

The Anatomy, Life Habits, and Later Development of a New Species of Enteropneust, *Harrimania planktophilus* (Hemichordata: Harrimaniidae) From Barkley Sound

CHRISTOPHER B. CAMERON

Department of Biological Sciences, CW 405 Biological Sciences Building, University of Alberta, Edmonton, Alberta T6G 2E9, Canada

Abstract. A new species of enteropneust, *Harrimania planktophilus*, lives intertidally and subtidally in mixed sediments in Barkley Sound, British Columbia, Canada. *H. planktophilus* has a long proboscis skeleton extending into the pharyngeal region. The collar (mesosome) has complete dorsal and ventral mesenteries. The trunk (metasome) has four distinct regions that can be recognized externally: the branchial region, esophageal region, hepatic region, and an undifferentiated intestinal region leading to the anus. The dorsal pharynx is large and has long gill slits without synapticles. Posterior to the gills is a constriction followed by a short esophageal region and a long gonadal region. The paired dorsolateral gonads extend almost to the end of the trunk. Eggs in the ovaries appear amber yellow, and the testes appear slightly paler. The trunk terminates at an anus with a well-developed sphincter muscle. *H. planktophilus* forms long sinuous burrows that are semipermanent and shared. Females deposit a tubular egg mass in a burrow in which the embryos develop directly into juveniles. Gastrulation appears to be by invagination, followed by a ciliated stage that has a telotrochal swimming band, suggesting that the ancestor to *H. planktophilus* developed *via* a tornaria larva. The juveniles emerge from the egg membrane with a ventral post-anal tail and assume an interstitial burrowing life habit. The post-anal tail, mode of development, small size and correlated simplification in body plan suggest that *H. planktophilus* is closely related to *Saccoglossus*, and together these worms may be sister taxa to the colonial

Pterobranchia. A taxonomic key is provided to the enteropneust genera, and to the species of *Harrimania*.

Introduction

The deuterostome phylum Hemichordata is composed of three extant classes: the Enteropneusta are solitary worms, the Pterobranchia are colonial tube-dwelling suspension feeders, and the Planktosphaeroidea are known only from their larvae. The enteropneusts and pterobranchs differ sufficiently in size and modes of reproduction and feeding that some investigators have considered the phylum Hemichordata to be polyphyletic (Jeffries, 1986; Nielsen, 1994). A recent molecular study strongly supports hemichordate monophyly and suggests an entirely new hypothesis for the evolution of the hemichordates (Cameron *et al.*, 2000). That study proposes that the pterobranchs are a sister taxon to the enteropneust family Harrimaniidae, and that this clade is a sister taxon to the enteropneust family Ptychoderidae (*i.e.*, enteropneust paraphyly). If this phylogeny is correct, then detailed studies of the morphology and development of the harrimaniid worms should yield important insights into the biology and evolution of the ancestral hemichordate and the colonial pterobranchs.

Thirteen genera are currently recognized from four families of Enteropneusta: the Ptychoderidae, including the familiar *Balanoglossus*; the monotypic hydrothermal vent Saxipendiidae (Franzén *et al.*, 1985; Woodwick and Sensenbaugh, 1985); the Spengeliidae; and the Harrimaniidae, including the genera *Protoglossus*, *Saccoglossus*, *Xenopleura*, *Stereobalanus*, and *Harrimania*. Harrimaniids are the most morphologically simple of the Enteropneusta; they lack hepatic sacs, synapticles joining adjacent gill bars, and

Received 9 November 2000; accepted 23 January 2002.

Current address: Department of Biology, University of Victoria, Victoria, BC V8W 2Y2, Canada. E-mail: jetsam@uvic.ca

nerve roots extending from the collar cord; about 20 species have been described from the family. To date, developmental observations of the family Harrimaniidae have been restricted to *Saccoglossus* (Davis, 1908; Burdon-Jones, 1952; Colwin and Colwin, 1953). *Saccoglossus pusillus* (Davis, 1908) is the only harrimaniid from the West Coast of North America for which developmental information exists.

Ricketts *et al.* (1985; originally published in 1939) remains the most comprehensive review of the Enteropneusta from the Pacific Coast of North America. The present status of the Enteropneusta from this region is briefly summarized here. From the family Harrimaniidae, *Saccoglossus* (formerly *Dolichoglossus*) *pusillus* (Ritter) (in Davis, 1908; Horst, 1930) is described from San Pedro and San Diego, California. In an earlier work, Ritter (1902) indicates that this same animal is abundant in Puget Sound. Recently, the *Saccoglossus* from Puget Sound was described by King *et al.* (1994), who found it conspecific with *S. bromophenolosus* (formerly identified as *S. kowalevskii*) from the Northeast Coast of the United States. Two other apparently undescribed species of *Saccoglossus* occur on the Pacific Coast; one is common subtidally in Barkley Sound, British Columbia (Cameron and Mackie, 1996; Cameron *et al.*, 1999), and the other is from the Santa Maria Basin (Woodwick, 1996). Molecular tools should help us to distinguish the morphologically similar species of this North American genus. *Stereobalanus* sp. has been discovered from the Santa Maria Basin (Woodwick, 1996), and *Harrimania maculosa* (Ritter, 1900) is described from Alaska.

Two undescribed species of *Schizocardium* (family Spengelidae) have been found on the coast of California: the first is from Mugu Lagoon (Ricketts *et al.*, 1985) (now a military base) and Newport Bay (Ricketts *et al.*, 1985) (which now has a highly modified and heavily populated shoreline), and the second is from the Santa Maria Basin (Woodwick, 1996). From the family Ptychoderidae, Ritter (1902) mentioned, but did not describe, *Balanoglossus occidentalis* from San Pedro, California. Other species of *Balanoglossus* have been collected from the Santa Maria Basin (Woodwick, 1996) and from the northeast coastline of the Sea of Cortez (author's collection). *Glossobalanus berkeleyi* (Willey, 1931), originally described from Nanaimo, British Columbia, is no longer present in Nanaimo Bay, but is found in Puget Sound, Washington (author's records). Ptychoderidae is the morphologically most complex of the enteropneust families, and the life cycle of its species usually includes a tornaria larva. Two species of tornaria larvae have been described from western North America (see Ritter and Davis, 1904), but the corresponding adult worms are unknown.

The purpose of the study reported here was to describe *Harrimania planktophilus*, a newly discovered member of the Harrimaniidae from the West Coast of North America.

The description includes—in addition to details of habitat and morphology—observations on the mode of development of *H. planktophilus*. Features of this development support the recent hypothesis (Cameron *et al.*, 2000) that the Harrimaniid worms are sister taxa to the colonial class Pterobranchia; the evidence also suggests that these taxa evolved from a worm-like ancestor with indirect development. See the appendix for a key to the families and genera of the Enteropneusta and the species of the genus *Harrimania*.

Materials and Methods

Harrimania planktophilus (Figs. 1 through 5) was collected from the intertidal sand flat at Cape Beale (48° 47' 30" N, 125° 12' 56" W) in Barkley Sound, Vancouver Island, Canada. Six worms were collected during February 1997, and another twenty-one were collected over the summer of 1999. All worms were transported to the Bamfield Marine Station, where they were kept in specimen bowls containing their natural sediment under a flow of fresh seawater. One or two worms at a time were removed from their bowls for histology. These animals were allowed to defecate their gut contents, and the external surface was cleaned of sediment. The cleaned worms were relaxed in 7% MgCl₂, then fixed in Bouin's solution, and dehydrated in a graded series of ethanol. Once in 100% ethanol, the animals were dissected into small pieces, transferred to xylene, and infiltrated with paraffin wax. Sections were cut on an American Optical Corporation 820 Spencer microtome, stained with either Delafield's hematoxylin or eriochrome-cyanin, and viewed and photographed with an Olympus OM-4T 35-mm camera mounted on an Olympus BH2 compound microscope.

I was unable to study unfertilized eggs, fertilization, and early cleavage stages preceding the late blastula stage. Blastulae were obtained on 19 February 1997 from a single egg mass maintained in an aquarium; about 70 embryos were in this tubular mass. Twenty of these embryos were removed and reared in finger bowls containing fresh seawater that was changed daily, but mortality was high. Consequently, the remaining 50 embryos were allowed to develop in the jelly mass, in the sediment, but an individual was removed periodically to document development. The temperature of the seawater during the observation period varied between 10 °C and 11 °C. The various stages were measured, photographed, described, and drawn.

Taxonomic Treatment

Harrimania Ritter 1900

Harrimania Ritter 1900: 112.

TYPE SPECIES: *Harrimania maculosa* Ritter 1900: 112–115.

Harrimania planktophilus new species
Figs. 1, 2, 3, 4.



Figure 1. *Harrimania planktophilus*. Total length of a relaxed and uncoiled animal is approximately 32 mm. c, collar; gp, gill pores; m, mouth; p, proboscis; t, trunk.

Description

External

Harrimania planktophilus is, in general, muscular and active. It is robust and can be collected intact, unlike many other species of enteropneust, which are fragile and easily broken. *H. planktophilus* (Fig. 1) is small compared with other enteropneusts. The longest individual was 6 cm in length when extended in the bottom of a specimen bowl, and the average specimen was approximately 3.5 cm. The proboscis is conical, is longer than broad, and about twice as long as the collar (Fig. 1, 2). It bears a middorsal groove that extends two-thirds the length of the proboscis from the posterior margin. In some animals, the proboscis skeleton is pigmented black and can easily be seen through the proboscis epithelium. The collar is shorter than it is wide, with a distinctive circumbuccal groove about halfway between anterior lip and posterior marginal ridge. The posterior marginal ridge of the collar is accompanied by a fine crease. Anteriorly the collar lip (see fig. 4, in Cameron, 2002) is muscular and contractile, altering the circumference and shape of the mouth. An epidermal pit is always present at the dorsal midline where the collar meets the trunk, and its perimeter is rust colored. The trunk can be separated into four regions: a long anterior branchial region; an esophageal region with a reddish esophageal organ; a short transparent

region, followed by a darkly pigmented hepatic region (there are no liver sacs); and an intestinal region that has a terminal anus with a sphincter muscle. In gravid males and females the gonads are a pair of large, irregular masses, located dorsolaterally and extending from the posterior pharyngeal region to the anus. The pharyngeal region has many ($36.2 \pm \text{SD } 9.7$; $n = 6$) pairs of large, muscular gill pores (Figs. 1, 5) that open to the outside in paired dorsolateral grooves. A strong exhalent water current was observed in animals acclimated to cold (5–7 °C) seawater. Under these conditions, the pores are expanded, and through the pores can be seen the tongue bars in the wall of the pharynx. In about 1 of 10 animals, the skeletal bars are black, rather than the more common collagenous opaque white. The pharyngeal region is 5 times the length of the extended proboscis. The esophageal region has a large bilobed muscular organ that is pigmented dark red, and posterior to this is a darkly stained hepatic region, and then a meandering gut that can be seen through the translucent body wall. The following measurements are from an average adult living specimen: length of proboscis, 2 mm; length of collar, 1 mm; length of pharynx, 9 mm; length of esophagus to anus, 20 mm; total, 32 mm.

Color (Ridgway, 1912): cream-colored proboscis (pl XVI, f-19); capucine orange collar (pl III, d-13); pale yellow-orange trunk (pl III, f-15), except for the esophageal

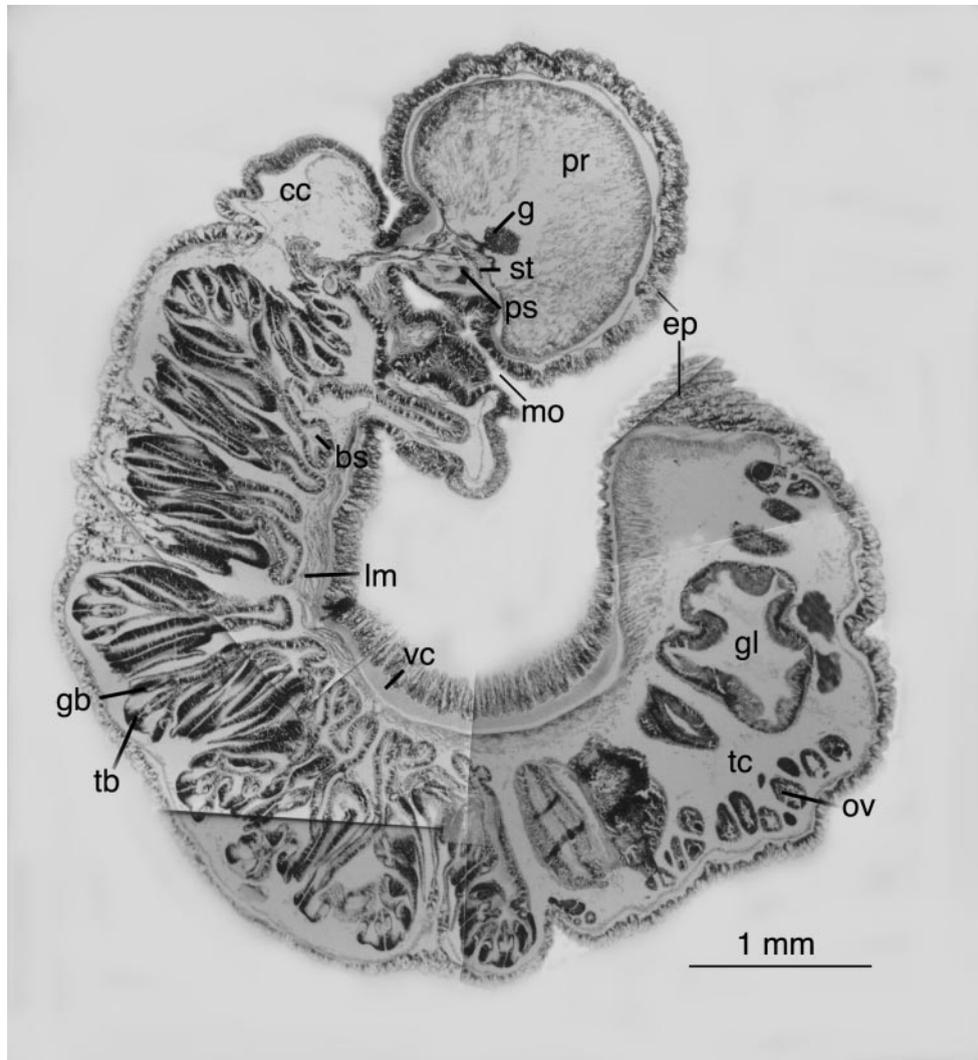


Figure 2. Light micrograph of a longitudinal section of *Harrimania planktophilus*. bs, branchial sac (= atrium); cc, collar coelom; ep, epithelium; g, glomerulus; gb, primary or gill bar; gl, gut lumen; lm, longitudinal muscles; mo, mouth; ov, ovaries; pr, proboscis; ps, proboscis skeleton; st, stomochord; tb secondary or tongue bar; tc, trunk coelom (or metacoel); vc, ventral cord.

organ, which is brick red (pl XIII, k-5), and the hepatic region, which is brownish olive (pl XXX, m-19). The eggs are amber yellow (pl XVI, b-21), and the male gonads are slightly paler.

Anatomical and histological characters

Proboscis. The epithelium of the proboscis is simple and columnar, with multiciliated cells and with glandular cells having basal nuclei. The nervous layer of the ectoderm is about equal in thickness to the underlying circular muscle layer, and thickening slightly under the dorsal proboscis groove. The longitudinal muscle fibers, in transverse section, are arranged in radiating plates, as in *Ptychodera* and *Stereobalanus* (Woodwick, 1996); there are fewer than 50

plates, with spaces between them, and they narrow near the center of the proboscis where the coelom is small or absent (Fig. 3A). There is no ventral proboscis mesentery; instead, a plate of connective tissue underlies the ventral stomochord and extends ventrally, nearly halfway to the epidermis, wedging itself between two radial muscle plates. This connective tissue is contiguous with the posterior portion of the proboscis stomochord (Fig. 3G) and becomes continuous with the proboscis skeleton posteriorly. The stomochord, which lacks the vermiform process of *Schizocardium* (Horst, 1939), is composed of columnar cells arranged radially around a central ciliated lumen, each cell with a large vacuole distally and a proximal nucleus. The ciliated lumen opens into the dorsal midline of the buccal collar, just

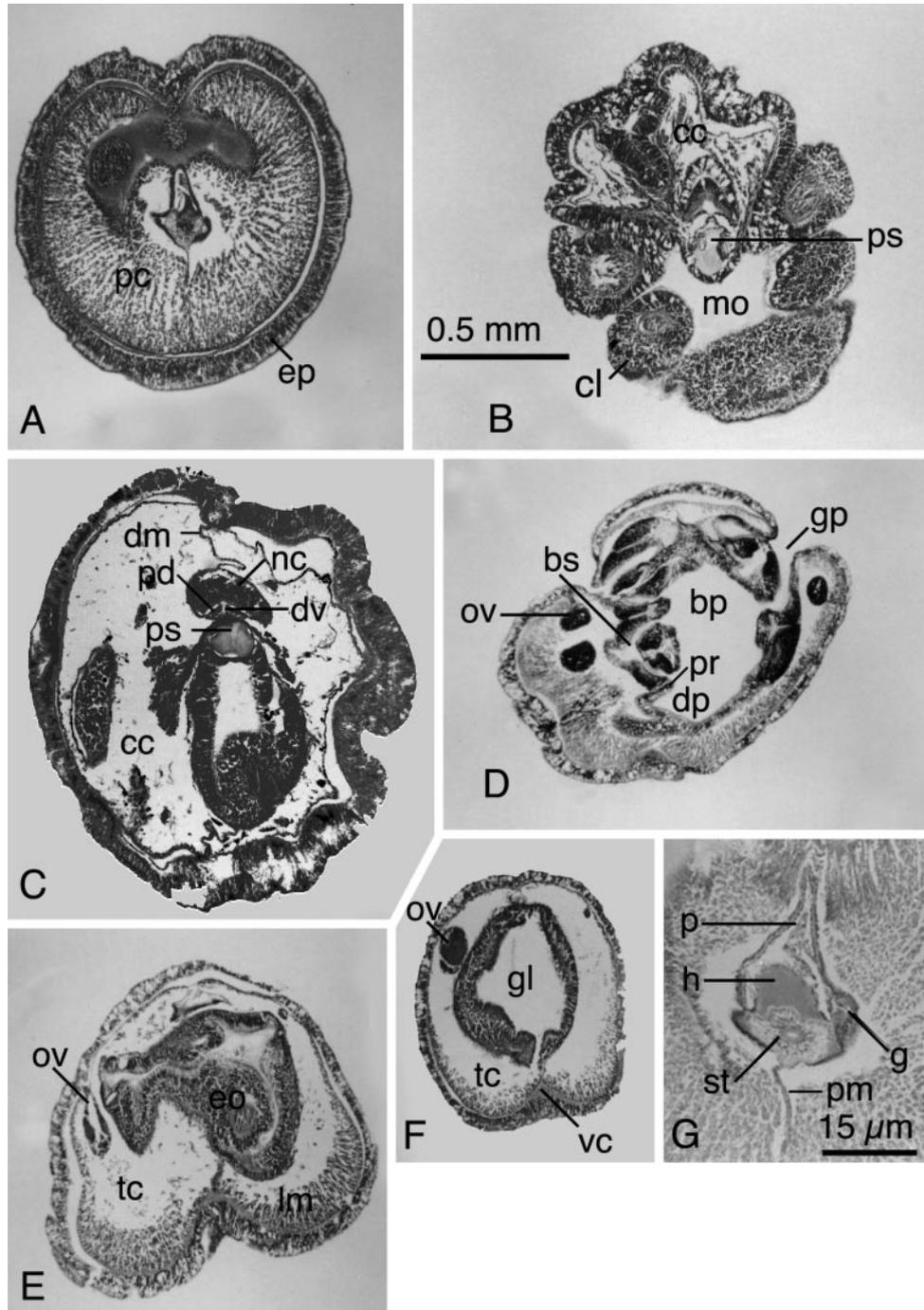


Figure 3. Light micrographs of transverse sections of *Harrimania planktophilus*. (A) Proboscis. (B) Proboscis stalk and anterior collar lip. (C) Collar. (D) Pharyngeal region of the trunk. (E) Esophageal region. (F) Intestinal region. (G) The stomochord is flanked by the heart-kidney coelomic complex, and has a central lumen. bp, branchial pharynx; bs, branchial sac (= atrium); cc, collar coelom; cl, collar lip; dm, dorsal mesentery; dp, digestive pharynx; dv, dorsal vessel; eo, esophageal organ; ep, epithelium; g, glomerulus; gl, gut lumen; gp, gill pore; h, heart; lm, longitudinal muscles; mo, mouth; nc, nerve cord; ov, ovaries; p, pericardial sac; pc, proboscis coelom (protocoel); pd, perihaemal diverticula; pm, proboscis mesentery; pr, parabranchial ridge; ps, proboscis skeleton; st, stomochord; tc, trunk coelom (metacoel); vc, ventral cord.

posterior to the position where the two proboscis skeleton crura bifurcate from the skeletal body. An extensive U-shaped glomerulus is situated on the wall of the heart and at the anterior tip of the stomochord. The pericardium, in transverse section, is a small wedge-shaped sac that extends dorsally from the heart, nearly halfway to the epidermis, between two longitudinal muscle plates (Fig. 3G).

The proboscis coelom is small to absent, is never divided completely, and extends posteriorly on the ventral and dorsal sides of the stomochord. Ventrally the coelom ends in a blind sac; dorsally it extends extremely posteriorly into the stalk and opens to the outside through a single proboscis pore just left of the dorsal midline of the proboscis stalk and almost into the anterior neuropore.

Collar. In some specimens, an anterior neuropore is located at the midline of the proboscis stalk where it meets the collar. A posterior neuropore is always present at the dorsal midline where the collar meets the trunk. Over the length of the collar nerve cord, one or two small lacunae may reside lateral to the midline. Giant nerve cells are absent. A dorsal mesentery is present throughout the length of the collar cord (Fig. 3C). Ventral to the collar cord, the perihæmal coelomic diverticula are well developed and extend from the trunk coelom for two-thirds the length of the collar; the mesentery between the diverticula is well defined. Anteriorly the diverticula become confluent, reduced in size, and enveloped in collagenous tissue that is continuous with the proboscis skeleton. In the collar, the dorsal blood vessel lies in the mesentery formed by the adjacent medial walls of the perihæmal diverticula (Fig. 3C). The collar coelom extends far anteriorly into the proboscis neck (Fig. 3B). Peribuccal spaces are apparently absent. The body of the proboscis skeleton (Fig. 3B) reaches beyond the middle of the collar before bifurcating into two crura that extend into the pharyngeal region. The stomochord consists of two distinct parts. The anterior pouch-shaped part has a central ciliated lumen that opens into the buccal cavity (Fig. 3G), as it does in all Hemichordata. The anterior part is continuous with a posterior gutter-shaped part, which projects posteriorly from the pore of the stomochord lumen, farther along the roof of the buccal cavity, before ending as two open troughs. The stomochord of *H. planktophilus* is thus similar in design to that of *Harrimania maculosa* (Ritter, 1900), but not as extensive.

Trunk. Poorly developed parabranchial ridges (Fig. 3D), formed by the ventral edge of the gill bars projecting into the pharynx (see fig. 2 in Cameron, 2002), separate the dorsal branchial pharynx from the ventral digestive pharynx (Fig. 3D). The gill slit skeletal bars are commonly opaque, but in 1 of every 10 animals, they are pigmented black. Peripharyngeal diverticula extend into the tongue bars only. No synapticles join the gill bars with the tongue bars (see figs. 1, 2 in Cameron, 2002). The atrial canal (branchial sac) (Fig. 3D, and see fig. 2 in Cameron, 2002) is U-shaped,

heavily ciliated, and leads from the pharyngeal gill slits to the outside *via* the ciliated gill pore. An epibranchial ridge is absent. *Harrimania planktophilus* is dioecious. Gonads, which are restricted to the posterior two-thirds of the trunk perivisceral coelom, begin in the pharyngeal region and extend into the intestinal region (Figs. 2, 3D). Gonadal pores occur dorsolaterally. Immediately behind the pharynx, the esophageal epithelium thickens laterally (Fig. 3E) and forms two opposing lobes, called here the esophageal organ; the lumen is correspondingly reduced in size, as compared with that of the intestine. The intestine meanders *en route* to the anus; it is lined with a simple epithelium, and hepatic sacs are absent. Two longitudinal muscles are broad and well-developed ventrolaterally, and narrow dorsally (Fig. 3E). Circular muscle is absent in this part of the body. The subepidermal nerve layer is thickened on the dorsal midline and even more so on the ventral midline (Fig. 3F). Thus, the midventral cord may be the main conductive channel in the trunk, as it is in a sympatrically occurring and apparently undescribed *Saccoglossus* sp. (Cameron and Mackie, 1996).

Diagnosis

The adult animal is distinguished by having a single proboscis pore, a glomerulus that extends frontally over the stomochord (Fig. 3G), and a proboscis skeleton that extends into the pharyngeal trunk. The stomochord begins in the buccal collar as a middorsal ridge; the ridge is maintained on the lateral sides by the proboscis skeletal crura. The opening of the stomochord canal is positioned just posterior to where the two crura meet to form the skeletal body. The collar has a complete dorsal mesentery (Fig. 3C). Animals may have as many as 54 pairs of gill pores. Gonads overlap with only a few of the most posterior gill pores, and the eggs are pigmented yellow.

Development

Embryos were found in aquaria at early blastula to mid-blastula stage and, therefore, information about gamete structure, egg maturation, events of fertilization, early cleavage stages, and blastulation is lacking. The coeloblastula (Fig. 4A) is 75 μm in diameter and heavily pigmented lemon yellow. It develops in a fertilization membrane 83 μm across that has a thin (1.5 μm) sticky jelly layer, thus development of the animal was often obscured by debris and sand adhering to the jelly coat. Gastrulation appears to proceed by invagination of the posterior (vegetal) pole, resulting in a hemispherical shape that persists for about 12 h (Fig. 4B). The wide archenteron greatly reduces the blastocoel. The blastopore begins to constrict, changing the shape of the gastrula, once again into a sphere (Fig. 4C). Over a period of 24 h the blastopore shrinks, finally persisting as a teardrop shape. There are no external cilia.

The development of cilia initiated rotation within the

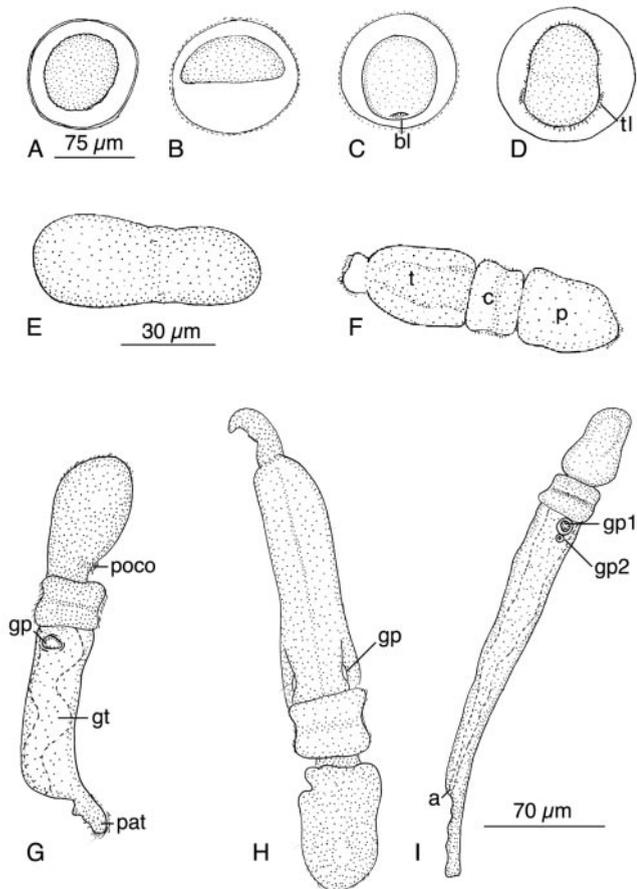


Figure 4. Diagram of the later development of *Harrimania planktophilus*. (A) Blastula. (B) Gastrulation showing obliteration of the blastocoel. (C) Unciliated gastrula. (D) Ciliated embryo with a diffuse ciliated telotrochal band. (E) Newly hatched juvenile. (F) Juvenile with distinct body regions. (G) Juvenile at the first gill pore stage, lateral view. (H) Juvenile at the first gill pore stage, dorsal view. (I) Juvenile at the second gill pore stage, lateral view. a, anus; bl, blastopore; c, collar; gp, gill pore; gp1, first gill pore; gp2, second gill pore; gt, gut; p, proboscis; pat, post-anal tail; poco, pre-oral ciliary organ; t, trunk; tl, telotroch.

embryonic chorion 42 h after the embryos were discovered (Fig. 4D). In addition to rotating, the “larvae” propel themselves in the apical direction. A dimple is all that remains of the closed blastopore. The spherical larvae are 72 μm in diameter. A wide telotrochal band forms on the otherwise uniformly ciliated ball; there is no ciliated apical tuft. Elongation by the constriction of a midventral groove is followed by the hatching of a 90- μm -long, benthic pre-juvenile stage (Fig. 4E). As with the rest of the development, hatching is asynchronous. Of 20 individuals that were extracted from the burrow egg mass and reared in culture, only a single animal hatched. Of about 50 embryos left in the burrow, 14 survived development into juveniles. The fate of the remaining eggs is uncertain, but predation cannot be ruled out because sympatrically occurring species were

present in the sediment. Often the jelly layer is shed shortly before the embryo hatches from the transparent chorion.

Elongation and differentiation of the body into a distinct proboscis, collar, and trunk was complete by day 8 for the remaining few animals in culture (Fig. 4F). The lengths of these body regions were 55.0 μm , 18.5 μm , and 75.0 μm , respectively. The animal could locomote on a plastic dish one body length every 2 s. Forward locomotion is guided by a ciliated muscular proboscis. Posterior locomotion is equally efficient and accomplished by a reversal in ciliary beat. Ciliation is most apparent on the proboscis tip, the collar, and the end of the tail. The pre-oral ciliary organ developed later, after about 12 days. The posterior ventral tail becomes post-anal with the completion of the alimentary canal. The tail extends from the ventral trunk and is heavily ciliated and glandular. The tail can adhere to sediments, plastic, and glass. Like the adults, the juveniles move away from light.

The first pair of gill clefts (perforating the pharyngeal epithelium) and gill pores (perforating the ectoderm) are large and appeared on day 15 (Fig. 4G). The circular clefts are lined with long cilia and lack any sign of a skeleton. When the juvenile is viewed perpendicular to its body axis, ciliary waves are seen to pass in a clockwise direction on the left side of the body and counterclockwise on the right side of the body. In other words, the metachronal ciliary beat, around the rim of each pair of clefts, is in a bilaterally symmetrical pattern. At day 17, the juveniles had a second pair of gill pores and a ventral post-anal tail (Fig. 4I). Eventually a gill skeleton develops around the perimeter of the gill cleft, elongating it into a dorsoventrally elongated gill slit (data not shown).

Biology

Harrimania planktophilus is an active infaunal burrower. Individuals were found in two locations in Barkley Sound, subtidally in the Ross Islets (49° 52' N, 125° 10' W) and in the low intertidal of a protected beach adjacent to the eastern slope of Cape Beale (48° 47' 30" N, 125° 12' 56" W). At Cape Beale, *H. planktophilus* was collected at the extreme low tides that occur during the morning hours in spring and summer, and during the evening in fall and winter (Canadian Tidal Tables). *H. planktophilus* was typically found in a mixture of biogenic calcium carbonate debris and sorted sands with a low concentration of organics. The carbonate fraction of the Cape Beale deposits in the lower tidal range typically contains more than 30% barnacle plates together with fragments of gastropods and, in lower abundance, echinoids, bryozoa, and foraminifera.

H. planktophilus creates sinuous interconnected tunnel systems that do not seem to approach the epibenthos. They are strongly photonegative and lack the odor of bromophenols. In aquaria, more than one animal would frequent a

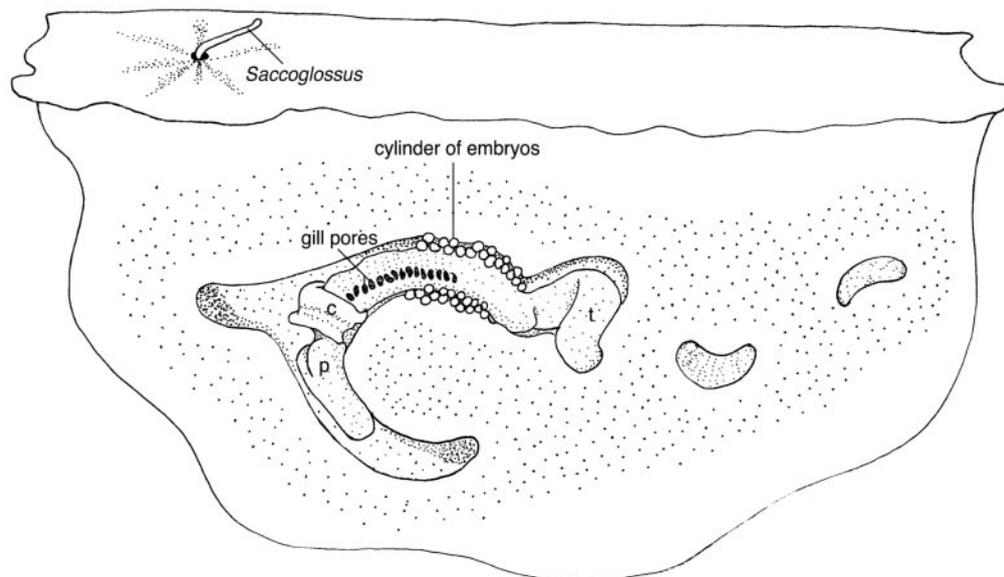


Figure 5. Diagram of *Harrimania planktophilus* in situ with a cylindrical mass of embryos. A tentatively undescribed species of enteropneust, *Saccoglossus* (a surface deposit feeder), is sympatric with *H. planktophilus* at Cape Beale. c, collar; p, proboscis; t, trunk.

single burrow system. Embryos are deposited in the burrow; the egg mass is cylindrical and has a hollow center, through which adult worms and water currents may pass (Fig. 5). Embryos stick to each other with a thin, sticky jelly coat. The face of the embryonic chorion directed towards the lumen of the burrow is clean, whereas the face directed towards the burrow wall is coated with sediment and detritus.

H. planktophilus ingests sediments that it traps in mucus on its proboscis (Fig. 5) and transports back to the mouth with cilia. It also feeds on organic matter in interstitial pore water, employing its pharyngeal cilia to propel water into its mouth and out of its gill pores (Cameron, 2002). Although debris acquired from pore water may constitute a small amount of the total gut content, it may have a significant nutritional role.

Etymology

The species name is Latin and means “plankton loving,” reflecting the animal’s ability to suspension feed on plankton in interstitial pore water.

Holotype

Adult female: British Columbia, Vancouver Island, Barkley Sound, Cape Beale protected beach, north side adjacent to the “gap,” 48° 47′ 30″ N, 125° 12′ 56″ W, 20 September 1999, C. B. Cameron.

Sectioned specimen deposited at the Royal British Columbia Museum (RBCM); holotype number 001-00109-001.

Paratypes

Two specimens of undetermined sex deposited at the RBCM; paratype number 001-00109-002.

Evidence the Genus *Harrimania* is Monophyletic

There are three families of Enteropneusta—four if *Saxipendium coronatum*, the deep-sea hydrothermal vent enteropneust (Woodwick and Sensenbaugh, 1985), constitutes a family of its own. *Harrimania planktophilus* belongs to the family Harrimaniidae, of which there are about 20 described species from the genera *Saccoglossus*, *Protoglossus*, *Stereobalanus*, *Xenopleura*, and *Harrimania*. Harrimaniids have no liver sacs, no synapticles joining the primary and secondary gill bars, and no nerve roots in the collar cord mesentery. *Harrimania* is distinguished from *Saccoglossus* by having a short proboscis (Figs. 1, 2), and from *Xenopleura* by having large pharyngeal pores (Fig. 1). *Harrimania* is distinguished from *Protoglossus* by having a more developed gill skeleton. *Harrimania* is distinguished from *Stereobalanus* by lacking a pair of genital wings and gill pores fused to a common dorsal slit (Ritter, 1900).

Two other species of *Harrimania* are known: *H. maculosa* (Ritter, 1900) and *H. kupfferi* (Spengel, 1901, in Horst, 1939); both have, like *Stereobalanus*, paired proboscis pores. *H. maculosa* is commonly found intertidally in Alaska at Kodiak, Prince William Sound, Orca, and Valdez. This thick, dark brown acorn worm is about 12 cm long and emits a strong bromophenol odor (Ritter, 1900). It does not burrow as do most enteropneusts, but lies under stones after

the fashion of some holothurians. In all of these respects *H. maculosa* is unlike the species described here, *H. planktophilus*. Perhaps the most distinctive feature of *H. maculosa* is its two-part stomochord. *H. maculosa* always has two proboscis pores, and has an epibranchial ridge (Ritter, 1900).

H. kupfferi is from Scandinavia (Öresund near Hellebaek, Kattegatt, east of Laessö Island) and Greenland (Horst, 1939). *H. kupfferi* is 8–9 cm long, with a pale-colored proboscis and collar, reddish gonads, and a brown trunk. It differs from *H. planktophilus* in that its glomerulus halves do not connect frontally. The crura of the proboscis skeleton are not as extensive, extending to the posterior end of the collar, whereas in *H. maculosa* and *H. planktophilus* they extend into the trunk. *H. kupfferi* has no mesentery separating the dorsal and ventral collar coelomic cavities, and the dorsal pharynx, with up to 40 gill pores, is smaller than the ventral pharynx (Horst, 1939). In contrast, the dorsal pharynx of *H. planktophilus* is larger than the ventral pharynx (Fig. 3D) and has as many as 54 pairs of gill pores.

Phylogenetic Relationships

The presence of a telotrochal swimming band (Fig. 4D) during the development of *Harrimania planktophilus* suggests that the harrimaniids had an ancestor that developed indirectly. Within the family Harrimaniidae, *Harrimania* is a sister taxon to *Stereobalanus* (Reinhard, 1942) a conclusion based on the shared presence of paired protoel ducts and pores. Molecular sequence data suggest that the harrimaniid worms are more closely related to the colonial class Pterobranchia (including the genera *Rhabdopleura* and *Cephalodiscus*), which also have paired protoel ducts and pores, than they are to the ptychoderid worms (*Balanoglossus* and *Ptychodera*) (Cameron *et al.*, 2000). The evolution of the pterobranchs from a harrimaniid ancestor may have involved a reduction in body size and a switch to coloniality. Harrimaniids and pterobranchs, when compared to ptychoderid enteropneusts, show a reduction and disappearance of coelomic diverticula, gonads, and gill slits. The pterobranchs show the most extreme reduction in the size and complexity of these structures. The ventral post-anal tail of juvenile *H. planktophilus* (Fig. 4G–I) may be homologous to the stalk that joins pterobranch zooids. The phylogenetic position (Cameron *et al.*, 2000) and ontogeny of *H. planktophilus* support the idea that the harrimaniid worms are intermediate in form between a complex and indirectly developing worm-like ancestral body plan, and the simple and directly developing body plan of the colonial class Pterobranchia. See the appendix for a key to the families and genera of the Enteropneusta and the species of the genus *Harrimania*.

Acknowledgments

Dr. A. R. Palmer provided support (NSERC Operating Grant A7245 [A.R.P.]) for this project. I gratefully ac-

knowledge Dr. A. Martel for providing his microscope, Dr. R. Marx for help with translating the taxonomy text for *H. kupfferi*, and the staff of the Bamfield Marine Station for providing facilities.

Literature Cited

- Burdon-Jones, C. 1952.** Development and biology of the larvae of *Saccoglossus horsti* (Enteropneusta). *Proc. R. Soc. Lond. B. Biol. Sci.* **236**: 553–589.
- Cameron, C. B. 2002.** Particle retention and flow in the pharynx of the enteropneust worm *Harrimania planktophilus*: the filter-feeding pharynx may have evolved before the chordates. *Biol. Bull.* **202**: 192–200.
- Cameron, C. B., and G. O. Mackie. 1996.** Conduction pathways in the nervous system of *Saccoglossus* sp. (Enteropneusta). *Can. J. Zool.* **74**: 15–19.
- Cameron, C. B., G. O. Mackie, J. F. F. Powell, and N. M. Sherwood. 1999.** Gonadotropin-releasing hormone in mulberry cells of *Saccoglossus* (Hemichordata: Enteropneusta). *Gen. Comp. Endocrinol.* **114**: 2–10.
- Cameron, C. B., J. R. Garey, and B. J. Swalla. 2000.** Evolution of the chordate body plan: new insights from phylogenetic analyses of deuterostome phyla. *Proc. Natl. Acad. Sci. USA* **97**: 4469–4474.
- Caulley, M., and F. Mesnil. 1904.** Contribution à l'étude des Entéropeustes. *Protobalanus (n.g.) koehleri*. *Zool. Jahrb. Anat.* **20**: 227.
- Colwin, A. L., and L. H. Colwin. 1953.** The normal embryology of *Saccoglossus kowalevskii* (Enteropneusta). *J. Morphol.* **92**: 401–453.
- Davis, B. 1908.** The early life history of *Dolichoglossus pusillus*. *Univ. Calif. Publ. Zool.* **4**: 187–226.
- Delle Chiaje, S. 1829.** Memorie sulla storia e notomia degli animali senza vertebre del Regno di Neapel. *Napoli* **4**: 1–72.
- Eschscholtz, F. 1825.** Bericht über die zoologische Ausbeute der Reise von Kronstadt bis St. Peter und Paul. *Oken's Isis* **6**: 64–67.
- Franzén, Å., K. H. Woodwick, and T. Sensenbaugh. 1985.** Spermiogenesis and ultrastructure of spermatozoa in *Saxipendium coronatum* (Hemichordata, Enteropneusta), with consideration of their relation to reproduction and dispersal. *Zoomorphology (Berl.)* **105** (5): 302–307.
- Gilchrist, J. 1925.** *Xenopleura vivipara*, g. et sp. n. (Enteropneusta). *Q. J. Microsc. Sci.* **69**: 555–573.
- Horst, C. J. van der. 1930.** Observations of some Enteropneusta. Papers from Dr. Th. Mortensen's Pacific expedition, 1914–1916, II. *Vidensk. Medd. Dan. Naturhist. Foren.* **87**: 135–200.
- Horst, C. J. van der. 1939.** Hemichordata. *Kl. Ordn. Tierreichs* **4**: 1–737.
- Jeffries, R. P. S. 1986.** *The Ancestry of the Vertebrates*. British Museum, London.
- King, G. M., C. Giray, and I. Kornfield. 1994.** A new hemichordate, *Saccoglossus bromophenolus* (Hemichordata: Enteropneusta: Harrimaniidae), from North America. *Proc. Biol. Soc. Wash.* **107**: 383–390.
- Nielsen, C. 1994.** *Animal Evolution: Interrelationships of the Living Phyla*. Oxford University Press, Oxford. 467 pp.
- Punnett, R. C. 1903.** The Enteropneusta. *The Fauna and Geography of the Maldives and Laccadive Archipelagoes*, J. S. Gardner, ed. Vol. 2, part 2. Cambridge Univ. Press, Cambridge.
- Reinhard, E. G. 1942.** *Stereobalanus canadensis*, a little known Enteropneusta from the coast of Maine. *J. Wash. Acad. Sci.* **32**: 309–311.
- Ricketts, E. F., J. Calvin, J. W. Hedgpeth, and D. W. Phillips. 1985.** *Between Pacific Tides*. 5th ed. Stanford University Press, Stanford. 652 pp.
- Ridgway, R. 1912.** *Color Standards and Color Nomenclature*. Washington, DC. Published by the author.
- Ritter, W. E. 1900.** Papers from the Harriman Alaska Expedition. II. *Harrimania maculosa*, a new genus and species of Enteropneusta from Alaska, with special regard to the character of its notochord. *Proc. Wash. Acad. Sci.* **2**: 111–132.

- Ritter, W. E. 1902.** The movements of the Enteropneusta and the mechanisms by which they are accomplished. *Biol. Bull.* **3**: 255–261.
- Ritter, W. E., and B. M. Davis. 1904.** Studies on the ecology, morphology, and speciology of the young of some Enteropneusta of western North America. *Univ. Calif. Publ. Zool.* **1**: 171–210.
- Schimkewitsch, W. M. 1892.** *Über die Beziehungen zwischen den Enteropneusta und Acrania.* Verlag der St.-Petersburger Naturforschergesellschaft, St. Petersburg.
- Spengel, J. W. 1893.** Die Enteropneusten des Golfes von Neapel. *Fauna und Flora des Golfes von Neapel.* Monograph 18. Friedländer, Berlin.
- Spengel, J. W. 1901.** Die Benennung der Enteropneusten-Gattungen. *Zool. Jahrb. Abt. Syst.* **15**: 209–218.
- Willey, A. 1898.** On *Ptychodera flava* Eschscholtz. *Q. J. Microsc. Sci.* **40**: 7–9.
- Willey, A. 1931.** *Glossobalanus berkeleyi*, a new enteropneust from the West Coast. *Trans. R. Soc. Canada* **5**: 19–28.
- Woodwick, K. H. 1996.** Hemichordata: Enteropneusta. Pp. 251–259 in *Taxonomic Atlas of the Benthic Fauna of the Santa Maria Basin and the Western Santa Barbara Channel.* Vol. 14. J. A. Blake, P. H. Scott, and A. Lissner, eds. Santa Barbara Museum of Natural History, Santa Barbara, CA.
- Woodwick, K. H., and T. Sensenbaugh. 1985.** *Saxipendium coronatum*, new genus, new species (Hemichordata: Enteropneusta): the unusual spaghetti worms of the Galápagos Rift hydrothermal vents. *Proc. Biol. Soc. Wash.* **98**: 351–365.

Appendix

Keys to the families and genera of the Enteropneusta, and to the species of the genus Harrimania

- A. Stomochord with a vermiform process; pericardium and glomerulus with paired anterior diverticula more or less developed **Spengeliidae**
- (a) Liver sacs and synapticles present; gill slits almost equaling the pharynx in depth, so that the ventral, non-pharyngeal part of the pharynx is reduced to a mere groove; nerve roots absent; pericardial diverticula long *Schizocardium*, Spengel (1893)
- (b) Liver sacs absent; ventral part of pharynx well developed; pericardial diverticula short
- (i) Synapticles and nerve roots absent,
- (a) Peribuccal spaces *Willeyia*, Punnett (1903)
- (B) Without peribuccal spaces *Glandiceps*, Spengel (1893)
- (ii) Synapticles present; nerve roots present or absent; genital region with dermal pits *Spengelia*, Willey (1898)
- B. Stomochord with no vermiform process; pericardium simple; ventral part of pharynx large, and sometimes more or less separated from the branchial part
- (a) Liver sacs, synapticles, and nerve roots present **Ptychoderidae**
- (i) Genital wings well developed
- (a) Atrium opening by long slits. *Ptychodera*, Eschscholtz (1825)
- (B) Atrium opening by small pores *Balanoglossus*, Delle Chiaje (1829)
- (ii) Genital wings hardly developed *Glossobalanus*, Spengel (1893)
- (b) Liver sacs, synapticles, and nerve roots absent. **Harrimaniidae**
- (i) With many atria
- (a) Proboscis long; one proboscis pore
- (1) burrowing *Saccoglossus*, Schimkewitsch (1892)
- (2) non-burrowing *Saxipendium*, Woodwick & Sensenbaugh (1985)
- (B) Proboscis short
- (1) one proboscis pore; viviparous *Xenopleura*, Gilchrist (1925)
- (2) usually two proboscis pores
- (a) Two pairs of genital wings *Stereobalanus*, Spengel (1901)
- (b) No genital wings *Harrimania*, Ritter (1900)
- (I) Proboscis skeleton extending into trunk, with collar mesenteries
- Extensive branchial ridge and collar stomochord. *H. maculosa*, Ritter (1900)
- Reduced branchial ridge and reduced collar stomochord. *H. planktophilus*, Cameron
- (II) Proboscis skeleton not extending into trunk, without collar mesenteries *H. kupfferi*, Spengel (1901)
- (ii) Without atria *Protoglossus*, Caullery & Mesnil (1904)