living colonies often brightly colored (Fig. 14b)	*Botryllus* (=*Botrylloides*) sp./spp.
	Zooids do not form systems; all atrial apertures open directly to outside; pharynx with 4 or more internal longitudinal vessels per side; living colonies variously colored	3
3(2)	Tiny, pale green zooids buried in clear tunic; pharyngeal wall lacks folds; 4 internal longitudinal vessels on each side (Fig. 15)	*Symplegma oceania* Tokioka, 1961
	Zooids not tiny, not deeply buried in common tunic; tunic tough but rather thin and often translucent in life, though it may be coated with sediment; pharyngeal wall with 2 or 4 folds and about 30 internal longitudinal vessels per side	4
4(3)	Zooids taller than broad; siphons prominent, close together; all 4 pharyngeal folds well developed; longitudinal stigmata	5
	Zooids shorter than broad; siphons low, rounded, well separated; 2 of 4 folds on each side of pharynx represented only by groups of close-set internal longitudinal vessels; transverse stigmata	*Eusynstyela transversalis* Tokioka, 1963
5(4)	Tunic leathery, often with epibionts; tangled colony of very densely packed, distorted zooids tightly adhering to each other by much of their tunics; siphons thick, long, with 4 terminal lobes and 4 conspicuous purplish longitudinal stripes; about 20 stubby hermaphroditic gonads on right atrium wall, fewer on left wall (Fig. 16)	*Polyandrocarpa sagamiensis* Tokioka, 1953

Tunic tough, smooth but sand-encrusted, not overgrown by epibionts; bouquet-like colony of densely packed club-shaped zooids connected basally by common tunic; siphons raised but rather delicate; cleaned siphons may show 8 dark stripes (2 per siphonal lobe); 5–10 stubby hermaphroditic gonads on right atrium wall, 3–5 on left wall .... *Polyandrocarpa zorritensis* Van Name, 1931

6(1) Gonads elongate, each consisting of a tubular ovary and few to many testis lobes clearly separate from the ovary but arranged around its sides and posterior end 7

Gonads long and tubular or short and ovoid; each gonad consists of ovary and testis lobes encased tightly together inside a membrane 8

7(6) Tunic surface mostly hairy; one elongate ovary and its associated testis lobes on each side of body; known only from deep water (600 m) off O‘ahu and Maui .... *Styela izuana hawaiiensis* Tokioka, 1967

Gray or tan and smooth to dark brown and deeply wrinkled; tunic surface not hairy; siphons striped purple and white; 2 elongate ovaries and their associated testis lobes on each side of body; in bays and harbors (Fig. 19) .... *Styela canopus* Savigny, 1816 [formerly *Styela partita* (Stimpson, 1852)]

8(6) (note: three choices)

Dark, deeply wrinkled tunic, warts around bases of siphons; siphons prominently striped purple-brown and white; elongate and tubular gonads at least three times longer than wide; usually 3 gonads on right, 2 on left (Fig. 18) .... *Cnemidocarpa areolata* (Heller, 1878)

Bright red body; colorless, sand-encrusted tunic; gonads less elongate than *C. areolata* but still sausage-like; 7 gonads on right side, 4 on left in sole specimen examined; a few specimens taken once offshore near Honolulu Harbor .... *Cnemidocarpa* sp.

Pale brown; stubby to fairly elongate gonads very numerous on and in atrium wall on both sides of body; many prominent endocarps, often one overlying the stomach (Fig. 17) .... *Polycarpa aurita* (Sluiter, 1890)

One or more *Botryllus* (=*Botrylloides*) species with linear systems are common in our harbors and occasional on reefs. These colonies are flat and encrusting, often several centimeters across. The zooids, only 2–3 mm long, are arranged in elongate double rows flanking common cloacal canals that meander and branch (Fig. 14b). The inflated ends (ampullae) of blood vessels in the tunic are prominent, especially along the growing margins of a colony. Living colonies are very colorful—red, orange, purplish, light and dark green, or shades of brown, often with vividly contrasting bands of gold or white between the zooids' siphons. Preserved animals turn brownish or dark purplish brown. Hawaiian botryllinids have not been studied or even tentatively identified. Since many species show color polymorphism, their variety surely exceeds their taxonomic diversity here. Surprisingly, species of *Botryllus* with stellate systems, diverse and widespread in quiet waters around most of the world, have not been found in Hawai‘i.

Symplegma oceania forms inconspicuous, thinly encrusting colonies (Fig. 15a). The zooids, 2–3 mm long, lie ventral side down in a transparent sheet of tunic and project both their oral and atrial siphons directly to the colony surface. Living colonies are almost white or pale green-yellow, often with tan, reddish, or yellowish bands around the zooids' apertures. In *Symplegma* as in botryllinids, the tunic contains prominent blood vessels that end in swollen ampullae, most noticeably at the growing margins of the colony (Fig. 15a). Zooids have four internal longitudinal pharyngeal vessels and 9–11 transverse rows of stigmata per side (Fig. 15b). The stomach has a prominent caecum and tubular connections with the proximal part of the intestine (Fig. 15c). Colonies have been found at Kāne‘ohe Bay, Pearl Harbor, and Honolulu Harbor, growing on such substrates as solitary tunicates, worm tubes, boat hulls, floats, and dock pilings. Elsewhere, *S. oceania* occurs widely in the temperate and tropical Indo-Pacific; it has recently become established on floats in San Diego Bay, California (G. Lambert, pers. comm.). The closely related *S. viride* Herdman, 1886 lives in the tropical western Atlantic.

Two polyzoinid genera that closely resemble each other, *Eusynstyela* and *Polyandrocarpa*, occur in Hawai‘i. These genera differ most consistently in the structure of their gonads. In *Eusynstyela* the multiple gonads each have only two (occasionally only one) testis lobes; in *Polyandrocarpa* each gonad contains several to many testis lobes.

Eusynstyela transversalis forms encrusting colonies whose zooids grow to 4 mm long and lie well separated from each other. The thin tunic may be partly obscured by attached materials. The zooids' nearly globular gonads, up to eight per side, each have two testis lobes. The only colonies of this species found so far in Hawai‘i were growing on *Ascidia sydneiensis* that was dredged from 50–80 m off Lahaina, Maui, during the 1902 *Albatross* Hawaiian explorations; they were first detected decades later by Tokioka when he was examining the *Albatross* collections. The species name *transversalis* refers to the extraordinary transverse (instead of the usual longitudinal) orientation of the pharyngeal stigmata, which nonetheless lie in regular transverse rows.

Colonies of the widespread *Polyandrocarpa sagamiensis* have zooids that reach 2 cm across and 3 cm high, packed together so tightly that the tunics of individual zooids partly fuse (Fig. 16b). Living colonies—tan, brown, orange-brown, or yellowish gray—form encrusting masses 10 cm or more across; when relaxed they are veritable thickets of projecting siphons. Each four-lobed aperture is marked with four prominent purple-brown patches; conspicuous tubercles often decorate the inner surface of the oral siphon. The body wall is very thin and fragile. There are 20–30 oral tentacles; the dorsal tubercle's ciliated slit is transverse and straight or has a shallow U-shape. The pharynx has four folds per side, each of which carries about a dozen internal longitudinal vessels; the interfold spaces carry five to seven vessels apiece (Fig. 16a). The stomach has a dozen external grooves and lacks a pyloric caecum. In a single zooid the gonads, in which ovaries and testis lobes are tightly juxtaposed, range in shape from globes to elongate sacs;

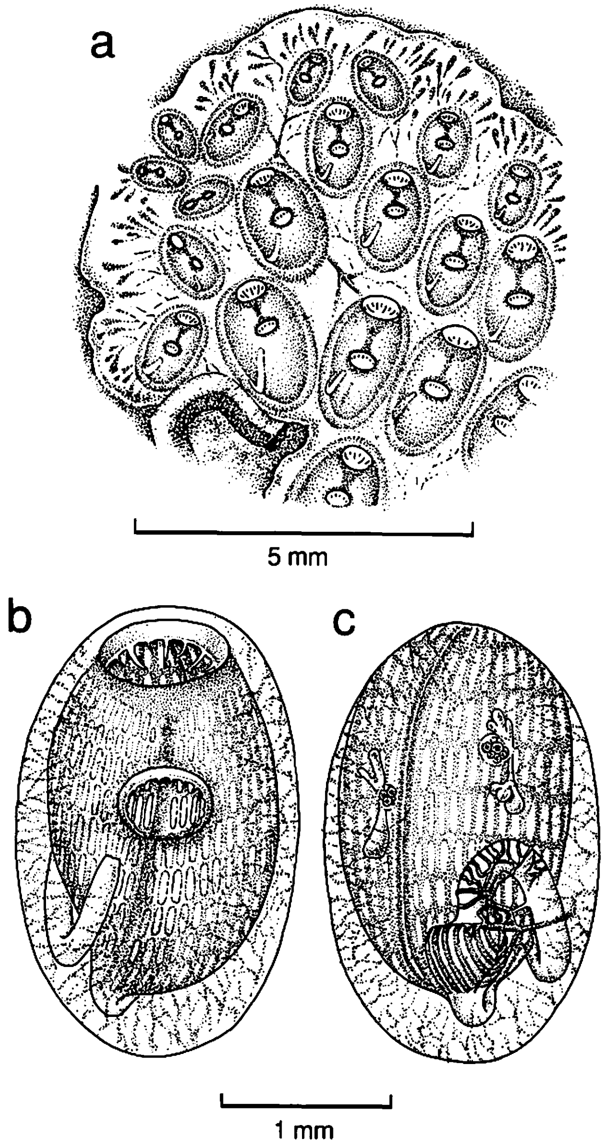

Figure 15.—*Symplegma oceania:* a, portion of a colony; b, dorsal view of individual zooid with mantle intact; c, ventral view of same intact zooid, showing elaborate gland connecting stomach and proximal intestine, and pleurogonid gonads.

they number 20 or even 30 on the right side, somewhat fewer on the left (Fig. 16c). The atrium wall lacks endocarps.

Polyandrocarpa sagamiensis is common on floating docks in Kāne'ohe Bay, and what is probably the same species occurs in similar habitats in Pearl Harbor. It was first found in Japan (Sagami Bay, hence its name). Kott (1964, 1985), reporting it from Australia, synonymizes *P. rollandi* Tokioka, 1961 from New Caledonia with this species; but C. Monniot (1987b) argues that New Caledonian *P. rollandi*

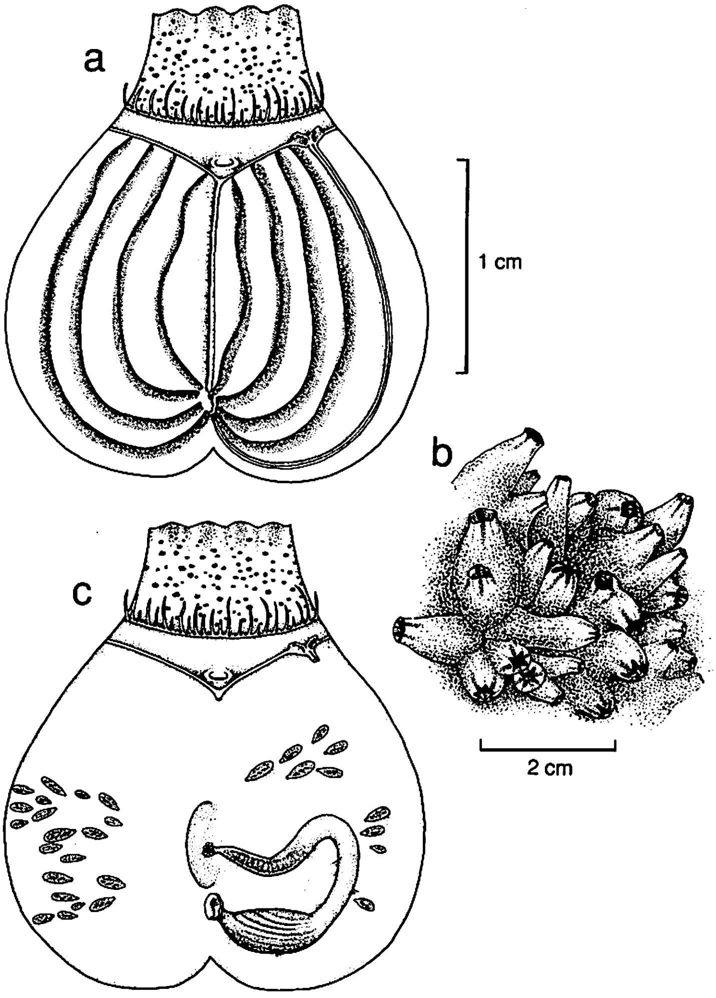

Figure 16.—*Polyandrocarpa sagamiensis:* a, zooid opened along mid-ventral line, pharynx left intact (endostyle at far right of drawing); note papillose interior of oral siphon, oral tentacles, dorsal tubercle, and pharyngeal folds; b, portion of a colony; c, zooid with pharynx removed to show gut loop and many small hermaphroditic gonads.

is a good species in its own right. He notes that Kott's 1985 drawing of an oddly sand-encrusted colony of *P. sagamiensis* looks quite unlike the much cleaner *P. rollandi. Polyandrocarpa sagamiensis* in Hawai'i also usually has a fairly unsilted tunic, though it is often heavily overgrown with epibionts. What appears to be *P. sagamiensis* lives in Mauritius (as *P. anguinea* in Vasseur 1967).

Polyandrocarpa zorritensis has been found recently in Hawai'i by H. Koyama and identified by T. Nishikawa (G. Lambert, pers. comm.). This species differs in appearance from *P. sagamiensis* in the following ways: its colonies are dense but less massive because their club-shaped zooids adhere to each other only near their bases and not well up along the sides of their tunics; its zooids (about 2 cm high) have tough but thin tunics that usually are silt-encrusted and without epibionts; and their rather short, four-lobed siphons may have eight dark stripes instead of four. Internally, its stubby little gonads are only half as numerous as those of *P. sagamiensis*. *Polyandrocarpa zorritensis* has been found growing in midwinter on an ecological survey raft in Pearl Harbor, on floats at the Coconut Island Marine Laboratory in Kāne'ohe Bay, and in the Kewalo Bay marina, all on O'ahu. Beyond Hawai'i, the species has a bizarre distribution: Peru, southern California, and the Mediterranean. Perhaps it has joined the ascidian species that are traveling in the hulls of ships from port to port around the world.

The zooids of *Polyandrocarpa* and bodies of *Polycarpa* are so alike that they cannot be assigned to one genus or the other by their anatomy. Rather, generic assignment depends on their growth habit: *Polyandrocarpa* species bud to form colonies, *Polycarpa* species do not. By custom, species of ambiguous growth habits are assigned to *Polycarpa;* some species of *Polycarpa* live in tightly packed aggregations that strongly resemble colonies. In fact, in several *Polycarpa* species the body undergoes strange bouts of evisceration, and in a few of them (e.g., *P. nigricans* Heller, 1878; *P. papillata* Sluiter, 1885) this event is followed by single or multiple regeneration of new bodies to produce one or several replacements within the old tunic—in effect, colonies (C. Monniot 1987b). One can surmise at least that the genera *Polyandrocarpa* and *Polycarpa* are very closely related. Putting them in separate subfamilies (colonial Polyzoinae, solitary Styelinae) is surely misleading and even casts doubt on the systematic value of the subfamilies themselves.

Pale brown individuals of what P. Kott (pers. comm.) has kindly identified for us as *Polycarpa aurita* live on floats in Kāne'ohe Bay, subtidally in the Honolulu Harbor channel, and in the seawater run-off system at Honolulu's Kewalo Marine Laboratory. Specimens rarely exceed 3 or 4 cm. Divested of their rough, somewhat cartilaginous tunics, fresh specimens from the Kewalo laboratory show surprising color: a gold patch on each side of the intersiphonal mid-dorsal line, contrasting with a purplish mantle. Both the oral and atrial siphons have a very papillose lining. About 50 oral tentacles ring the entrance to the pharynx. The very large, cushion-like dorsal tubercle has an incurled C-shaped or W-shaped ciliated slit (Fig. 17e, f). On each side of the pharynx, the four folds carry at least a dozen internal longitudinal vessels apiece, with a few vessels in the interfold spaces. The stomach has nearly 20 internal folds, most of which are scarcely visible externally as grooves. A large pyloric caecum projects from the stomach, and a prominent tube links the stomach with the proximal intestine (Fig. 17g, i). In many large specimens, dozens of gonads two to three times as long as wide are easily discernible in the atrium wall (Fig. 17a, c). In small animals the gonads are stubby (Figs. 17b, d), not elongate as others have reported of this species. The atrium wall has many hernia-like endocarps among the gonads (Fig. 17a, c); two or three large ones beside or inside the gut loop tend to overgrow the stomach (Fig. 17h).

We are confident of Kott's identification of this *Polycarpa* as *P. aurita;* she is thoroughly familiar with that species. But confusing differences exist between

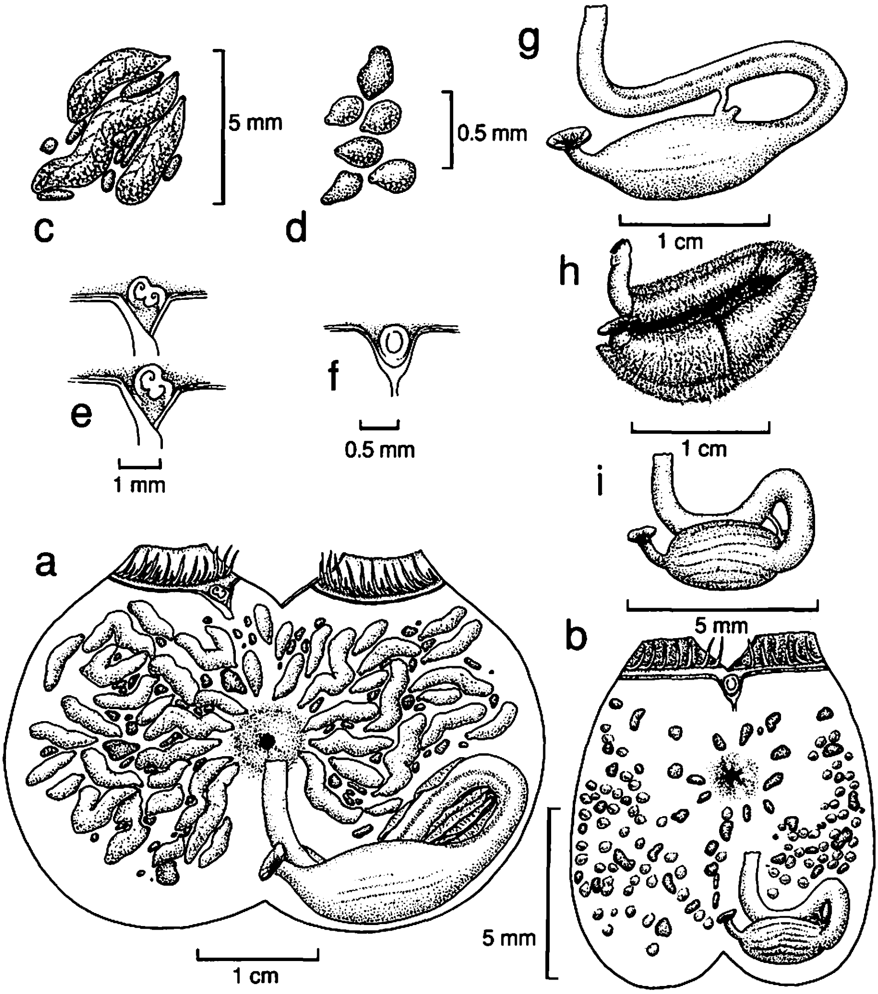

Figure 17.—*Polycarpa aurita:* a, large specimen opened along mid-ventral line, pharynx removed, showing oral tentacles, dorsal tubercle, gut loop, gonads, and endocarps; very large endocarps in curve of gut loop have been cut away to show gut loop more fully, but flaplike ones alongside gut are visible; b, smaller specimen similarly prepared; c, various gonads from (a); d, various gonads from (b); e, dorsal tubercles and ciliated slits from large specimens; f, dorsal tubercle and ciliated slit from small specimen; g, h, i, gut loops from three specimens; caecum and stomach–intestine connection are clear in (g) and (i), from which large endocarps in curve of gut loop have been removed; those endocarps, left in place, obscure gut loop in (h).

the Hawaiian specimens and those described from elsewhere. Most troubling, Hawaiian animals have far fewer internal longitudinal vessels on their pharyngeal folds than any descriptions of *P. aurita* indicate. But accounts suggest that this is a highly variable species, for they disagree even about the presence or lack of a pyloric caecum (e.g., compare Kott 1985 and C. Monniot 1987b). No report mentions the striking gold plaques between the siphons of our specimens. In this as well as some other ascidian species, Hawaiian animals evidently do not reach the large sizes attained elsewhere—12 cm in Australia, 10 cm in New Caledonia. *Polycarpa aurita* has been found in the western Atlantic (Caribbean, Gulf of Mexico) as well as in the Indo-Pacific (Australia, Malaysia, the Philippines, Indonesia, Micronesia, Melanesia).

What appears to be a mere variant of *Polycarpa aurita* lives in the seawater run-off system at the Kewalo Marine Laboratory in Honolulu, under rocks in tidepools at Hālona, O'ahu, and on the island of Hawai'i under nearshore rocks at Nāpō'opo'o in Hōnaunau Bay. The brown, leathery, corrugated, papillose tunic is encrusted with worm tubes, sponges, small mytilid molluscs, and other solitary ascidians. Large specimens (reaching 4 cm) may have 60–70 oral tentacles. The dorsal tubercle's ciliated slit is like an almost closed horseshoe. Gonads in small specimens are very stubby. In small specimens about 15 stomach grooves are visible externally, but they are more obscure in larger specimens.

In *Cnemidocarpa* the gonads are fewer and usually much more elongate than in *Polycarpa*, but structurally similar: the testes are encased tightly with the ovaries in sausage-like units. At least two species of *Cnemidocarpa* occur in Hawai'i. An as-yet-unidentified species is small (1 cm or less) with a bright red body within a colorless, sand-encrusted tunic. Very few specimens have been taken, and these only once, from 15 m deep beside the ship channel a quarter mile out from Honolulu Harbor; they were growing on *P. aurita*. The siphons are separated by at least half the length of the body. A 1 cm specimen has about a dozen oral tentacles; the ciliated slit is a simple hole in the dorsal tubercle. Its four pharyngeal folds per side carry 6–12 internal longitudinal vessels apiece; one vessel runs in each interfold space. There are 16–18 rows of stigmata. The gut is tightly recurved on itself; the stomach has a very long, completely curled pyloric caecum. This specimen has seven rather stubby (for *Cnemidocarpa*) parallel gonads on its right side and four on its left. Small endocarps are scattered about the atrium wall.

Hawai'i's other *Cnemidocarpa* is *C. areolata*. Large specimens may reach a length of 4 cm and have a dark brown to whitish tunic with deep wrinkles that crisscross to throw the surface into irregularly shaped raised areas. Smaller animals have a smoother tunic, but all specimens bear warts, especially around the bases of the siphons, and may be encrusted with debris and epibionts. The tunic is 1–3 mm thick and tough but flexible. A striking tetramerous pattern of broad purple-brown and narrow white stripes radiates from the top of each siphon and is often the easiest external clue to this species' identity in Hawai'i (Fig. 18a) and sometimes even to its otherwise encrusted presence at all. There are 20–30 oral tentacles; the ciliated slit of the dorsal tubercle is U-shaped. The four pharyngeal folds on each side carry a dozen or more internal longitudinal vessels apiece; each interfold space has a few such vessels. Along with these traits, the number and shape of the gonads (Fig. 18b) is enough to identify this species in Hawai'i. It grows on docks in Kāne'ohe Bay and was found occasionally on the walls of tanks in the old Honolulu Aquarium, presumably from the settlement of larvae entering the aquarium's seawater intake at Waikīkī. *Cnemidocarpa areolata* is a widespread

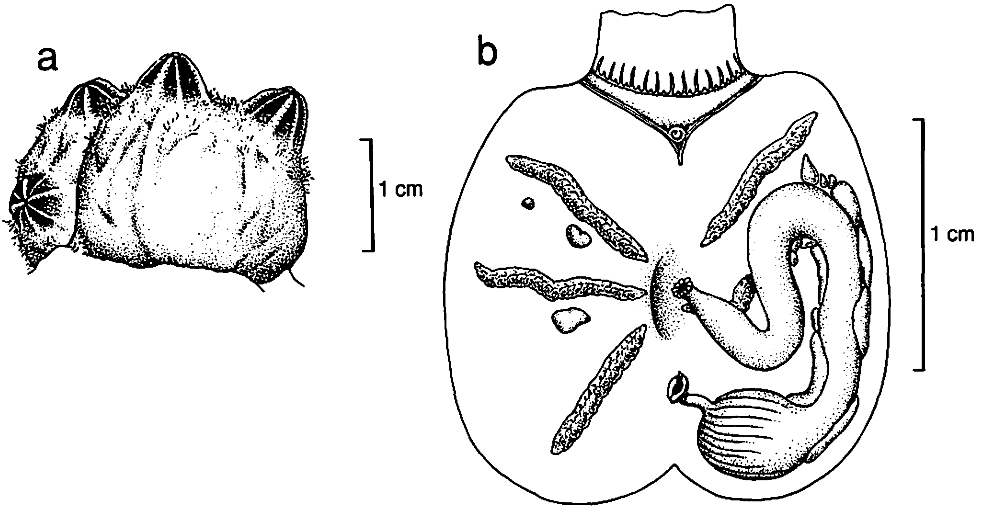

Figure 18.—*Cnemidocarpa areolata:* a, two entire, slightly contracted specimens on substrate; b, specimen opened along mid-ventral line, pharynx removed, showing oral tentacles, dorsal tubercle, gut loop, gonads, a few small endocarps amid gonads, and large flaplike endocarps alongside gut loop.

Indo-Pacific ascidian, found in Japan, the Philippines, Australia, Micronesia, and Melanesia. C. Monniot (1983b) also reports it from Guadeloupe, in the tropical western Atlantic, a record disputed by Kott (1992b).

What we call *Cnemidocarpa areolata* goes by several names, for experts disagree about our species' type specimen. We cannot resolve the debate here, but it illustrates a difficulty that can bedevil taxonomy. C. and F. Monniot (1984, also C. Monniot 1988) examined Heller's type specimen of *C. areolata* and found that each of its several sperm ducts opens through a little cluster of papillae, not by the single orifice per duct found in the widespread ascidian called *C. areolata.* They believe that Heller's (1878), and probably also Herdman's (1906), *C. areolata* grows only in Sri Lanka; for them, what Hartmeyer (1919) described from Australia as *C. valborg* is what grows widely in the western Pacific and is misidentified as Heller's Sri Lankan *C. areolata.* Nishikawa (1991) accepts the Monniots' case and goes a step farther, equating *C. valborg* with the Japanese *Styela irene* Hartmeyer, 1906, which would make *C. valborg* into *C. irene* (Hartmeyer, 1906). But Kott (1985, 1992b) cuts through all these revisions. She believes that Heller's *C. areolata* is the widespread species, that Hartmeyer's *C. valborg* (and, accepting Nishikawa's synonymies, Hartmeyer's *S. irene,* too) is synonymous with it, and that perhaps Heller simply by bad luck chose as the type specimen of this "variable species" (Kott 1985) an animal with aberrant sperm ducts.

This arcane biology of words may seem beyond the pale for naturalists. But it isn't. If we hope to assess the world's biodiversity accurately and to compare its taxa reliably, we must agree on species' names and on our evidence for their identity. Confusion we would not tolerate in our taxonomy of prominent vertebrates should not be allowed, either, in that of less familiar taxa. So our grasp of the sea's inhabitants will be a little firmer when this disagreement is resolved. Meanwhile, according to what paper one reads, any of three currently used *Cnemidocarpa* species names—*areolata*, *valborg*, and even *irene*—may refer to our local one.

Styela is a large, diverse genus of solitary but sometimes tightly aggregated styelids. The tunic varies in thickness and opacity, but it is tough and strong even when thin. The siphonal apertures are four-lobed, the pharynx has four folds on each side, and one or more gonads on each side bulge into the atrium from the body wall. A *Styela* gonad lacks the sausage-like compactness of those in *Polycarpa* or *Cnemidocarpa*. Instead, its many testis lobes lie only loosely associated with the long, sinuous ovaries. *Styela izuana hawaiiensis* is a Hawaiian subspecies of a form previously described from Japan. It is known only from a few specimens dredged in deep water. Its identifying traits are listed in the above key to the family.

Styela canopus, better known by its discarded synonym *Styela partita*, is the common styelid of Hawaiian inshore waters. Some specimens reach a height of 3 cm, but most are smaller. The ovoid body is attached posteriorly and tapers toward its free end, which bears the siphons. The tunic is gray or tan and relatively smooth in smaller animals, but rusty brown and ridged and creased in larger individuals. The short, rough, and somewhat papillated siphons have purple and white stripes down their length. This species has about 24–30 oral tentacles and a C-shaped ciliated slit in the dorsal tubercle. In a 1 cm specimen, each of the pharyngeal folds carries six to nine internal longitudinal vessels, and each interfold space carries one to three vessels; 2 cm specimens may have 15 or more vessels per fold and four per interfold space. *Styela canopus* is a highly variable species in outward appearance; internally, however, it usually can be recognized quickly by its stomach, which is noticeably longer and narrower (Fig. 19) than that of other Hawaiian styelids. Many Hawaiian specimens lack a gastric caecum but may have a prominent tubular cross-connection between the stomach and the proximal intestine. Two sinuous ovaries and associated testis lobes lie on each side of the body. The common sperm duct of each testis gathers the ducts from many lobes and rides along the atrial surface of an ovary to open near that ovary's gonopore. The atrium wall has many small endocarps.

Styela canopus is a common fouling organism in Pearl Harbor, Kāne'ohe Bay, and other quiet waters, where it attaches to boat hulls, docks, worm tubes, dead coral, and occasionally living surfaces such as other solitary ascidians. It occurs around the world in warm and temperate seas, doubtless spread by ships.

Family **Pyuridae**

This large family of solitary ascidians is represented in Hawai'i by two common species, *Herdmania momus* (Savigny, 1816) and *Microcosmus exasperatus* Heller, 1878, and an enigmatic one, *Microcosmus miniaceus* Sluiter, 1900. The genus *Pyura*, despite its wide abundance and variety, has not been found here.

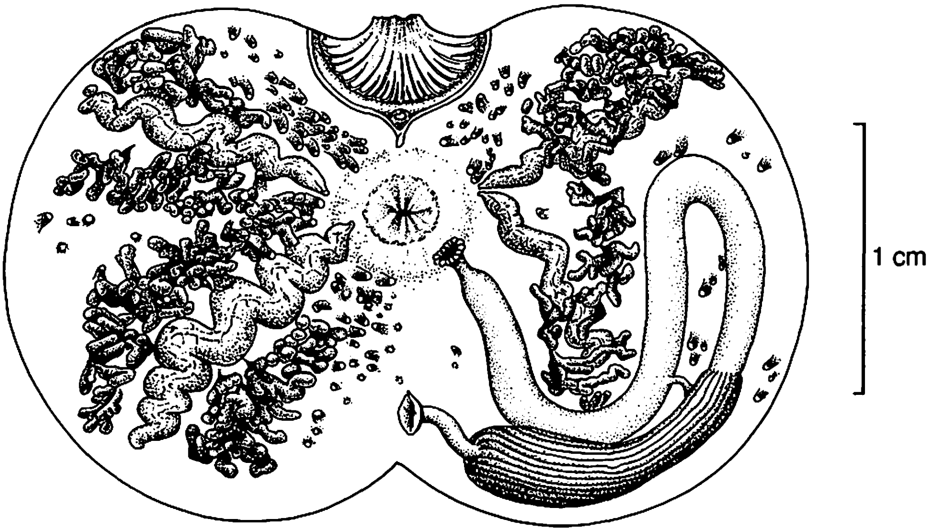

Figure 19.—*Styela canopus* opened along mid-ventral line, pharynx removed, showing oral tentacles, dorsal tubercle, gut loop with elongate stomach and gastro-intestinal connection, and sinuous ovaries with associated testis lobes. Sperm duct rides on each ovary, but most connections of testis lobes to sperm ducts have been lost in dissection. Small endocarps also lie in atrium wall. Drawing shows cut stumps of many blood vessels that had connected pharynx to mantle.

Pyurids look like especially robust solitary styelids. The tunic is usually tough and leathery, it is sometimes translucent (in *Herdmania*) but more often thick and opaque (in *Pyura* and *Microcosmus*), and it is often armored with warts and spines, especially within and around the four-lobed siphonal apertures. Some species nestle into crannies, with only their siphons exposed, and in many species the tunic is impregnated with sand or covered by debris or epibionts.

Pyurids have stout but rather feathery, fernlike, pinnately branched oral tentacles, in contrast to the styelids' thin, filiform tentacles. The dorsal tubercle's ciliated slit is generally prominent. The pharyngeal wall is deeply pleated into 6–10 longitudinal folds per side, rather than just the four or fewer of styelids. Many internal longitudinal vessels lie on the folds, and several run along most interfold spaces. The mid-dorsal line of the pharynx may have a smooth-edged dorsal lamina, as in *Microcosmus*, or carry a line of dorsal languets, as in *Pyura* and *Herdmania*.

Anteriorly the slim pyurid stomach is accompanied by hepatic or pyloric glands (the "liver"). These glands form lobulate or lamellar masses or dense, stout arborescent shapes of striking prominence and complexity. The gonads, attached to the atrium wall, are large and usually elaborate. Each has a long, slender ovary and many testis elements arranged around it. Usually there is one gonad on each side, the left one lying partly or wholly inside the gut loop. Endocarps protrude from the atrium wall of many pyurids but are rare or absent in the species recorded so far from Hawai'i.

Herdmania momus has tiny, needle-like, calcareous spicules scattered abundantly and widely in its mantle and in many other tissues (Lambert and Lambert 1987; Lambert 1992). They resemble monaxon sponge spicules but when highly magnified are seen to be composed of many rings of tiny spines (Fig. 20b). The validity of the genus has been hotly debated, since it has been defined by a single trait (these spicules) and has contained a single species. Recently, C. Monniot (1992) reversed his earlier position (1965 et seq.) and joined others (Kott 1985; Nishikawa 1991) in recognizing the genus on the basis of a suite of characters that includes its spicules, unlobed gonads, abundantly folded pharynx, and peculiar liver. And he has enlarged the genus to include both the widespread shallow-water *H. momus* and several recently described bathyal species from the western tropical Pacific.

Stout, spherical *Herdmania momus* may reach a diameter of several centimeters in Hawai'i. It adheres posteroventrally to the substrate. Smaller specimens are clean, smooth, translucent, and generally tinged with pink or purple; larger ones are wrinkled and more opaque but retain a ruddy cast to the body wall and some-

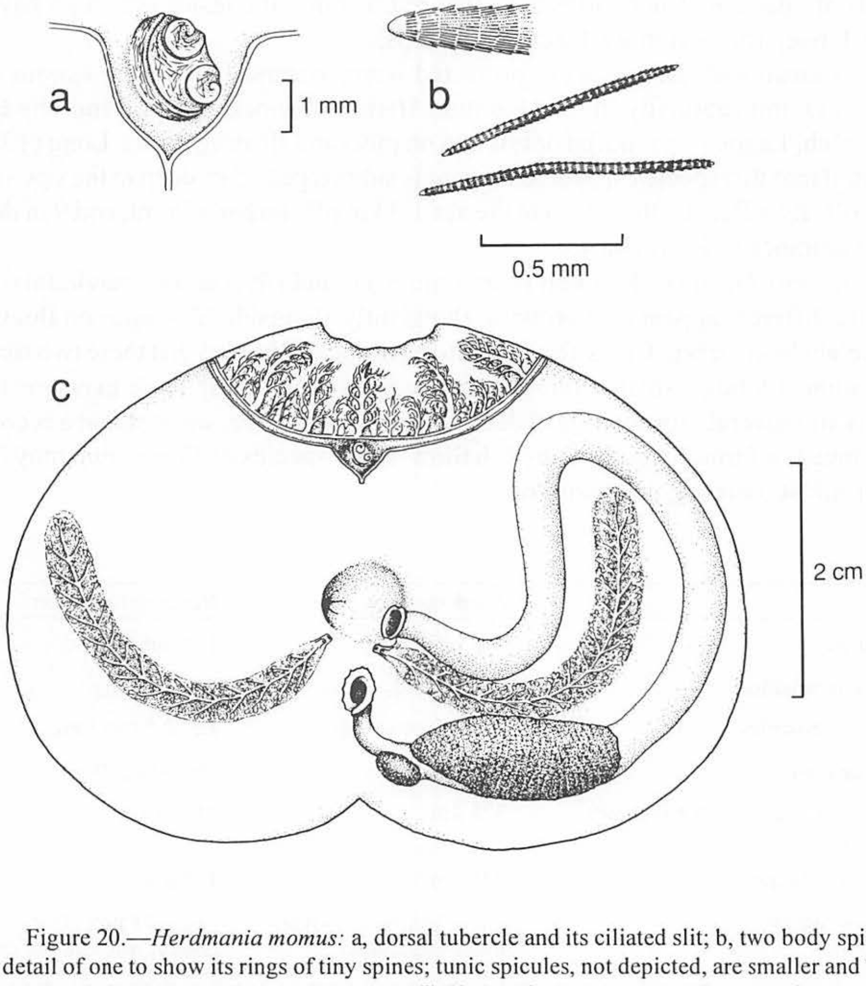

Figure 20.—*Herdmania momus:* a, dorsal tubercle and its ciliated slit; b, two body spicules, and detail of one to show its rings of tiny spines; tunic spicules, not depicted, are smaller and "have a mace-like ball of recurved spines at one end" (G. Lambert, pers. comm.); c, specimen opened along mid-ventral line, pharynx removed, showing oral tentacles, dorsal tubercle, gut loop, and gonads.

times also to the tunic. This color is often obscured by epibionts. Living specimens, even when brownish overall, have distinctive lengthwise stripes of orange-red or red and white down their siphons.

The *Herdmania momus* pharynx is guarded by a dozen or more large oral tentacles, plus fewer small ones. The tentacles vary from moderately pinnate to elaborately branched (Fig. 20c). The prominent dorsal tubercle looks like a cushion, usually with an inrolled U- or C-shaped ciliated slit (Fig. 20a). The pharynx has 6–12 folds per side, with about 10–25 internal longitudinal vessels per fold; only a few vessels run along each interfold space even in large specimens. The hepatic lobes, close to the esophagus, comprise two compact patches, one small and one large, of massed tubules on the wall of the widely curving gut loop. A long, sinuous gonad lies in the atrium wall on each side of the body, the left one entirely within the gut loop. In each gonad, dense rows of many small testis lobes tightly border either side of the long ovary and feed into a common sperm duct that rides along the ovary's atrial surface to open near the ovarian gonopore, close to the atrial aperture. The ovaries are often red or pink; the testes may also have a pinkish hue. The atrium wall lacks endocarps.

A cosmopolitan species in protected warm waters, *Herdmania momus* occurs on O'ahu subtidally in the Honolulu Harbor channel, and in Kāne'ohe Bay and Ke'ehi Lagoon on mudflat debris and on piers and floating docks. Long (1974) reported that this species appeared on panels submerged 15 m deep in the open sea 2 km off 'Ewa Beach, 30 m deep in the sea 1.3 km off Barber's Point, and 9 m deep in the entrance to Pearl Harbor.

In 1990, G. and C. Lambert (pers. comm.) found *Herdmania* individuals of a slightly different appearance growing abundantly alongside *H. momus* on floats at the Ke'ehi boat harbor. Cross-fertilization experiments showed that these two forms were almost totally isolated reproductively from each other. Their experimental results and several morphological details, summarized here, suggest that a second, introduced or formally undescribed shallow-water species of *Herdmania* may live in our midst, unrecognized until now.

	Herdmania sp.	*Herdmania momus*
tunic	4–6 mm thick	1–2 mm thick
tunic spicules	95 µm long	140 µm long
body spicules	1.5–2.5 mm long	1.5–2.5 mm long
egg color	pink	greenish yellow
egg diameter, with envelopes intact	375 µm	315 µm
ovum diameter	225 µm	190 µm
egg follicle cells	dia. = 13 µm; not clear; multi-vacuolated	dia. = 21 µm; clear; one big vacuole per cell
sperm	length of head = 8.8 µm overall length = 52.4 µm	length of head = 6.6 µm overall length = 53 µm

Unlike *Herdmania momus*, even small specimens of *Microcosmus exasperatus* are tough and leathery. These spherical animals grow to over 4 cm across. Rough outgrowths from the basal tunic help anchor the body to the substrate. The ruddy tunic is often covered with epibionts and debris. The well-separated siphons lack stripes, but their apertures often show an alternation of light and dark shallow marginal lobes. The inner surface of each siphon carries an armature of many tiny but stout, thornlike, flattened, noncalcareous spines.

The dozen large (and as many small) oral tentacles of *Microcosmus exasperatus* are fleshy and pinnate (Fig. 21c) but usually less elaborate than those of *H. momus*. The dorsal tubercle's ciliated slit forms a horseshoe shape with one or both ends inrolled (Fig. 21a). There are seven or eight pharyngeal folds on the left side, seven to nine on the right (Fig. 21a). In specimens 1 cm across, most folds carry 8–12 internal longitudinal vessels, and one to three vessels run along each interfold space. Large specimens have many more vessels per fold but still only a few per interfold space. The dorsal lamina is a smooth-edged membrane. The narrow gut loop has compact, tightly adhering, lamellar "liver" lobes (Fig. 21b) and an intestine that loops back close to the eosophagus. On each side of the body an elongate gonad is divided into a few prominent, well-separated lobes, in each of which the ovarian and testicular elements are closely combined (Fig. 21b). Each gonad's male and female gonopores lie close together near the atrial aperture. A distinguishing character of *Microcosmus* is that the left gonad drapes over the intestine and so lies only partly within the gut loop. In Hawaiian specimens, the atrium wall lacks endocarps.

Microcosmus exasperatus has been found in the tanks and seawater run-off trenches at the Kewalo Marine Laboratory. It grows regularly on floats and docks in Kāne‘ohe Bay. The species probably occurs worldwide in the tropics, having been found in the Caribbean and throughout the Indo-West Pacific. It reaches temperate waters in Australia and eastern Asia.

Sluiter (1900) described *Microcosmus miniaceus* from Pearl Harbor, but it has not been found again. He collected three slightly pedunculate specimens and dissected one. The 2 cm specimen he opened had seven pharyngeal folds per side. He does not report the number of internal longitudinal vessels per fold but does remark that there are only three such vessels per interfold space. Sluiter described and depicted club-shaped papillae projecting into the pharynx "everywhere between the folds," especially prominently from the internal longitudinal vessels. The pharynx of a 2 cm *M. exasperatus* may have this number of folds and interfold vessels, but it lacks the internal papillae that so struck Sluiter.

Regrettably, without the spur of modern records, we have not reexamined Sluiter's specimens. Nor, apparently, did C. Monniot in preparing his review (1965) of the family Pyuridae. Until this is done, we agree with Monniot that *Microcosmus miniaceus* should be accepted as a species, not put aside as aberrant specimens of *M. exasperatus*, a species Sluiter surely knew well. It is tantalizing that even such a well-visited place as Pearl Harbor should not yield new specimens of *M. miniaceus* in this century, nor have any pharyngeally papillated specimens of *M. exasperatus* been found there. We can only urge that the search be pressed—as, indeed, we urge it be pressed in new ways for ascidians of all sorts throughout Hawai‘i.

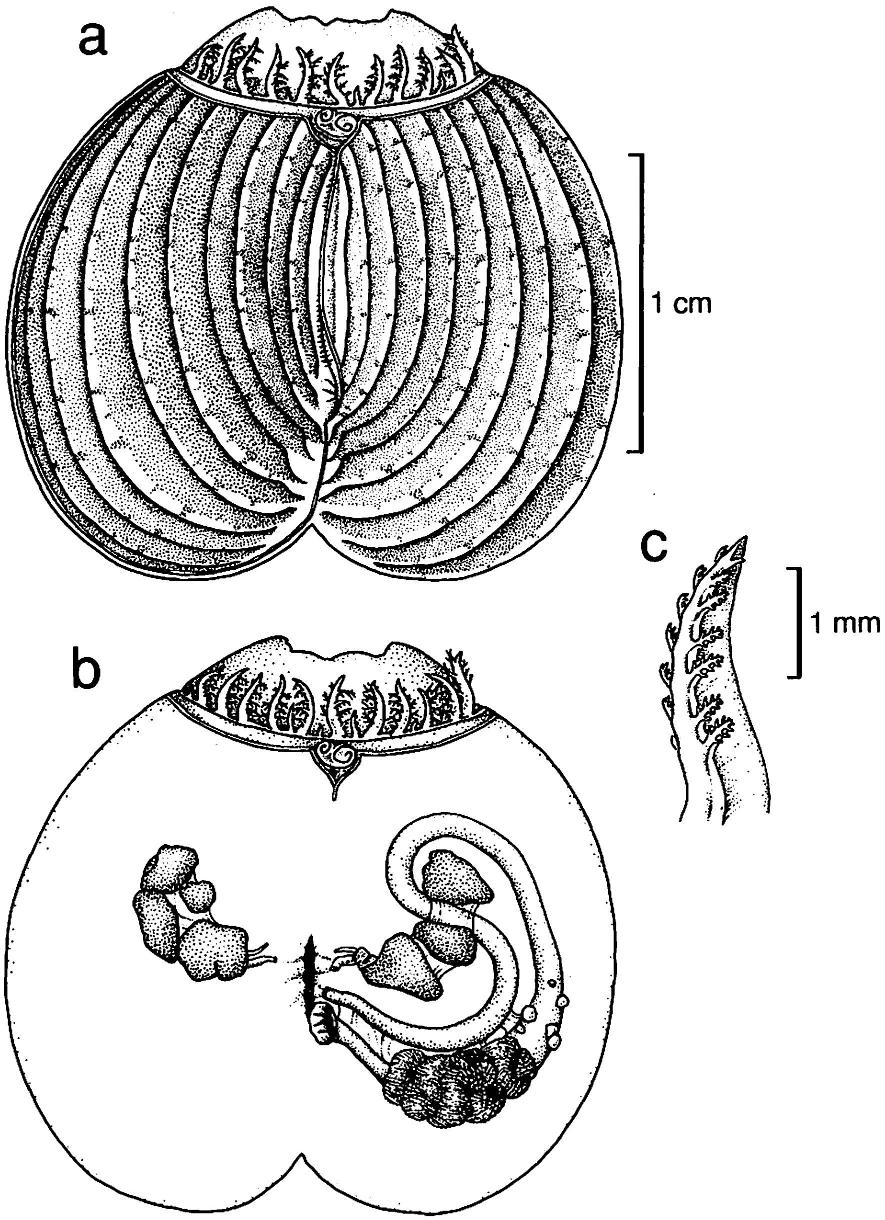

Figure 21.—*Microcosmus exasperatus:* a, specimen opened along mid-ventral line, pharynx left intact (endostyle at far left of drawing); note oral tentacles, dorsal tubercle and its ciliated slit, pharyngeal folds, and dorsal lamina; b, same specimen with pharynx removed to show gut loop and gonads (cf. *Herdmania momus*, Fig. 20c); c, oral tentacle.

REFERENCES

Includes uncited continuations of regional series; bibliographic compilation through 1995.

Abbott, D. P. 1954. Phylum Chordata: sub-phylum Urochordata (=Tunicata). In *Intertidal Invertebrates of the Central California Coast* (2nd edition) (R. I. Smith, F. A. Pitelka, D. P. Abbott, and F. M. Weesner, eds.), pp. 298–310. Berkeley: Univ. Calif. Press.

Abbott, D. P. 1975. Phylum Chordata: introduction and Urochordata. In *Light's Manual: Intertidal Invertebrates of the Central California Coast* (3rd edition) (R. I. Smith and J. T. Carlton, eds.), pp. 638–655. Berkeley: Univ. Calif. Press.

Ballan-Dufrançais, C., A. Y. Jeantet, and M. Truchet. 1995. La formation des spicules de Didemnidae (Ascidiacea). *Canad. J. Zool. 73*:1647–1656.

Berrill, N. J. 1950. *The Tunicata with an Account of the British Species*. 354 pp. London: Ray Soc.

Brien, P. 1948. Embranchement des Tuniciers. In *Traité de Zoologie* (P. Grassé, ed.) vol. 11, pp. 553–930. Paris: Masson.

Carlton, J. T., and J. B. Geller. 1993. Ecological roulette: the global transport of nonindigenous marine organisms. *Science 261*:78–82.

Caullery, M. 1909. Récherches sur les synascidies du genre *Colella* et considérations sur la famille des Distomidae. *Bull. Sci. France Belg. 42*:1–59, pl. 1.

Eldredge, L. G. 1967. A taxonomic review of the Indo-Pacific didemnid ascidians and descriptions of twenty-three Central Pacific species. *Micronesica 2*:161–261.

Flood, P. R., and A. Fiala-Medioni 1981. Ultrastructure and histochemistry of the food trapping mucous film in benthic filter-feeders (ascidians). *Acta Zool. 62*:53–65.

Gaertner, C. F. von 1774. Zoophyta . . . In *Spicilegia Zoologica* . . . (P. S. Pallas *et al.*) *10*:17–41, pls. 1 and 4. Berlin: G. A. Lange [citation compiled from Hartmeyer 1909–11, Hopkinson 1913, Michaelsen 1915, and Kott 1985].

Galigher, A. E., and E. N. Kozloff. 1964. *Essentials of Practical Microtechnique*. 484 pp. Philadelphia: Lea & Febiger.

Garstang, W. 1891. Note on a new and primitive type of compound ascidian. *Ann. Mag. Nat. Hist.*, ser. 6, *8*:265–268.

Godeaux, J. 1974. Introduction to the morphology, phylogenesis, and systematics of lower Deuterostomia. In *Chemical Zoology* (M. Florkin and B. T. Sheer, eds.) vol. 8, pp. 3–60. New York: Academic Press.

Goodbody, I. 1974. The physiology of ascidians. *Adv. Mar. Biol. 12*:1–149.

Hartmeyer, R. 1909–11. Ascidien [treatise begun by O. Seeliger]. In *Klassen und Ordnungen des Tier-Reichs* (H. G. Bronn, ed.) vol. 3, suppl., pp. 1281–1773. Leipzig: C. F. Winter.

Hartmeyer, R. 1919. Ascidien. In *Results of Dr. E. Mjöbergs Swedish Scientific Expeditions to Australia 1910–13. Kungl. Svenska Vetensk.-Akad. Handl. 60*:1–150, pls. 1–2.

Hartmeyer, R. 1924. Ascidiacea, Part II. *The Danish Ingolf-Expedition* vol. 2, pt. 7, pp. 1–278. Copenhagen: Hagerup.

Heller, C. 1878. Beiträge zur nähern Kenntniss der Tunicaten. *Sitzungsb. K. Akad. Wiss., Vienna, 77*:83–110, pls. 1–6.

Herdman, W. A. 1880. Preliminary report on the Tunicata of the Challenger expedition. I. Ascidiadae. *Proc. Roy. Soc. Edinburgh 10*:458–472.

Herdman, W. A. 1881. Preliminary report on the Tunicata of the Challenger expedition. III. Cynthiidae. *Proc. Roy. Soc. Edinburgh 11*:52–88.

Herdman, W. A. 1882. Report on the Tunicata. Part I. Ascidiae Simplices. In *Report on the Scientific Results of the Voyage of H.M.S. Challenger during the Years 1873–76* (C. Wyville Thomson and J. Murray, eds.), Zoology, *6*:1–296, pls. 1–37.

Herdman, W. A. 1886. Report on the Tunicata. Part II. Ascidiae Compositae. In *Report on the Scientific Results of the Voyage of H.M.S. Challenger during the Years 1873–76* (C. Wyville Thompson and J. Murray, eds.), Zoology, *14*:1–432, pls. 1–49, map.

Herdman, W. A. 1906. On the Tunicata. *Report to the Government of Ceylon on the Pearl Fisheries of the Gulf of Manaar*, pt. 5, suppl. rpt. 39:295–348, pls. 1–9. London: Royal Soc.

Hopkinson, J. 1913. *A Bibliography of the Tunicata 1469–1910.* 288 pp. London: Ray Soc.

Humason, G. L. 1979. *Animal Tissue Techniques* (4th ed.). 661 pp. San Francisco: W. H. Freeman.

Huus, J. 1937. Ascidiaceae. In *Handbuch der Zoologie* (W. Kukenthal and T. Krumbach, eds.) vol. 5, pt. 2, pp. 545–692. Berlin: W. de Gruyter.

Jackson, J. B. C. 1979. Morphological strategies of sessile animals. In *Biology and Systematics of Colonial Organisms* (G. Larwood and B. R. Rosen, eds.), pp. 499–555. New York: Academic Press.

Kniprath, E., and F. Lafargue. 1980. Spicule formation in the Didemnidae (compound ascidians). In *The Mechanisms of Biomineralization in Animals and Plants: Proc. 3rd Intl. Biomin. Symp.* (M. Omori and N. Watabe, eds.), pp. 31–36. Tokyo: Tokai Univ. Press.

Kott, P. 1952a. Observations on compound ascidians of the Plymouth area, with descriptions of two new species. *J. Mar. Biol. Assoc. U. K. 31*:65–83.

Kott, P. 1952b. The ascidians of Australia I. Stolidobranchiata Lahille and Phlebobranchiata Lahille. *Austral. J. Mar. Freshw. Res. 3*:205–333.

Kott, P. 1957a. The sessile Tunicata. In *Sci. Repts. John Murray Exped. 10*:129–149.

Kott, P. 1957b. The ascidians of Australia II: Aplousobranchiata Lahille: Clavelinidae Forbes & Hanly and Polyclinidae Verrill. *Austral. J. Mar. Freshw. Res. 8*:64–110.

Kott, P. 1964. Stolidobranch and phlebobranch ascidians of the Queensland coast. *Papers Dept. Zool. Univ. Qld. 2*:127–152.

Kott, P. 1969. Antarctic Ascidiacea. *Antarctic Res. Ser.* vol. 13. *Amer. Geophys. Union Publ.* no. 1725, pp. 1–239.

Kott, P. 1980. Algal-bearing didemnid ascidians in the Indo-West-Pacific. *Mem. Queensland Mus. 20*:1–47, pls. 1–4.

Kott, P. 1981a. The ascidians of the reef flats of Fiji. *Proc. Linn. Soc. N. S. W. 105*:147–212.

Kott, P. 1981b. Replication in the Ascidiacea: an adaptive strategy in the coral reef environment. *Proc. 4th Intl. Coral Reef Symp.*, Manila 1981, *2*:725–733.

Kott, P. 1985. The Australian Ascidiacea Part I, Phlebobranchia and Stolidobranchia. *Mem. Queensland Mus. 23*:1–440.

Kott, P. 1989a. The family Hexacrobylidae Seeliger, 1906 (Ascidiacea, Tunicata). *Mem. Queensland Mus. 27*:517–534.

Kott, P. 1989b. Form and function in the Ascidiacea. *Bull. Mar. Sci. 45*:253–276.

Kott, P. 1990. The Australian Ascidiacea Part 2, Aplousobranchia (1). *Mem. Queensland Mus. 29:*1–266.

Kott, P. 1992a. The Australian Ascidiacea Part 3, Aplousobranchia (2). *Mem. Queensland Mus. 32*:375–620.

Kott, P. 1992b. The Australian Ascidiacea, Supplement 2. *Mem. Queensland Mus. 32*:621–655.

Kott, P. (in prep.) The Australian Ascidiacea Part 4, (Didemnidae). *Mem. Queensland Mus.*

Kott, P., and I. Goodbody. 1982. The ascidians of Hong Kong. In *Proc. First Intl. Mar. Biol. Workshop: The Marine Flora and Fauna of Hong Kong and Southern China, Hong Kong, 1980* (B. S. Morton and C. K. Tseng, eds.), pp. 503–554. Hong Kong: Hong Kong Univ. Press.

Lafargue, F., and E. Kniprath. 1978. Formation des spicules de Didemnidae (Ascidies Composées). I. L'apparition des spicules chez l'oozoïde après la métamorphose. *Mar. Biol. 45*:175–184.

Lahille, F. 1886. Sur la classification des tuniciers. *C. R. Acad. Sci. Paris 102*:446–448.

Lahille, F. 1888. Etude systématique des tuniciers. *C. R. Assoc. Franç. Avanc. Sci., Toulouse 16* (ann. 1887):667–677.

Lambert, G. 1992. Ultrastructural aspects of spicule formation in the solitary ascidian *Herdmania momus* (Urochordata, Ascidiacea). *Acta Zool. 73*:237–245.

Lambert, G., and C. Lambert. 1987. Spicule formation in the solitary ascidian *Herdmania momus. J. Morph. 192*:145–159.

Linnaeus (Linné), C. 1767. *Systema Naturae*, 12th ed., vol. 2., pt. 2. Stockholm.

Long, E. R. 1974. Marine fouling studies off O'ahu, Hawai'i. *Veliger 17*:23–36.

Michaelsen, W. 1914. Über einige westafrikanische Ascidien. *Zool. Anz. 43*:423–432.

Michaelsen, W. 1915. Tunicata. In *Beiträge zur Kenntnis der Meeresfauna Westafrikas* (W. Michaelsen, ed.), pp. 321–518, pls. 16–19, map. Hamburg: L. Friederichsen.

Millar, R. H. 1964. South African ascidians collected by Th. Mortensen, with some additional material. *Vidensk. Meddel. Dansk Naturh. Foren. 127*:159–180.

Millar, R. H., 1971. The biology of ascidians. *Adv. Mar. Biol. 9*:1–100.

Millar, R. H. 1975. Ascidians from the Indo-West Pacific region in the Zoological Museum, Copenhagen (Tunicata, Ascidiacea). *Steenstrupia 3*:205–336.

Millar, R. H. 1977. Ascidians (Tunicata: Ascidiacea) from the northern and north-eastern Brazilian Shelf. *J. Nat. Hist. 11*:169–223.

Milne-Edwards, H. 1841. Observations sur les Ascidies Composées des Côtes de la Manche. 110 pp., 8 pls. Paris: Fortin-Masson (and in *Mém. Acad. Sci. Inst. France 18*:217–326, pls. 1–8).

Monniot, C. 1965. Etude systématique et évolutive de la famille des Pyuridae (Ascidiacea). *Mém. Mus. Natn. Hist. Nat.*, Paris, (A), *36*:1–203.

Monniot, C. 1970. Ascidies phlébobranches et stolidobranches. In *Campagne de la "Calypso" au large des côtes atlantiques de l'Amérique du Sud (1961–1962). Ann. Inst. Océanogr., Paris, 47*:33–59.

Monniot, C. 1972. Ascidies phlébobranches des Bermudes. *Bull. Mus. Natn. Hist. Nat., Paris*, 3e sér., Zoologie, *61*:939–948.

Monniot, C. 1983a. Ascidies littorales de Guadeloupe II. Phlébobranches. *Bull. Mus. Natn. Hist. Nat., Paris*, 4e sér., (A), *5*:51–71.

Monniot, C. 1983b. Ascidies littorales de Guadeloupe IV. Styelidae. *Bull. Mus. Natn. Hist. Nat., Paris*, 4e sér, (A), *5*:423–456.

Monniot, C. 1987a. Ascidies de Nouvelle-Calédonie I. Phlébobranches du lagon. *Bull. Mus. Natn. Hist. Nat., Paris*, 4e sér., (A), *9*:3–31.

Monniot, C. 1987b. Ascidies de Nouvelle-Calédonie II. Les genres *Polycarpa* et *Polyandrocarpa*. *Bull. Mus. Natn. Hist. Nat., Paris*, 4e sér, (A), *9*:275–310.

Monniot, C. 1988. Ascidies de Nouvelle-Calédonie IV. Styelidae (suite). *Bull. Mus. Natn. Hist. Nat., Paris*, 4e sér., (A), *10*:163–196.

Monniot, C. 1989. Ascidies de Nouvelle-Calédonie VI. Pyuridae et Molgulidae. *Bull. Mus. Natn. Hist. Nat., Paris*, 4e sér., (A), *11*:475–507.

Monniot, C. 1990. Ascidies de Nouvelle-Calédonie VIII. Phlébobranches (*suite*). *Bull. Mus. Natn. Hist. Nat., Paris*, 4e sér, (A), *12*:491–515.

Monniot, C. 1991. Ascidies de Nouvelle-Calédonie X. Stolidobranches (*suite*). *Bull. Mus. Natn. Hist. Nat., Paris*, 4e sér., (A), *13*:3–37.

Monniot, C. 1992. Ascidies de Nouvelle-Calédonie. XI. Phlébobranches et stolidobranches du plateau des Chesterfield. *Bull. Mus. Natl. Hist. Nat., Paris*, 4e sér., (A), *14*:3–22.

Monniot, C., and F. Monniot. 1972. Clé mondiale des genres d'Ascidies. *Arch. Zool. Exp. Gén. 113*:311–367.

Monniot, C., and F. Monniot. 1976. Ascidies de la côte du Mozambique. *Rev. Zool. Afr. 90*:357–392, pl. 6.

Monniot, C., and F. Monniot. 1984. Ascidies littorales de Guadeloupe VII. Espèces nouvelles et complémentaires à l'inventaire. *Bull. Mus. Natn. Hist. Nat., Paris*, 4e sér, (A), *6*:567–582.

Monniot, C., and F. Monniot. 1987. Les ascidies de Polynésie française. *Mém. Mus. Natn. Hist. Nat.*, Paris, (A), *136*:1–155.

Monniot, C., and F. Monniot. 1990. Revision of the class Sorberacea (benthic tunicates) with descriptions of seven new species. *Zool. J. Linn. Soc. London 99*:239–290.

Monniot, C., and F. Monniot. 1994. Additions to the inventory of eastern tropical Atlantic ascidians: arrival of cosmopolitan species. *Bull. Mar. Sci. 54*:71–93.

Monniot, C., F. Monniot, and F. Gaill. 1975. Les Sorberacea: une nouvelle classe de tuniciers. *Arch. Zool. Exp. Gén. 116*:77–122.

Monniot, C., F. Monniot, and P. Laboute. 1991. *Coral Reef Ascidians of New Caledonia*. 248 pp. Paris: ORSTOM.

Monniot, F. 1969. Sur une collection d'ascidies composées de Dakar. *Bull. Mus. Natn. Hist. Nat., Paris*, 2e sér., *41*:426–457.

Monniot, F. 1972. Ascidies aplousobranches des Bermudes. Polyclinidae et Polycitoridae. *Bull. Mus. Natn. Hist. Nat., Paris*, 3e sér., Zoologie, *61*:949–962.

Monniot, F. 1983a. Ascidies littorales de Guadeloupe I. Didemnidae *Bull. Mus. Natn. Hist. Nat., Paris*, 4e sér, (A), *5*:5–49.

Monniot, F. 1983b. Ascidies littorales de Guadeloupe V. Polycitoridae. *Bull. Mus. Natn. Hist. Nat., Paris*, 4e sér., (A), *5*:999–1019.

Monniot, F. 1987. Ascidies de Nouvelle-Calédonie III. Polyclinidae du lagon. *Bull. Mus. Natn. Hist. Nat., Paris*, 4e sér., (A), *9*:499–535.

Monniot, F. 1988. Ascidies de Nouvelle-Calédonie V. Polycitoridae du lagon. *Bull. Mus. Natn. Hist. Nat., Paris*, 4e sér., (A), *10*:197–235.

Monniot, F. 1989. Ascidies de Nouvelle-Calédonie VII. Les genres *Atriolum* et *Leptoclinides* dans le lagon sud. *Bull. Mus. Natn. Hist. Nat., Paris*, 4e sér., (A), *11*:673–691.

Monniot, F. 1990. Ascidies de Nouvelle-Calédonie IX. Le genre *Trididemnum*. *Bull. Mus. Natn. Hist. Nat., Paris*, 4e sér., (A), *12*:517–529.

Monniot, F. 1992. Ascidies de Nouvelle-Calédonie XII. Le genre *Lissoclinum* (Didemnidae) dans le lagon sud. *Bull. Mus. Natn. Hist. Nat., Paris*, 4e sér. (A), *14*:565–589.

Monniot, F. 1993. Ascidies de Nouvelle-Calédonie XIII. Le genre *Polysyncraton* (Didemnidae). *Bull. Mus. Natn. Hist. Nat., Paris*, 4e sér., (A), *15*:3–17.

Monniot, F. 1994a. Ascidies de Nouvelle-Calédonie XIV. Le genre *Diplosoma* (Didemnidae). *Bull. Mus. Natn. Hist. Nat., Paris*, 4e sér., (A), *16*:3–11.

Monniot, F. 1994b. Ascidies de Nouvelle-Calédonie XV. Le genre *Didemnum*. *Bull. Mus. Natn. Hist. Nat., Paris*, 4e sér, (A), *16*:299–344.

Nishikawa, T. 1984. Ascidians from the Truk Islands, Ponape Island and Majuro Atoll (Tunicata, Ascidiacea). *Proc. Japn. Soc. Syst. Zool. 27*:107–140.

Nishikawa, T. 1986. Ascidians from the Gilbert and Solomon Islands and Nauru. I. Perophoridae, Ascidiidae and Corellidae. *Proc. Japn. Soc. Syst. Zool. 32*:30–78.

Nishikawa, T. 1990. The ascidians of the Japan Sea. I. *Publ. Seto Mar. Biol. Lab. 34*:73–148.

Nishikawa, T. 1991. The ascidians of the Japan Sea. II. *Publ. Seto Mar. Biol. Lab. 35*:25–170.

Nishikawa, T. 1992. The ascidians of the Japan Sea. III. *Publ. Seto Mar. Biol. Lab. 35*:303–334.

Oka, A. 1927a. Ascidiacea. In *Figuraro de Japanaj Bestoj* [cited in T. Tokioka 1953, 1954; and in P. Kott 1957b, 1990].

Oka, A. 1927b. Über eine *Perophora* aus Japon. *Proc. Imperial Acad.*, Japan *3*:558–560.

Pizon, A. 1908. Ascidies d'Amboine. *Rev. Suisse Zool. 16*:195–240, pls. 9–14.

Redikorzev, V. 1927. Zehn neue Ascidien aus dem fernen Osten. *Zool. Jahrb.* (*Syst.*) *53*:373–404.

Ritter, W. E. 1893. Tunicata of the Pacific coast of North America. I. *Perophora annectens* n. sp. *Proc. Cal. Acad. Sci.*, ser. 2, *4*:37–85, pls. 1–3.

Rowe, F. W. E. 1966. A review of the genus *Diplosoma* Macdonald, 1859, (Ascidiacea: Didemnidae) with a description of the proposed neotype of *Diplosoma listerianum* (Milne Edwards), 1841. *Ann. Mag. Nat. Hist.*, ser. 13, *9*:457–467.

Ruppert, E. E. 1990. Structure, ultrastructure and function of the neural gland complex of *Ascidia interrupta* (Chordata, Ascidiacea): clarification of hypotheses regarding the evolution of the vertebrate anterior pituitary. *Acta Zool. 71*:135–149.

Savigny, M. J. C. L. de. 1816. Recherches anatomiques sur les ascidies composées et sur les ascidies simples. In *Mémoires sur les Animaux sans Vertèbres* vol. 2, 240 pp., 24 pls. Paris.

Sluiter, C. P. 1885. Über einige einfachen ascidien von der Insel Billiton. *Natuurk. Tijdschr. Nederl. Ind. 45*:160–232, pls. 1–9.

Sluiter, C. P. 1890. Die Evertebraten aus der Sammlung des königlichen naturwissenschaftlichen Vereins in Niederlädisch Indien in Batavia. Zugleich eine Skizze der Fauna des Java-Meeres mit Beschreibung der neuen Arten. Ascidiae simplices. *Natuurk. Tijdschr. Nederl. Ind. 50*:329–348, pls. 1–2.

Sluiter, C. P. 1895. Tunicaten. In *Zoologische Forschungsreisen in Australien und dem Malayischen Archipel.* (R. Semon, ed.) vol. 5, pt. 2. *Denkschr. Med. Naturwiss. Gesellsch. Jena, 8*:163–186, pls. 6–10, addendum, pp. 325–326, pl. 22.

Sluiter, C. P. 1898. Beiträge zur Kenntniss der Fauna von Süd-Afrika. II. Tunicaten von Süd-Afrika. *Zool. Jahrb.* (*Syst.*) *11*:1–64, pls. 1–7.

Sluiter, C. P. 1900. Tunicaten aus dem Stillen Ocean. *Zool. Jahrb.* (*Syst.*) *13*:1–35, pls. 1–6.

Sluiter, C. P. 1904. Die Tunicaten der Siboga-Expedition. I. Die socialen und holosomen Ascidien. *Siboga-Expeditie 56a*:1–126, addendum, pls. 1–15.

Sluiter, C. P. 1909. Die Tunicaten der Siboga-Expedition. II. Die merosomen Ascidien. *Siboga-Expeditie 56b*:1–112, pls. 1–8.

Stimpson, W. 1852. Several new ascidians from the coast of the United States. *Proc. Boston Soc. Nat. Hist. 4*:228–232.

Stimpson, W. 1855. Descriptions of some new Invertebrata [from the Chinese and Japanese Seas]. *Proc. Acad. Nat. Sci. 7*:385–394 (ascidians, pp. 387–388) [seq. to another paper of the same title, ibid., *7*:375–384 (ascidians, pp. 377–378)].

Tokioka, T. 1950. Ascidians from the Palao Islands. I. *Publ. Seto Mar. Biol. Lab. 1*:115–150, pls. 9–10.

Tokioka, T. 1953. *Ascidians of Sagami Bay.* 315 pp, 79 pls, map. Tokyo: Iwanami Shoten.

Tokioka, T. 1954a. Contributions to Japanese ascidian fauna VII. Invertebrate fauna of the intertidal zone of the Tokara Islands, VII. Ascidians *Publ. Seto Mar. Biol. Lab. 3*:239–264, pls. 18–37.

Tokioka, T. 1954b. Contributions to Japanese ascidian fauna IX. Re-descriptions of Oka's species found in "Figuraro de Japanaj Bestoj." *Publ. Seto Mar. Biol. Lab. 4*:69–74, pl. 4.

Tokioka, T. 1961. Ascidians collected during the Melanesia expedition of the Osaka Museum of Natural History. I. Ascidians presented by Dr. R. L. A. Catala of the aquarium of Noumea. *Publ. Seto Mar. Biol. Lab. 9*:103–138, pl. 5.

Tokioka, T. 1963. A new remarkable synstyelid, *Polyandrocarpa (Eusynstyela) transversalis* n. sp., from Hawai'i. *Publ. Seto Mar. Biol. Lab. 11*:101–103, pl. 3.

Tokioka, T. 1967. Pacific Tunicata of the United States National Museum. *U.S. Natl. Mus. Bull. 251:1–247.*

Traustedt, M. P. A. 1882. Vestindiske Ascidiae simplices, Første Afdeling (Phallusiadae). *Vidensk. Meddel. Naturh. Foren. Kjøbenhavn* ann. *1881*:257–288, pls. 4–5.

Van Name, W. G. 1902. The ascidians of the Bermuda Islands. *Trans. Conn. Acad. Arts Sci. 11*:325–412, pls. 46–64.

Van Name, W. G. 1912. Simple ascidians of the coasts of New England and neighboring British provinces. *Proc. Boston Soc. Nat. Hist. 34*:439–619, pls. 43–73.

Van Name, W. G. 1918. Ascidians from the Philippines and adjacent waters. *U.S. Natl. Mus. Bull. 100,* vol. 1(pt.2):49–174, pls. 23–33.

Van Name, W. G. 1921. Ascidians of the West Indian region and southwestern United States. *Bull. Amer. Mus. Nat. Hist. 44*:283–494.

Van Name, W. G. 1924. Ascidians from Curaçao. (Bijdragen tot de kennis der fauna van Curaçao.) *Bijdr. Dierk. 23*:23–32.

Van Name, W. G. 1930. The ascidians of Porto Rico and the Virgin Islands. *Sci. Surv. Porto Rico and the Virgin. Is. 10*:401–512, pls. 5–8. New York: N. Y. Acad. Sci.

Van Name, W. G. 1931. New North and South American ascidians. *Bull. Amer. Mus. Nat Hist. 61*:207–225.

Van Name, W. G., 1945. The North and South American ascidians. *Bull. Amer. Mus. Nat. Hist. 84*:1–476, pls. 1–31.

Vasseur, P. 1967. Contribution a l'étude des ascidies de l'Ile Maurice (Archipel des Mascareignes, Océan Indien). *Rec. Trav. Sta. Mar. Endoume* suppl. 6(hors sér):101–139.

Vázquez Otero, M. E. 1993. *Estudio Faunistico Autoecológico y Biogeográfico de los Tunicados Bentónicos de la Ria de Ferrol (Galicia).* Ph.D. dissertation. Universidade de Santiago de Compostela, Spain. 434 pp.

Verrill, A. E. 1871. On the distribution of marine animals on the southern coast of New England. *Amer. J. Sci. Arts,* ser. 3, *2*:357–362.

Weesner, F. M. 1960. *General Zoological Microtechniques.* 230 pp. Baltimore: Williams & Wilkins.

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