The columellar muscle of prosobranch gastropods: morphological zonation and its functional implications

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Abstract. The arrangement of muscle and connective tissue fibers in the columellar muscle of three prosobranch gastropods, *Calliostoma euglyptum, Littorina littorea,* and *Ilyanassa obsoleta,* was analyzed. The columellar muscle in the three snails is a flat, sheet-like muscle that originates on the columella of the shell and inserts on the operculum. The muscle was found to comprise a densely packed three-dimensional array of muscular and connective tissue fibers, supporting the hypotheses of previous authors that it functions as a muscular hydrostat. The fiber arrangement is not uniform throughout the entire muscle; instead it includes three regions that grade into one another. Nearest its origin, the muscle consists primarily of longitudinal muscle fibers with few dorsoventral or transverse fibers, and no oblique fibers. The middle portion of the columellar muscle in the region posterior to the head consists of longitudinal, dorsoventral, and transverse fibers wrapped by two opposed layers of oblique fibers. The third region of the columellar muscle extends through the foot and includes longitudinal, transverse, and dorsoventral fibers with a layer of oblique fibers on the dorsal surface. A narrow band of spherical spaces divides the columellar muscle into dorsal and ventral halves in this region. The morphological zonation within the muscle suggests that different regions perform different functions. In addition, compared to a muscle that is circular in cross section, the elliptical cross-sectional shape may increase the muscular force necessary to twist the foot relative to the shell and may decrease the resistance to buckling.

Additional key words: muscular antagonism, muscular hydrostat, hydrostatic skeleton, Mollusca

One familiar aspect of most coiled snails is their ability to retract the soft parts into the shell when disturbed. In coiled gastropods, the muscle responsible for retraction, the columellar muscle, originates on the columella of the shell and extends into the foot, where it inserts on the operculum. Although the function of the columellar muscle in retraction is straightforward, the mechanisms of elongation of the columellar muscle and protraction of the body from the shell are less obvious.

Early studies of the anatomy of the gastropod foot and visceral mass noted that the longitudinal fibers of the columellar muscle were divided into bundles by muscle fibers oriented perpendicular to the axis of the muscle (Küttler 1913; Weber 1926; Rotarides 1941). In a study of locomotion of *Bullia digitalis,* Trueman & Brown (1976) observed a similar three-dimensional array of muscle fibers in the columellar muscle and hypothesized that such an arrangement could provide a mechanism for muscular antagonism. Later, Brown & Trueman (1982) briefly surveyed metapodia from five prosobranchs and two pulmonates and found a three-dimensional network of muscle fibers in the columellar muscles of each. Finally, Voltzow (1990) described a three-dimensional array of muscle fibers in the columellar muscle in the foot of *Busycon contrarium* and in the pedal retractor muscle of *Haliotis kamtschatkana.*

A tightly packed three-dimensional array of muscle fibers has been observed in a variety of organs including the arms, tentacles, and mantle of cephalopod molluscs, the tongue of many vertebrates, the trunk of elephants, and in other gastropod organs such as the tentacles (see Kier & Smith 1985; Kier 1988; Smith & Kier 1989; Marshall et al. 1989). In such a system, called a muscular hydrostat, the contraction of the muscle fibers arranged in one orientation can antagonize the others. The mechanical principle is straightforward and relies on the fact that these structures con-

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sist of incompressible tissue and liquid. Because the structure is constant in volume, decrease in one dimension caused by shortening of fibers with a given orientation must result in increase in another dimension and elongation of the fibers running parallel to that dimension.

Contraction of the muscle fibers perpendicular to the long axis will decrease the cross section and increase the length of the columellar muscle, pushing the snail's body out of the shell or elevating the shell above the foot (Trueman & Brown 1976; Brown & Trueman 1982; Kier 1988; Voltzow 1990). Contraction of the longitudinal muscle fibers coupled with relaxation of muscle fibers perpendicular to the long axis will tend to shorten the columellar muscle, generating force for retraction or clamping of the shell to the substratum. Contraction of obliquely arranged muscle layers generates force for twisting of the columellar muscle, for instance when the shell is twisted relative to the foot during locomotion. The direction of twisting depends on the handedness of the contracting oblique muscle layer. Simultaneous contraction of opposite handed oblique muscle layers increases the torsional resistance of the columellar muscle (see Kier & Smith 1985).

Activity of antagonistic muscle fibers or application of force by a muscular hydrostat causes the pressure to increase. Indeed, Trueman & Brown (1985) presented experimental evidence in support of this mechanism in the abalone Haliotis midae, which has a three-dimensional arrangement of muscle fibers in the pedal (or shell) retractor muscle. They showed that shell elevation, analogous to protraction of the foot out of the shell in a more coiled gastropod, is accompanied by an increase in pressure in the pedal retractor muscle (Trueman & Brown 1985). Increases in intramuscular pressure in the foot associated with lifting, twisting, and retraction of the shell were also reported by Voltzow (1986) for Busycon contrarium and Haliotis kamtschatkana.

Although the previous studies cited above provide an overview of the anatomy and function of the columellar muscle (primarily in the portion of the columellar muscle located in the foot), a detailed description of the arrangement of the muscle and connective tissue fibers of the columellar muscle over the entire length from its origin on the columella of the shell to its insertion in the foot is lacking. (For a description of the ultrastructure of the muscle fibers see Huddart et al. 1977; Plesch 1977; Frescura & Hodgson 1989, 1990, 1992; Trueman & Hodgson 1990). In this study, we provide such a description for three species representing three grades of prosobranch gastropod evolution and show a previously undescribed zonation of the muscle along its length. We also discuss the func-

tional implications of this morphological specialization.

Methods

Animals

Specimens of Calliostoma euglyptum A. ADAMS 1854, Littorina irrorata (SAY 1822), and Ilyanassa obsoleta (SAY 1822) were obtained from Cape Fear Biological Supply (Southport, NC, USA). Additional specimens of L. irrorata and I. obsoleta were collected at Beaufort, North Carolina, USA. These species were chosen for the study as representatives of three grades of evolution in prosobranch gastropods: an archaeogastropod, C. euglyptum; a mesogastropod, L. irrorata; and a neogastropod, I. obsoleta.

The snails were maintained in artificial seawater (Instant Ocean, Aquarium Systems Inc, Ohio, USA) in aquaria for up to two months prior to fixation and sectioning.

Histology

A total of 18 snails (6 C. euglyptum, 4 L. irrorata, 8 I. obsoleta) were anesthetized in a 1:1 (v/v) mixture of seawater and 7.5% $MgCl_2 \cdot 6H_2O$ in distilled deionized water (Messenger et al. 1985) for 24 to 72 hours, and fixed in 10% formalin in seawater for 48 hours. The snails were treated overnight (DeCal, National Diagnostics, Atlanta, GA, USA) to decalcify the shells. The visceral masses of most specimens were then dissected away, leaving intact only the foot, columellar muscle, and, in the case of I. obsoleta, the muscle that retracts the siphon. The visceral masses of a few snails were left intact so that internal organs could be used for reference to determine orientation of the columellar muscle.

The fixed tissue was dehydrated in a graded series of ethyl alcohol, cleared (Histo-Clear, National Diagnostics, Atlanta, GA, USA), and embedded in Paraplast (Monoject Scientific, St. Louis, MO, USA). The tissue blocks were serially sectioned (10 μm thickness) in 3 mutually perpendicular planes with a rotary microtome. Alternate slides were stained with Milligan's trichrome and with picro-ponceau with Weigert iron hematoxylin according to procedures adapted from Humason (1979) by Kier (1992). Each staining procedure provides clear differentiation between collagenous connective tissue and muscle.

The serial sections were examined with brightfield, phase contrast, and polarized light microscopy, and drawings of representative sections were made using a camera lucida. Photomicrographs of individual sections were made with polarized light and brightfield illumination.

Movements and behavior

The three snails analyzed in this study, as well as *Busycon contrarium* and *Fasciolaria tulipa,* were observed in aquaria. Particular attention was paid to movements of the foot during locomotion, to twisting of the foot relative to the shell during locomotion and in righting movements, to shell elevation, and to protraction of the snail from its shell.

Theoretical calculations

To explore the mechanical consequences of the observed elliptical cross-sectional shape of the columellar muscle, we compared two simple geometrical shapes. The first, a cylinder of elliptic cross section, approximates the shape of the columellar muscle observed in this study. A major to minor axis ratio of 4:1 was chosen for the elliptical cross section. The actual range of major axis to minor axis ratios in the columellar muscles we measured was from 4:1 to 10:1 (or higher). The ratio of 4:1 was selected as a conservative value for our calculations. The second shape was a right circular cylinder, which approximates the shape of the pedal retractor muscle of monoplacophorans and abalones. For the calculations, the cross-sectional area, length, and mechanical properties of the material of each were identical.

Two aspects of the mechanical properties of the shapes were compared. First, we calculated the net torsional moment that could be generated by an oblique muscle layer wrapping each model. We assumed that a torsional force was applied tangential to the surface of each model and the force was integrated around the shape. The force must be integrated since, for the elliptical model, the torsional moment depends on the distance from the central longitudinal axis (e.g., the torsional moment is larger at the major vertex than at the minor vertex) (see Wainwright et al. 1976). To make the results of the calculations size and force independent, the torsional moment values for the elliptical model were divided by the values for the circular model.

The second comparison was a calculation of the critical Euler buckling force, and it was made in order to evaluate the ability of the two shapes to support the weight of the shell as a compressive load (see Wainwright et al. 1976). The second moment of area for each cross-sectional shape was obtained from Wainwright et al. (1976). The results of these calculations were made size and force independent by dividing the values for the elliptical model by the values for the circular model.
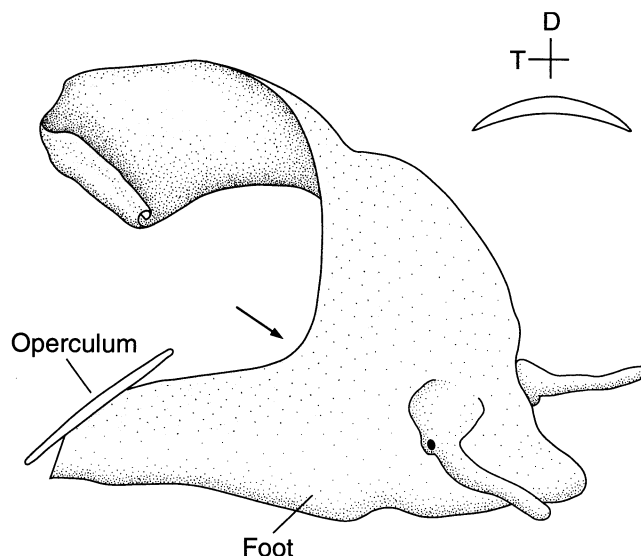

Fig. 1. A generalized schematic diagram of a snail showing the relation between foot and columellar muscle. The columellar muscle (shaded) turns ~½ whorls from origin to insertion. The 90° bend of the muscle is indicated by the arrow. Not shown is the small proportion of fibers that insert on the anterior ventral sole of the foot. The viscera are also excluded for clarity. In the upper right is a schematic diagram of a cross section through the columellar muscle. The orientations of dorsoventral (D) and transverse (T) muscle fibers are indicated.

Results

Movements and behavior

Except for *Littorina irrorata,* which spent most of the time immobile just above the water surface, the snails were capable of rapid shell elevation and twisting of the shell relative to the foot during locomotion. The widest range of twisting of the foot relative to the shell was noted when the snails attempted to right themselves after being flipped over. We estimate that snails were able to twist the foot as much as 50° relative to the shell. Most snails also rapidly cupped and flared the margins of the foot and twisted the foot relative to its long axis during attempts at righting. The anterior portion of the foot was not observed to be in contact with the aperture of the shell during protraction.

General morphology

The columellar muscle in all three species is a flat muscle that has its origin on the columella of the shell (Fig. 1). (We use the term "origin" as it is used in other descriptions of muscular anatomy to denote the site of attachment of the muscle that does not move, or moves less, than the moveable site of attachment

called the "insertion"). The muscle spirals ~1½ whorls from its origin on the shell to its insertion on the operculum. Upon entering the foot, it bends ~90° and extends posteriorly along the dorsal portion of the foot to insert on the operculum (Fig. 1). A small proportion of columellar muscle fibers branch from the main mass as it enters the foot and extend anteriorly to insert on the anterior sole of the foot. The interdigitation of muscle fibers in the foot with fibers from the columellar muscle, combined with the overall complexity of fiber orientations in this region, make it difficult to determine the precise points of insertion.

Before entering the foot, the columellar muscle is crescentic in cross section, with the convex side in contact with the columella of the shell. Excluding the portion that inserts on the anterior sole of the foot, the columellar muscle is oriented in contact with the columella; within the foot, the convex surface is located adjacent to the foot's dorsal surface. The columellar muscle is tapered slightly; the cross-sectional area of the muscle near its origin on the columella is less than the cross-sectional area of the muscle immediately dorsal to the foot. At its thickest point along the midline of the foot, the columellar muscle comprises ~30–40% of the dorsoventral thickness of the foot of a snail anesthetized and fixed as described above.

Microanatomy of the columellar muscle

Terminology. Choosing appropriate terminology for the various muscle and connective tissue fiber orientations in the columellar muscle and for the muscle itself is difficult because the muscle spirals, the axis of the helix changes, and the orientations of the axes of the helix are oblique to the body axes of the snail. To simplify the terminology, we define the orientations as they appear in the portion of the muscle that extends through the foot. We denote the muscle and connective tissue fibers that are parallel to the long axis of the columellar muscle as longitudinal. In addition to the longitudinal fibers, two categories of muscle and connective tissue fibers are oriented perpendicular to the long axis. The shortest perpendicular fibers extend from the convex surface of the columellar muscle to the concave surface and are referred to as dorsoventral (Fig. 1, D). The longer perpendicular fibers cross the dorsoventral fibers approximately at right angles and are termed transverse (Fig. 1, T). Note that the "dorsoventral" fibers are truly dorsoventral only in the portion of the columellar muscle located in the foot. In the description that follows, keep in mind that the convex surface of the columellar muscle is adjacent to the columella as the muscle spirals down from its origin

and is dorsal in the portion of the columellar muscle located in the foot.

The morphology of the columellar muscles of the species studied was similar and, unless noted, the morphological descriptions (and Figs. 1, 2, 4, 6) apply to all three species.

Morphology of the columellar muscle near its origin. Near its origin on the columella, the columellar muscle consists primarily of longitudinal muscle and connective tissue fibers (Figs. 2, 3). The connective tissue fibers and the endomysium surrounding individual muscle fibers are birefringent and exhibit staining reactions typical of collagen. Only a few scattered dorsoventral and transverse muscle or connective tissue fibers are present. A thin layer of connective tissue, ~2% of the thickness of the cross section of the columellar muscle, is present on the convex side of the muscle (Fig. 2). Most of the connective tissue fibers in this layer are longitudinally oriented, but there is a thin, overlying band of transverse connective tissue (Fig. 2). The layers of transverse and longitudinal connective tissue extend to the lateral margins of the columellar muscle and are thickest at the center of the cross section. A very thin layer of transverse connective tissue, ~1% of the thickness of the cross section of the columellar muscle, is present on the concave side of the muscle (Fig. 2).

Morphology of the columellar muscle immediately dorsal to the foot. The columellar muscle continues down into the mesopodium posterior to the head. In the region immediately dorsal to the foot, the core of the columellar muscle consists of longitudinal muscle fibers divided into bundles by regularly spaced sheets of dorsoventral muscle fibers (Figs. 4, 5a,b). This core of longitudinal and dorsoventral muscle is surrounded on both the convex and concave surfaces by a layer of transverse muscle fibers (Figs. 4, 5a). In addition, scattered transverse muscle fibers are observed within the core. In *Ilyanassa obsoleta*, a narrow band of transverse muscle fibers, ~2% of the thickness of the cross section of the columellar muscle, is also present in the core of longitudinal and dorsoventral fibers (Fig. 5a).

In sections perpendicular to the longitudinal muscle, scattered diagonal connective tissue and muscle fibers were observed to extend through the core at oblique angles to the dorsoventral muscle fibers (Fig. 5a). This three-dimensional muscular array is wrapped on the convex side by two oblique muscle layers (each roughly 5% of the thickness of the cross section), one wrapped as a left-hand helix and the other wrapped as a right-hand helix (Figs. 4, 5b). The oblique muscle layers include connective tissue fibers. In the region of the muscle immediately dorsal to the 90° bend in the

columellar muscle, the muscle is wrapped on all sides by the two oblique muscle layers. The fiber angle, the angle between the oblique fibers and the long axis of the columellar muscle, was between 45° and 50°. A thin layer (about 3% of the thickness of the cross section) of longitudinal muscle and connective tissue overlies the oblique muscles on the convex side.

The outermost layer on the convex side consists of a thin band of transverse muscle and connective tissue that is ~2% of the thickness of the cross section. A thin layer of transverse muscle and connective tissue fibers is also present on the concave side of the columellar muscle. This transverse layer comprises about 2% of the thickness of the cross section of the muscle.

In *I. obsoleta,* the muscle that retracts the siphon and the columellar muscle are contiguous in the region posterior to the head and just dorsal to the mesopodium. This narrow muscular connection consists of a core of longitudinal muscle fibers crossed by a few muscle fibers oriented perpendicular to the longitudinal fibers.

Morphology of the columellar muscle in the foot. The columellar muscle enters the dorsal portion of the mesopodium just posterior to the buccal mass and extends along the dorsal portion of the foot to insert on the operculum. The core of the columellar muscle consists primarily of longitudinal muscle fibers divided into bundles by regularly spaced sheets of dorsoventral fibers (Figs. 6, 7a,b). Transverse muscle fibers are present within a band of spherical spaces (see below) and at the dorsal and ventral margins of the columellar muscle (Figs. 6, 7a). The transverse fibers near the center of the muscle are not organized into distinct sheets like the dorsoventral fibers but extend in an irregular band through the center of the muscle, parallel to the frontal plane. Scattered diagonal connective tissue and muscle fibers extend through the core at oblique angles to the dorsoventral muscle fibers in transverse planes (Fig. 7a). Many of these diagonal fibers appear to branch and extend from the transverse muscle band described above.

The core of longitudinal and dorsoventral muscle is bordered on the ventral side by a thin layer of transverse muscle reinforced with connective tissue (Figs. 6, 7a), which constitutes about 2% of the thickness of the cross section of the muscle. On the dorsal side are two obliquely arranged layers of muscle fibers, one arranged as a right-hand and the other as a left-hand helix, located adjacent to the core of the muscle (Figs. 6, 7a). The fiber angle (measured relative to the longitudinal muscle fibers) of the oblique muscle layers is 45° to 50°. Each layer of oblique muscle makes up ~6% of the thickness of the cross section of the columellar muscle. Outside the oblique muscle layers is
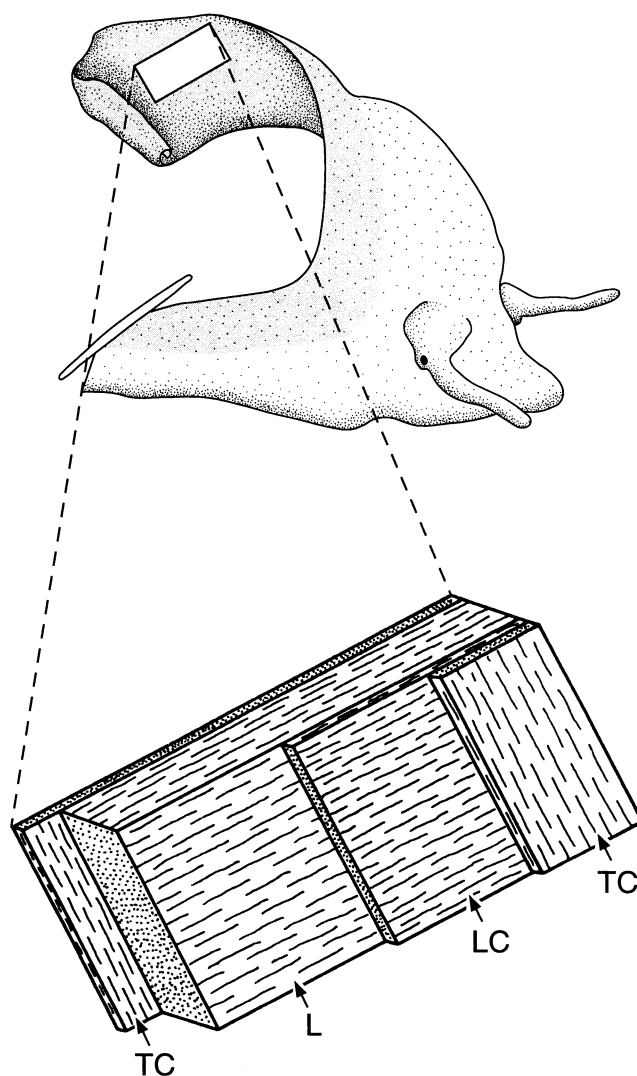

Fig. 2. Schematic diagram of the columellar muscle near its origin on the columella of the shell, showing the arrangement of muscle and connective tissue. The diagram is representative of all three species studied. The outline of the block diagram corresponds to the outline on the columellar muscle. The face of the block diagram closest to the viewer is the convex side of the muscle and is in contact with the columella of the shell. Most of the muscle fibers are oriented longitudinally (L). Longitudinal (LC) and transverse (TC) connective tissue fibers are present on the convex side of the muscle. A thin transverse band of connective tissue is present on the concave side of the muscle.

a thin layer of longitudinal muscle and connective tissue fibers (Fig. 6). Dorsal to the thin longitudinal layer is a thin layer of transverse muscle and connective tissue fibers (Fig. 6). The thin longitudinal and transverse muscle and connective tissue layers occupy roughly 3% and 2% of the thickness of the cross section of the muscle, respectively. Overlying the entire
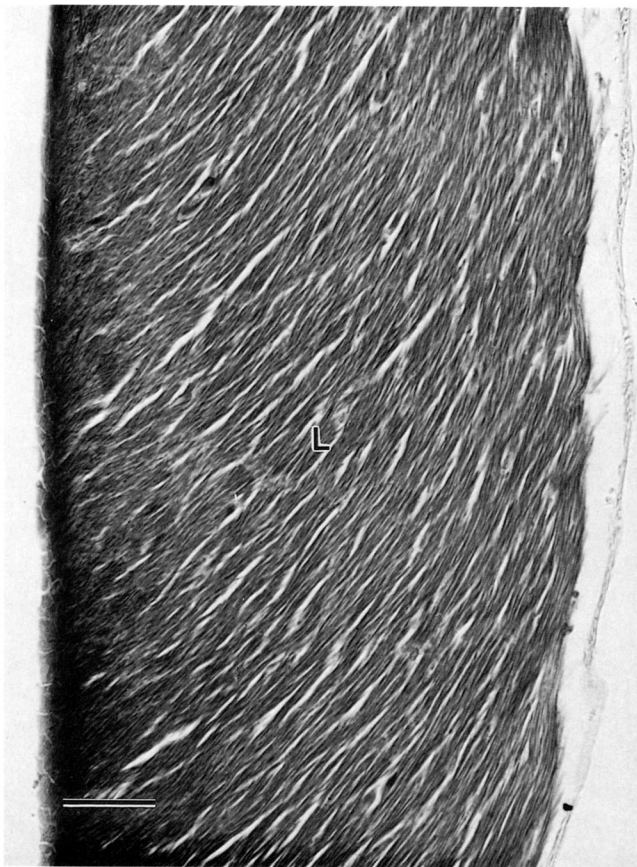
Fig. 3. Columellar muscle of *Calliostoma euglyptum* near its origin on the columella of the shell showing the core of longitudinal (L) muscle fibers characteristic of this region. Neither dorsoventral nor transverse muscle fibers are visible. The dark region at the left edge of the columellar muscle is a thin layer of connective tissue. Scale bar, 50 μm. Section orientation is slightly oblique to the longitudinal muscle fibers. Photomicrograph, brightfield illumination.

dorsal surface of the columellar muscle is a thin epidermis.

The columellar muscle, where it extends posteriorly from the 90° bend through the foot to the operculum, contains a distinct layer of spherical spaces (Figs. 6, 7a). Little cellular structure is visible in the lumen of the spaces and thus we tentatively identify them as hemocoelic spaces. Nonetheless, glandular tissue has been misidentified as hemocoelic spaces in previous studies of gastropod foot structure (see Grenon & Walker 1978, 1982). Definitive characterization of these spaces awaits appropriate histochemical tests (Grenon & Walker 1978). This layer of spherical spaces comprises 10–40% of the thickness of the cross section of the muscle. In addition, the number and size of the spherical spaces increase from the central region to the posterior region of the foot. Few longitudinal muscle fibers occur within the layer of spherical spac-

es, although both dorsoventral and transverse muscle fibers project through the layer (Figs. 6, 7a).

Near the operculum, the sponge-like layer of spherical spaces becomes less distinct, with fewer spherical spaces and more connective tissue fibers. The ordered arrangement of the columellar muscle also breaks down near the operculum. Although many connective tissue and muscle fibers remain oriented more or less longitudinally, others are oriented in a variety of directions in this region.

Summary of results

In summary, the fiber arrangement within the columellar muscle is not uniform throughout. Instead, the muscle includes three regions that grade into one another. The portion of the columellar muscle nearest its origin on the columella of the shell consists primarily of longitudinal muscle fibers. There are few dorsoventral or transverse fibers and no oblique muscles. The region of the columellar muscle posterior to the head, extending dorsally from the 90° bend in the muscle to the radular sac, consists of longitudinal, dorsoventral, and transverse fibers wrapped by two opposed layers of oblique muscles. In the foot, the columellar muscle consists of a three-dimensional muscular array, but oblique muscles are restricted to the dorsal surface. A narrow band of spherical spaces divides the columellar muscle into dorsal and ventral halves.

Discussion

The columellar muscle is not a single muscle; instead, it is an assemblage of muscle and connective tissue fibers that vary in orientation but that act in concert to effect complex, controlled movements. In this study, we consider those muscle and connective tissue fibers peripheral to the main mass of the columellar muscle (specifically, the oblique muscles and associated connective tissues of the body wall) to be part of that assemblage. The oblique muscle and connective tissue layers are integral to the functioning of the columellar muscle.

The columellar muscles of the three gastropod species studied are similar in morphology and include regions of tightly packed muscle fibers arranged in three mutually perpendicular orientations. This morphology is consistent with previous proposals that the columellar muscle functions as a muscular hydrostat (Trueman & Brown 1976; Brown & Trueman 1982; Kier 1988; Voltzow 1990).

Zonation of the columellar muscle

The muscular arrangement in the columellar muscle is not uniform from origin to insertion and instead in-

cludes three regions that grade into one another: (1) near the origin of the muscle on the columella of the shell; (2) in the region immediately dorsal and posterior to the head, extending ventrally to the foot; (3) in the foot. This zonation of morphology suggests that the three regions serve different functional roles, as proposed below.

In the region of the columellar muscle near its origin on the columella, the predominance of longitudinal fibers and the paucity of dorsoventral and transverse muscle fibers suggest that this region of the muscle functions primarily in retraction of the body of the snail into the shell and contributes little to protraction. The absence of oblique muscles indicates that this region does not function in twisting movements of the foot relative to the shell.

In the portion of the columellar muscle immediately dorsal and posterior to the head, and extending ventrally to the 90° bend just dorsal to the foot, the longitudinal muscle fibers are compartmentalized by regularly spaced bands of dorsoventral muscle fibers. The longitudinal fibers retract the snail into the shell. The presence of sheets of dorsoventral and transverse muscle fibers suggests an important role in protraction of the snail and in shell elevation during locomotion and burrowing (Trueman & Brown 1976). The oblique muscle layers most likely function primarily in twisting movements of the foot relative to the shell. Since both right- and left-handed layers are present, twisting in either direction can be generated and was observed. Gastropods can twist the foot relative to the shell through a large, semicircular arc. For example, Hazlett (1989) reported that *Trochus intexus* twists 30°–40° in one direction then 40°–60° back in the other direction in response to contact with a hermit crab. The snails in our study, particularly *Ilyanassa obsoleta*, were capable of remarkable twisting movements, with angles estimated to be as high as 50° in each direction.

The morphology of the columellar muscle in the foot is similar to that of the preceding region except for the presence of the spherical spaces and the restriction of oblique muscle layers to the dorsal surface only. Although the longitudinal muscle fibers may play a role in retraction of the snail into the shell, their primary function is probably to shorten the foot before it is withdrawn into the shell. The sheets of dorsoventral and transverse muscle fibers probably function to elongate the foot. Contraction of the dorsoventral fibers coupled with maintenance of tension by the transverse fibers and relaxation by the longitudinal fibers would lengthen the foot. Contraction of the dorsoventral fibers with concurrent relaxation of the transverse fibers and maintenance
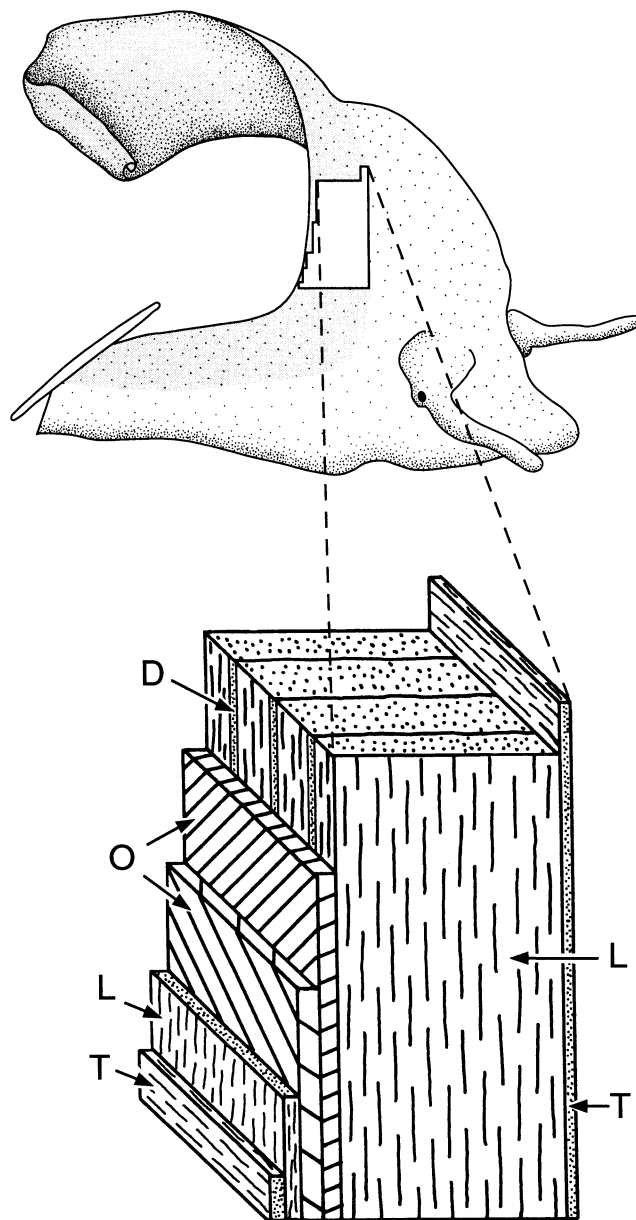

Fig. 4. Schematic diagram of the columellar muscle in the region immediately dorsal to the foot and posterior to the buccal mass showing the arrangement of muscle and connective tissue. The outline of the block diagram corresponds to the outline on the columellar muscle. The left side of the block diagram corresponds to the convex side of the columellar muscle. The majority of the muscle fibers are longitudinal (L). The longitudinal fibers are compartmentalized by regularly spaced sheets of dorsoventral (D) muscle fibers. A layer of transverse (T) muscle fibers is present on the concave surface of the columellar muscle. Two opposed layers of oblique (O) muscles are adjacent to the core of longitudinal and dorsoventral fibers on the convex side. A layer of longitudinal (L) muscle and connective tissue fibers is adjacent to the oblique muscles on the convex side. A thin layer of transverse (T) muscle and connective tissue is present at the surface of the convex side.
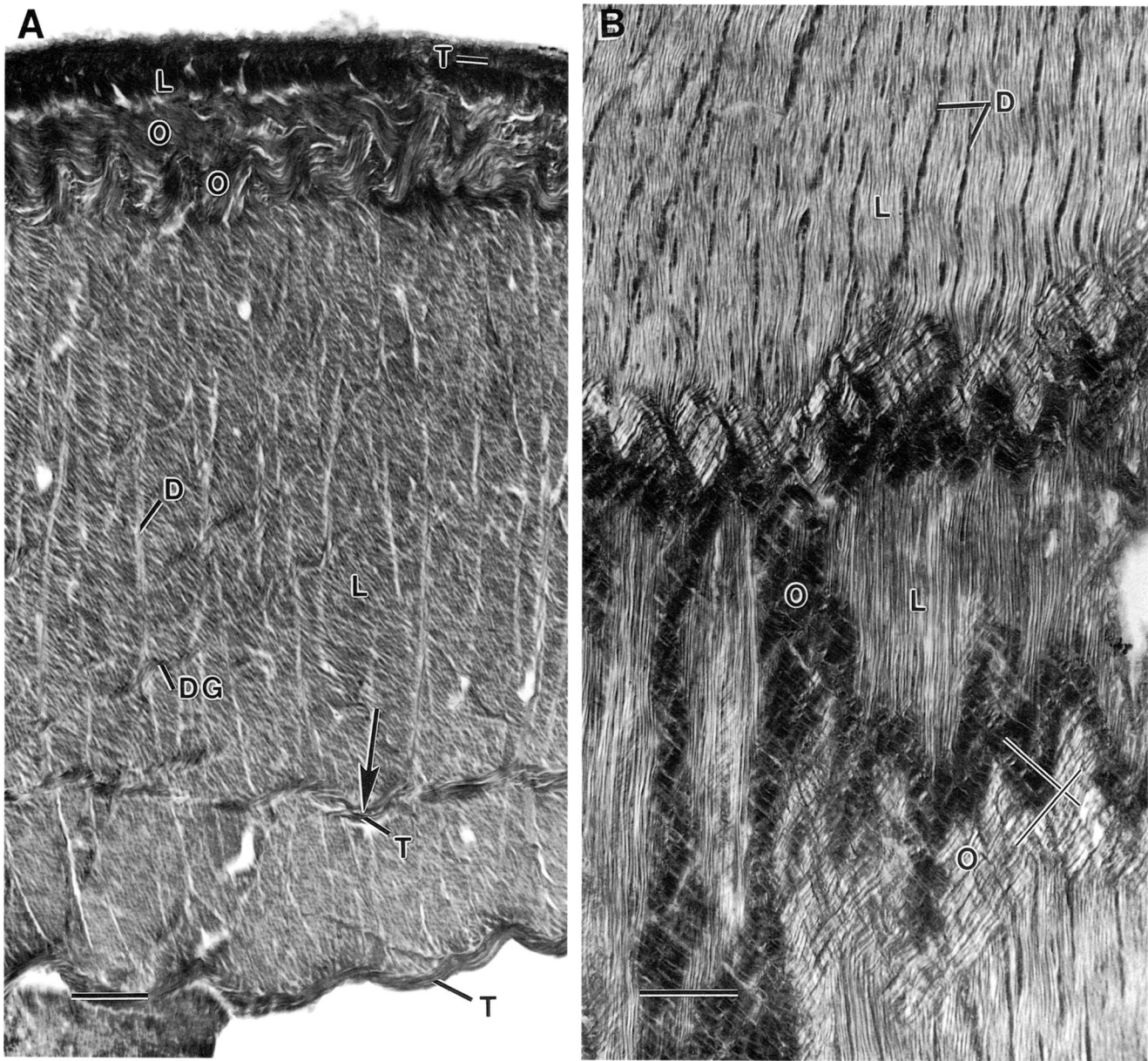
Fig. 5. A. Cross section of the columellar muscle in the region immediately dorsal to the foot and posterior to the buccal mass showing the band of transverse (T) muscles (arrow) characteristic of *Ilyanassa obsoleta*. Dorsoventral muscle, D; diagonal muscle and connective tissue, DG; longitudinal muscle, L; oblique muscle, O. Scale bar, 50 μm. Photomicrograph, brightfield illumination. **B.** Grazing section of the columellar muscle in the region immediately dorsal to the foot and posterior to the buccal mass of *I. obsoleta* showing the oblique (O) muscle layers. The crossed lines indicate the approximate trajectories of the oblique muscle and connective tissue fibers. Also visible in the section is the core of longitudinal (L) and dorsoventral (D) muscle fibers. Scale bar, 50 μm. Photomicrograph, polarized light.

of tension by the longitudinal fibers would widen the foot perpendicular to its long axis. The oblique muscle layers probably function in twisting of the foot relative to, not the shell, but the long axis of the foot. Contraction of the diagonal muscle and connective tissue fibers would tend to curl the lateral edges of the foot ventrally.

During protraction out of the shell, the foot is not in contact with the aperture of the shell. This observation is consistent with a similar observation in *Bullia digitalis* by Brown & Trueman (1982), and, as noted by these authors, does not support the hypothesis that snails inflate the foot with hemolymph so that the foot drags the animal out of the shell (Brown 1964; Dale 1974). For an alternative explanation of protraction in pulmonate gastropods, see Dale (1974).

Connective tissue in the columellar muscle

The presence of connective tissue surrounding individual fibers and groups of fibers in gastropod columellar muscles, in particular near the site of origin on the shell, has been noted previously (Frescura & Hodgson 1989; Voltzow 1990). The abundant collagen near the origin on the columella probably serves as a tendinous attachment to the shell, while connective tissue fibers in the columellar muscle transmit the force of muscular contraction, limit deformation, and provide structural rigidity (Trueman & Hodgson 1990; Voltzow 1990). Although it has been suggested that collagen in the columellar muscle may function as a "catch connective tissue" analogous to that in echinoderms (Motokawa 1988), experimental evidence for such a mechanism in molluscan collagen is lacking (Frescura & Hodgson 1989).

Functional implications of non-circular cross section

Not all prosobranch gastropods have columellar muscles that are elliptical in cross section. In members of the Haliotidae, the pedal retractor muscle (the muscle homologous to the columellar muscle in more coiled prosobranch gastropods) is nearly circular in cross section. In monoplacophorans, the presumed ancestors of prosobranch gastropods (Runnegar & Pojeta 1985), the paired pedal retractor muscles are circular, or nearly circular, in cross section (Lemche & Wingstrand 1959). Why, then, is the columellar muscle in coiled gastropods elliptical in cross section? The shape may be a consequence of packing in a cramped shell. Indeed, some gastropods (e.g., Nerita; Vermeij 1993) remodel the inside of the shell to increase the volume. Alternatively, an elliptical cross section may confer some advantage when wrapped around the coiled columella of the shell. Regardless of the causes of the elliptical cross section of the columellar muscle, there are mechanical consequences for snails with regard to supporting the shell and the ease of performing twisting movements.

One consequence of an elliptical cross-sectional shape compared with a circular cross-sectional shape is a reduction in the force available for twisting of the columellar muscle around its long axis and, thereby, for twisting the foot relative to the shell. In the columellar muscle and in other muscular hydrostats, the oblique or helical muscle arrays are found in a peripheral position, i.e., wrapping the structure at a distance from its central longitudinal axis. Such an arrangement provides a larger torsional moment than if the oblique muscle arrays are more centrally located (Kier & Smith 1985). With other factors held constant, the far-
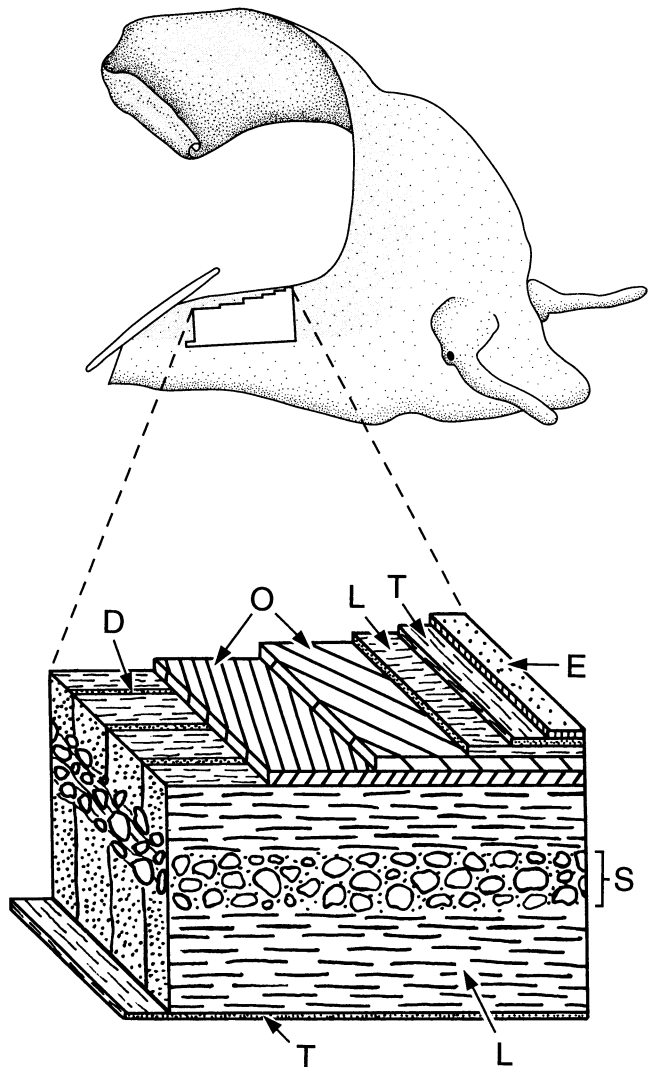

Fig. 6. Schematic diagram of the columellar muscle in the foot showing the orientations of muscle and connective tissue. The diagram is representative of all three species studied. The outline of the block diagram corresponds to the outline on the columellar muscle. The upper surface of the block diagram is the convex side of the muscle. The majority of the muscle fibers are longitudinal (L). The longitudinal fibers are compartmentalized by regularly-spaced sheets of dorsoventral (D) muscle fibers. A layer of transverse (T) muscle fibers is present on the concave surface of the columellar muscle. Two opposed layers of oblique (O) muscles are adjacent to the core of longitudinal and dorsoventral fibers on the convex side only. A layer of longitudinal (L) muscle and connective tissue fibers is adjacent to the oblique muscles on the convex side. A thin layer of transverse (T) muscle and connective tissue is present at the surface of the convex side. Note the band of spherical spaces (S) that runs through the central region.
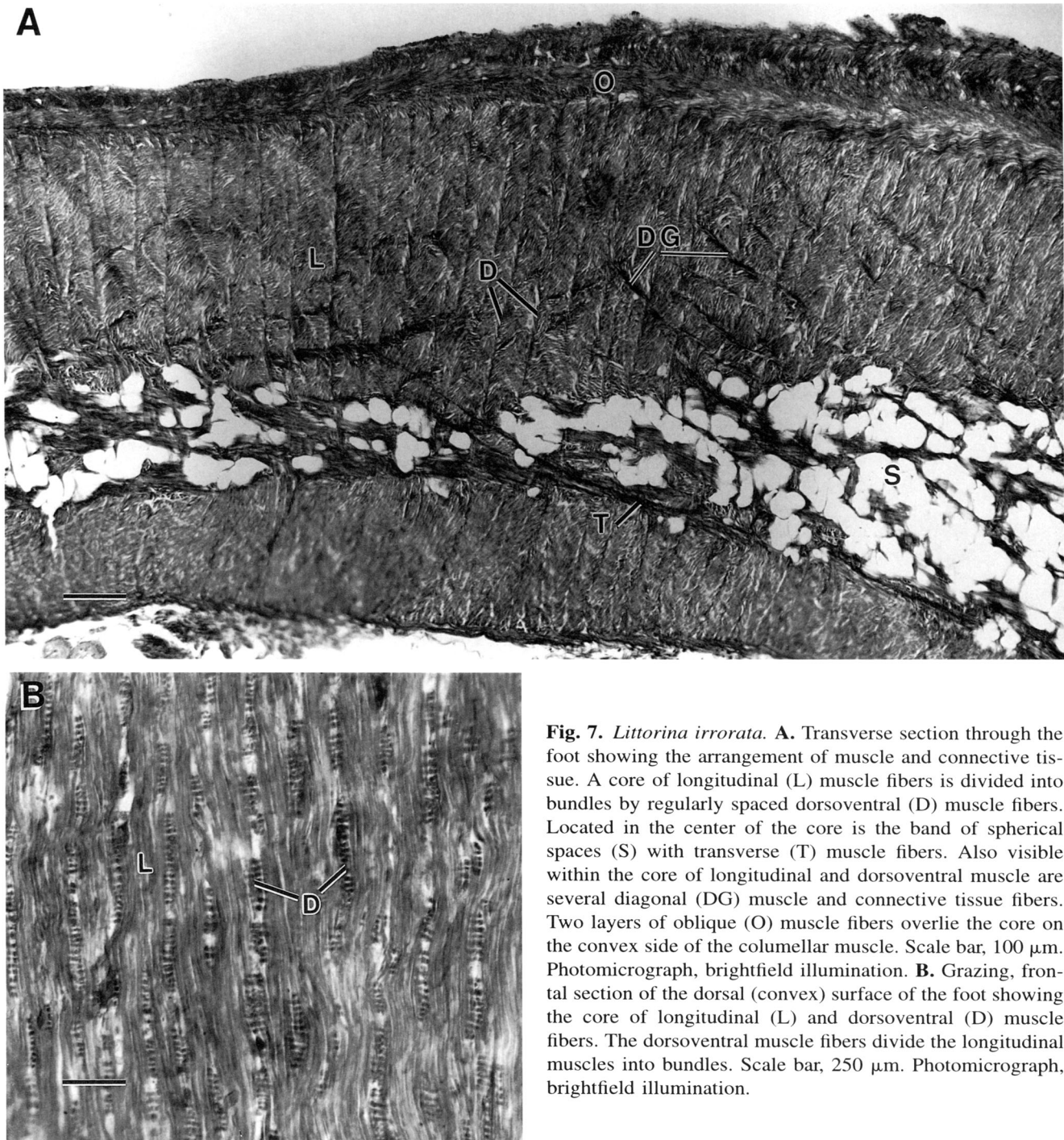
A

B

Fig. 7. *Littorina irrorata.* **A.** Transverse section through the foot showing the arrangement of muscle and connective tissue. A core of longitudinal (L) muscle fibers is divided into bundles by regularly spaced dorsoventral (D) muscle fibers. Located in the center of the core is the band of spherical spaces (S) with transverse (T) muscle fibers. Also visible within the core of longitudinal and dorsoventral muscle are several diagonal (DG) muscle and connective tissue fibers. Two layers of oblique (O) muscle fibers overlie the core on the convex side of the columellar muscle. Scale bar, 100 μm. Photomicrograph, brightfield illumination. **B.** Grazing, frontal section of the dorsal (convex) surface of the foot showing the core of longitudinal (L) and dorsoventral (D) muscle fibers. The dorsoventral muscle fibers divide the longitudinal muscles into bundles. Scale bar, 250 μm. Photomicrograph, brightfield illumination.

ther the muscle is from the axis of torsion, the lower the force that must be produced to cause torsion. It is useful to compare, therefore, the torsional moment available for two simplified columellar muscle shapes, one of circular cross-sectional shape (representative of the pedal retractor muscles of monoplacophorans and members of the Haliotidae), and another of elliptical

cross section (representative of the columellar muscle observed in tightly coiled prosobranchs).

For the comparison, we assume that the two shapes have the same cross-sectional area, that they consist of material of identical mechanical properties, that the torsional force is applied tangential to the surface and is integrated around the periphery, and that the major

to minor axis ratio of the shape with an elliptical cross section is 4:1. If the cross-sectional shape of the columellar muscle is approximated as an ellipse, a major to minor axis ratio of 4:1 is a conservative estimate of the ratio present in a real muscle (the actual range is from 4:1 to 10:1 or higher). The calculation suggests that the muscular force required to produce torsion is ~1.6 times higher in a columellar muscle of elliptical cross section than in one circular in cross section. Since a muscle that is circular in cross section requires lower muscular force to produce torsion, this suggests that oblique musculature should be present where the columellar muscle most approximates a circular cross-sectional shape. In the three species studied, the oblique musculature was thickest and completely encircled the columellar muscle in the region immediately dorsal to the foot; the columellar muscle in this region was the most circular in cross section. For *I. obsoleta*, the ratio of major to minor axis was only 4:1 in this region, compared with 10:1 (or higher) in the region near the origin and 5:1 in the foot.

Support of the shell

The elliptical cross section of the columellar muscle also has mechanical implications with respect to protraction of the body from the shell and support of the shell above the foot. Both of these functions involve loading of the columellar muscle in longitudinal compression. The ability of a structure to resist longitudinal compression depends, in part, on cross-sectional shape; the elliptical cross section of the columellar muscles examined in this study may reduce the compressive loads that can be resisted, when compared with a more circular cross section.

To examine this issue, we compared two simple shapes; one a cylinder of circular cross section, and the other a cylinder of elliptical cross section with a major to minor axis ratio of 4:1. All other aspects of the two shapes, e.g., mechanical properties of the material, length, and cross-sectional area are identical. We consider the models to be fixed at one end and free at the other, and loaded in compression by the mass of the shell. As the compressive load on a slender column increases, at some point the column will bow laterally under the load in a process called buckling. This point, called the critical Euler buckling force, depends on the length of the column, the Young's modulus of the column, the second moment of area of the cross section of the column, and the nature of the restraint at the ends of the column (Wainwright et al. 1976). For our comparison, the length, Young's modulus, and nature of loading are identical and thus any difference in the critical Euler buckling force depends only on the second moment of

area of the two cross sections. The second moment of area of a column of elliptical cross section with a major to minor axis ratio of 4:1 is 4 times smaller than a column that is circular in cross section, and thus the critical Euler buckling force for the column of elliptical cross section will also be 4 times smaller.

Although these calculations suggest that a change from a circular to a more elliptical cross section reduces the ability of the columellar muscle to resist compressive loads, we do not have sufficient information to calculate the actual critical Euler buckling force for the columellar muscle. To do so would require detailed knowledge of the mechanical properties of the various components of an active columellar muscle. In addition, the simple formula used here does not take into account the complex helical shape of the columellar muscle, potential support provided because the columellar muscle rests against the columella, and support from the head and buccal mass.

Generality of results and future directions

The three species of snails selected for this study represent a broad ecological range: *Calliostoma euglyptum* is carnivorous on epibenthic invertebrates and lives on subtidal hard substrata or sand (Ruppert & Fox 1988); *Littorina irrorata* is a grazer and is found almost exclusively on estuarine *Spartina* sp. or eel grass (Ruppert & Fox 1988; unpubl. observ.); *I. obsoleta* is a deposit feeder and lives on estuarine mud and sand flats (Ruppert & Fox 1988; unpubl. observ.). Broad ecological range implies nothing about phylogenetic diversity and, therefore, we do not claim that our findings are general for all prosobranch gastropods. However, our study does allow us to make some predictions of columellar muscle morphology based on the degree of shell coiling.

All else being equal, those animals with tightly coiled shells (and, presumably columellar muscles with non-circular cross-section) are likely to have proportionately more robust oblique muscle layers than animals with less coiled or straight shells. In addition, oblique muscles are likely to be located along the region of the columellar muscle that is most circular in cross section (relative to the rest of the muscle). In an animal with a tightly coiled shell, this region is likely to be where the columellar muscle enters the mesopodium. In an animal with a straight or barely coiled shell, oblique muscles may be located along the entire length of the muscle.

However, a definitive statement about the relationship between morphology and degree of coiling will require additional sampling of taxa that include the entire range of coiling geometry from the many lim-

pet-like forms to those with tightly coiled shells. In addition, future comparative studies conducted in the context of our emerging understanding of the phylogeny of gastropods (e.g., Taylor 1996) are needed in order to gain insight into the evolution of columellar muscle morphology and function. Finally, experimental studies are required in order to test the proposals for function presented here.

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