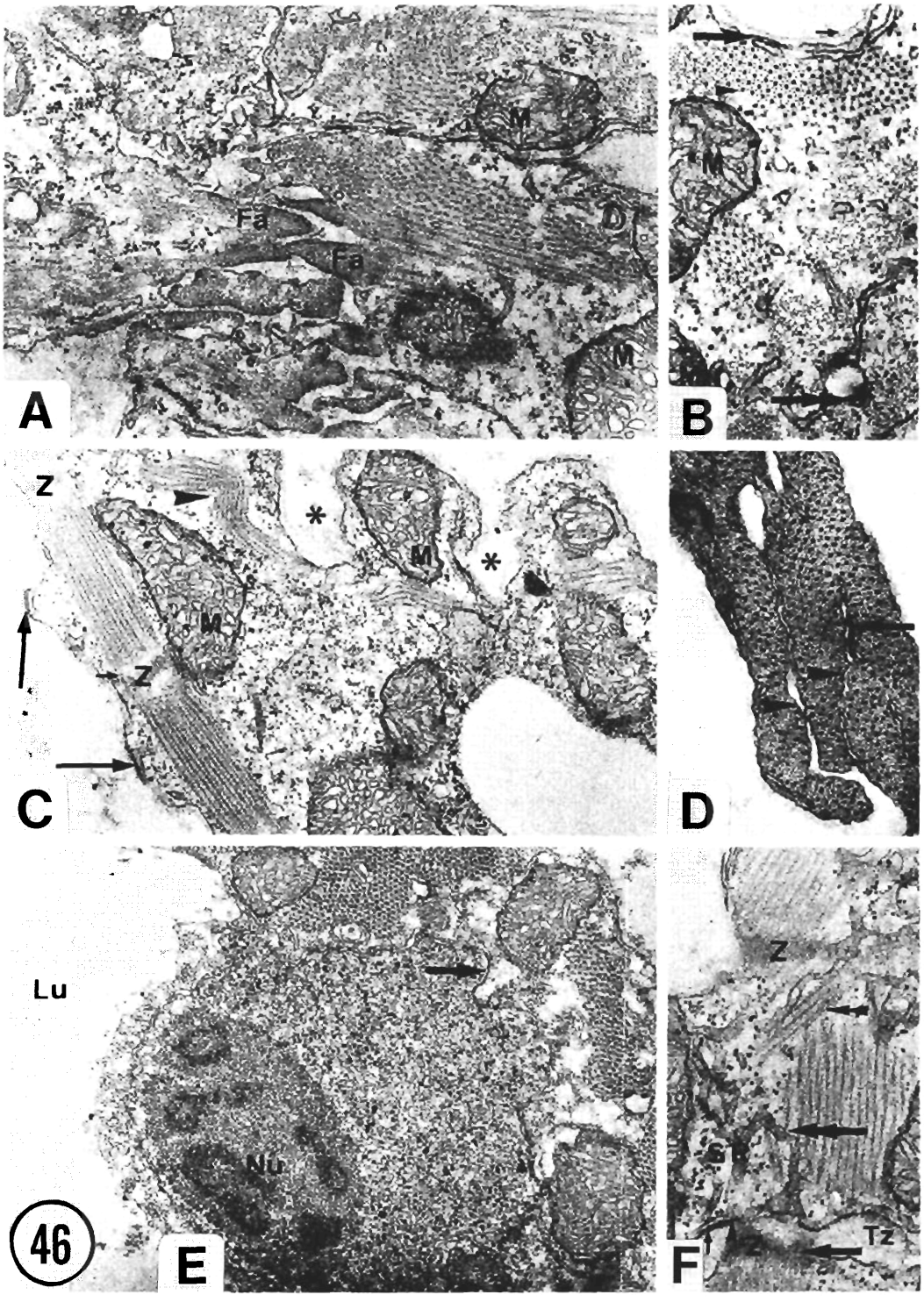


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C). Invaginations of sarcolemma occur only in Z-band levels on both sides of the myocardium (Figs. 46C, 47A). These invaginations begin as clefts or Tz-tubules (Steinsland, 1982) to which the Z-band material is anchored (Fig. 46F). These Tz-tubules are wide (up to 350 nm) and covered by a basal lamina (Fig. 47C). Steinsland (1982) stated that communications between the luminal and the pericardial sides occur. The Tz-tubules branch into longitudinal tubules (LT) that run within or between myofibrils (Fig. 47C). The LT system "is continuous with the T system of neighboring sarcomeres and the intercellular space in intercalated discs" (Steinsland, 1982). The sarcoplasmic reticulum (SR) does not encircle the myofibrils, but there are transverse tubules at the Z- and A–I-band levels (Figs. 46F, 47C) (Steinsland, 1982). The transverse SR tubules at the Z levels form "interior couplings" with Tz-tubules and are interconnected with LT, which they follow, and with which they form the interior couplings between tubules at A-band levels (Figs. 46F, 47A–C). The intercalated discs, which Steinsland (1982) found in connection with the ventral ridge, are of the "normal interdigitating type" found in other nonmalacostracans (Steinsland, 1982). Fasciae adherentes are common (Fig. 46A) and desmosomes, which lack a central dense line, are also present (Fig. 47B,E).

Thus, *Daphnia* has a thinner myocardium than does *Lepidurus; Lepidurus* also has "very unorganized contractile material and membrane systems" (Steinsland, 1982). The extensive, complicated system of T-tubules seen in *Lepidurus* differs slightly from the more regular Tz and LT system of *Daphnia*. *Branchipus* almost lacks an LT system, although there is a regular transverse system with invaginations at the Z-band level. The SR in *Daphnia* and in *Branchipus* is regular with transverse systems at the Z- and A–I-band levels, which form couplings with the exposed sarcolemma (Steinsland, 1982). Transverse tubules are interconnected with longitudinal tubules between or within myofibrils; the SR system in *Lepidurus* also makes some peripheral couplings. Attachment plaques, which Steinsland (1982) described as a "primitive condition found in the myocardial cells of molluscs," are known only in *Lepidurus* (Tjønneland et al., 1980). Other differences, such as the nature of the myofibril patterns, couplings at Z- and A-band levels, etc., are discussed by Steinsland (1982) and Økland et al. (1982).

Fig. 46. Heart of *Daphnia pulex*. (From Steinsland, 1982.) **A:** Cross section showing intercalated disc at ventral side of heart. Fascia adherentes (Fa) are common. ×26,000. **B:** Cross section of myofibrils showing low electron density of core of thick filaments (small arrowhead) and hexagonal arrangement of thin filaments around thick. Peripheral couplings between sarcoplasmic reticulum and exposed sarcolemma (arrow) and internal coupling with longitudinal T-tubule (double-headed arrow) are seen. Thin basal lamina (small arrows) is indicated. ×33,000. **C:** Longitudinal section of myofibrils showing distribution of contractile material (no polarization). Z-bands (Z) and I-band (small arrow) indicated. Some myofibrils appear bent (arrowhead). Peripheral couplings at A-band levels (arrows) and invaginations at Z-band (asterisks) are also seen. ×20,000. **D:** Cross section of myofibril showing membrane systems. Tz tubules (arrowheads) invaginate at Z/I-band level and make internal coupling with sarcoplasmic reticulum (arrow). ×34,500. **E:** Nucleus (Nu), with nuclear pore indicated (arrow), adjacent to lumen (Lu). No polarization of myocardium evident. ×20,000. **F:** Longitudinal section of myofibril showing distribution of sarcoplasmic reticulum (SR). Transverse SR system at the Z-band level (arrowhead), which makes internal couplings with Tz tubule (small arrow), and at the A-band level (double-headed arrow) indicated. Diffuse Z-band material (Z) is anchored to the Tz tubules (arrow). Note direction of contractile material (double arrowhead). Tz is a transverse tubule at the Z-band level. ×26,000. M, mitochondria.

## Circulation

Other than the heart, branchiopods have no true blood vessels, instead having an open or lacunar system, as do all Crustacea. However, there are recognizable channels that are, according to Greene (1924), "well marked and fairly definite," and the movement of the blood cells can be followed through the body and into the appendages (Greene, 1924; Vehstedt, 1940). Two mechanisms are responsible for movement of the blood: regular pumping of the heart musculature and general body movements, mostly those of the appendages. Heart contraction is rhythmic and peri-


Fig. 47. Heart of *Daphnia pulex*. (From Steinsland, 1982.) **A:** Longitudinal section of supercontracted sarcomeres showing wide Z-bands (Z) and invaginations at Z-band levels (asterisks) on both luminal and pericardial sides. Peripheral couplings between sarcoplasmic reticulum and exposed sarcolemma (large arrows) are seen. Internal couplings between sarcoplasmic reticulum (double-headed arrow) and transverse tubules (arrow) and with longitudinal tubular system (arrowhead) are indicated. ×20,000. **B:** Longitudinal section of sarcomere showing internal couplings between longitudinal tubule and sarcoplasmic reticulum (arrowhead). Note invagination at Z-band level (asterisk) and diffuse Z-bands (Z). Desmosome (arrow) indicated. ×33,000. **C:** Longitudinal section of myofibrils showing internal couplings between sarcoplasmic reticulum and Tz tubules (small arrows) and longitudinal tubular system (small arrowheads). The longitudinal tubular system branches off Tz (large arrowhead). Diffuse Z-bands (Z) in neighboring myofibrils not necessarily aligned. Basal lamina in Tz indicated (double-headed arrow). Note direction of contracted material (double arrowhead). ×20,000. **D:** Section showing H-bands (arrows). ×1,600. **E:** Cell border with desmosome (arrow). ×33,000. Lu, lumen of heart; M, mitochondria; Tz, transverse tubule at Z-band level; Z, Z-bands.

staltic, with the wave beginning at the posterior end and running anteriorly. Rate varies with size and external conditions; a rate of 125 beats per minute was recorded for *Artemia* under normal conditions (Greene, 1924), whereas *Daphnia*'s rate is 300 to 450 beats per minute (Kaestner, 1970; Postmes et al., 1989). Interestingly, heart rate in *Daphnia* is affected by changes in illumination even when compound and naupliar eyes are removed, suggesting myogenic heart control (see also Postmes et al., 1989). Conversely, illumination of the eyes alone elicits no response (Schultz, 1928; Peters, 1987; Ringelberg, 1987). Blood flows into the heart through the valved posterior and lateral ostia and out through the anterior ostium. The general direction of blood flow is from the anterior "aorta" into the head, and then posteriorly along the gut and into the caudal lymph spaces (Greene, 1924), with lateral blood flow into the appendages easily observed as well. In the "respiratory plates" (the epipods, discussed in the section on respiration), Greene (1924) thought that the open vascular lacunae were in the form of "quite definite capillary-like patterns," and he described the movement of blood cells into the limb epipod on one side and back via adjacent smaller spaces. Possibly what Greene saw was the reticulated pattern of the epithelial cells of the epithelium of these epipods (see Holliday et al., 1990), and if indeed movement of blood is visible around these cells, then the view of Moens et al. (1991) that the epipods do function in respiration by providing direct contact between the hemolymph and cuticle in intercelluar spaces receives support.

### Blood-Forming Organs

I have been unable to locate any detailed study of the organs that form new blood cells subsequent to that of Lochhead and Lochhead (1941), who discredited previous suggestions that new blood cells are formed by cellular division of circulating old cells. The blood-forming organs are, at least in *Artemia* and *Branchipus*, small (50–150 μm length), poorly defined nodules located at the base of each trunk appendage (Fig. 48A) (there are 22 in *Artemia*) (Cassel, 1937; Lochhead and Lochhead, 1941). Each nodule consists of a group of rounded cells, which differ in size and in nuclear activity (because of different stages of mitosis), surrounded by a thin membrane, probably connective tissue, approximately 1 μm thick and showing occasional discontinuities (Fig. 48A,B). There are no "stroma" cells, the cells within a nodule being all of one type, and in this respect the organs differ from similar blood-forming organs described in decapods (Lochhead and Lochhead, 1941; Criel, 1991a). In cells closer to the periphery of the nodule, the cytoplasm increases with respect to the nucleus, which appears smaller than in the inner (less active) cells. Occasionally "necrotic" nuclei, darker-staining and oval, are seen. In the spaces adjacent to the blood-forming organs are blood cells in intermediate stages of development, phagocytic storage cells, and muscles of the limb (Fig. 48A,B). Each nodule is located between the two nerves that run from the ventral segmental ganglion of that somite into the limb.

### Blood Cells

Circulating blood cells differ from those in the periphery of the blood-forming organs in having more cytoplasm and numerous inclusions, especially the refringent granules. Prior to development of these blood-forming organs, primary hemocytes, which resemble fat-storage cells and contain yolk droplets (Criel, 1991a), can be detected in early naupliar stages (Benesch, 1969).

Circulating blood cells are ameboid and vary considerably in size (Fig. 48C,D) (Criel, 1991a). The cell nucleus is small, and the cytoplasm is typically filled with large refringent granules of unknown composition (Lochhead and Lochhead, 1941; Criel, 1991a). These large inclusions appear in electron micrographs as an electron-dense crescent against a lighter background matrix (Fig. 48E) (Criel, 1991a). The capacity for phago-

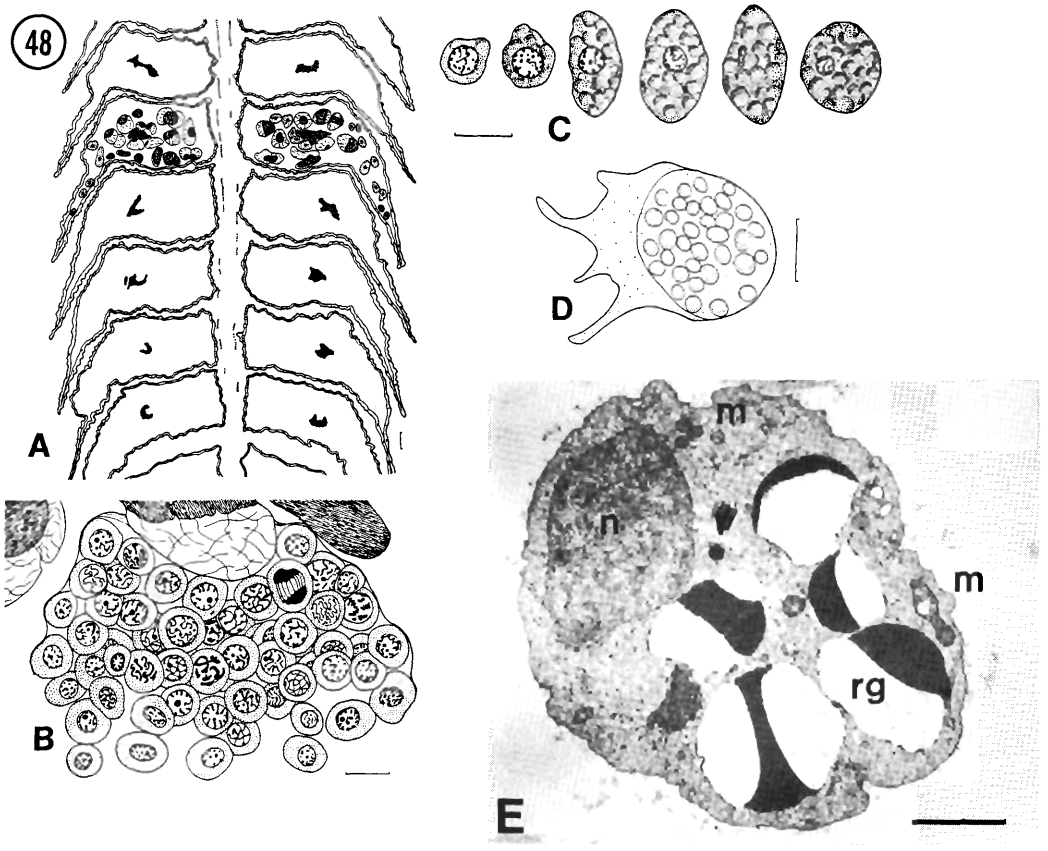


Fig. 48. Blood-forming organs and blood cells of *Artemia*. **A:** Frontal section through bases of six trunk limbs showing blood-forming organ in each limb. Muscles and phagocytic storage cells (latter with large black nuclei) shown in second limb from top. **B:** Blood-forming organ and adjacent muscles and sarcoplasm. Scale bar = 10 μm. **C:** Different types of blood cells seen in sectioned material. Scale bar = 10 μm. **D:** Blood cell as it appears when adhering to tissues in living animal. Scale bar = 5 μm. (A–D, from Lochhead and Lochhead, 1941.) E: TEM through *Artemia* blood cell. Scale bar = 2 μm. (Courtesy of G. Criel.) m, mitochondria; n, nucleus; rg, refringent granules.

cytosis attributed to the blood cells by earlier workers (e.g., by Kollman, 1925; Tyson, 1975) possibly results from confusion with the phagocytic storage cells (discussed with connective tissue); it is unlikely that true blood cells have the capacity for phagocytosis (Lochhead and Lochhead, 1941; Benesch, 1969; Criel, 1991a). Blood cells are actively involved in clotting and healing, as would be expected, but some workers have attributed to them some additional function in vitellogenesis (Lochhead and Lochhead, 1941). The blood cells contain no respiratory pigments, since these are extracellular in branchiopods (see section on respiration).

## DIGESTION AND ABSORPTION

Few areas of branchiopod biology have received so much attention and yet have been so misunderstood as the feeding mechanisms. The confusion has sprung mostly from generalizations that cannot encompass the variation among extant orders, such as assuming that all anostracans and all anomopods are filterers. The mechanics of feeding—i.e., the morphology and function of the mouthparts and associated feeding structures and the various feeding behaviors—are beyond the scope of this review, but a brief synopsis follows to emphasize the diversity of feeding modes among extant orders. Most anostracans are

filter feeders, but some species of *Chirocephalus* feed on larger particles; carnivory is known in species of *Branchinecta;* and the mode of feeding may change with ontogeny. Reviews of anostracan feeding can be found in the works of Cannon (1928, 1933, 1935a,b) and Fryer (1966, 1983). Notostracans are benthic scavengers or predators, with mouthparts designed for tearing or grasping (Horne, 1966; Fryer, 1988; Martin, 1989a). Feeding in the "conchostracans" orders was reviewed by Martin (1989a). Spinicaudatans include scrapers, detrital grazers, true filter feeders, and probably several other feeding types. Martin's review of laevicaudatans was erroneous; they do scrape and graze, but the antennae are in no way used in feeding and filtering does not occur (G. Fryer and J. Martin, unpublished data). Haplopods and onychopods are all planktonic or littoral (*Polyphemus*) predators, with apparent omnivory in some onychopods (R. Barnhisel, personal communication). Anomopods and ctenopods include a variety of feeding modes, including filtering, scraping, scavenging, and parasitism (Eriksson, 1934; Fryer, 1968, 1974, 1987a–c, 1991a; Lampert, 1987).

The branchiopod gut lies in the open hemocoelic cavity. The alimentary tract is basically, and probably primitively, a simple straight tube that is abruptly curved where the esophagus joins the midgut (e.g., anostracans and notostracans; Fig. 49A). But this simple design has been modified in those taxa with short bodies, so that the gut is C-shaped in some of the smaller bivalved taxa (Fig. 49B). It is even convoluted or looped in some of the anomopods (Fig. 49C,D). The gut is composed of three regions: the anterior foregut (= esophagus, stomodaeum), the midgut, and the hindgut (= rectum, proctodaeum) (see Dall and Moriarty, 1983, for a review of crustacean gut terminology).

The following discussion is based primarily on the works of Kikuchi (1971), Hootman and Conte (1974), Snyder and Wolfe (1980), Foster and Wolfe (1986), Criel (1991a), and especially Schrehardt (1987b), from which many of the figures were taken. All of the above studies are on *Artemia,* a filter feeder living in high salinity waters. Because in *Artemia* the gut is also employed in salt balance (Hootman and Conte, 1974; Conte, 1984; Holliday et al., 1990), possibly not the case in the other (freshwater) groups, the ultrastructure may not apply to other taxa (see Schultz and Kennedy, 1976, for *Daphnia*; Günzl, 1991, for *Alona affinis*).

### Foregut

The foregut (or esophagus) (Figs. 50, 51, 52A) is derived from stomodeal ectoderm and hence is lined with very thin cuticle (about 0.25 μm in *Artemia*). In *Artemia,* the only areas of the body with comparably thin cuticle are the hindgut and epipods, suggesting to Claus (1886) that respiration might occur across esophageal and hindgut cuticle, but this is unlikely considering the thickness of the foregut and hindgut epithelium (Schrehardt, 1987b). Epithelial cells underlying this thin layer of cuticle are cuboidal anteriorly, becoming more columnar as the esophagus approaches its junction with the midgut. Adjacent cells form apical zonulae adherentes and basal tight junctions (Schrehardt, 1987b). The nucleus of these cells is 6–7 μm long and contains a single conspicuous nucleolus (Schrehardt, 1987b, fig. 10). Cytoplasm tends to be electron-dense and contains ribosomes along the nuclear membrane, free ribosomes, cisternae of RER, mitochondria, and Golgi apparati (Fig. 50A–D). Small amounts of glycogen and lipids occur as inclusions (Fig. 50C). As the epithelial cells become closer to those of the midgut, a finely granular substance, the function of which is unknown but possibly connected with cuticle deposition, can be seen between the apical cell membrane and the cuticle (Fig. 51A,B). The basal lamina separating these cells from the hemocoel is finely granular and monolayered (Schrehardt, 1987b). Longitudinal and circular muscle fibers surround the esophagus and provide for its contraction. This is seen also in the conchostracans (e.g., see Martin, 1989a,

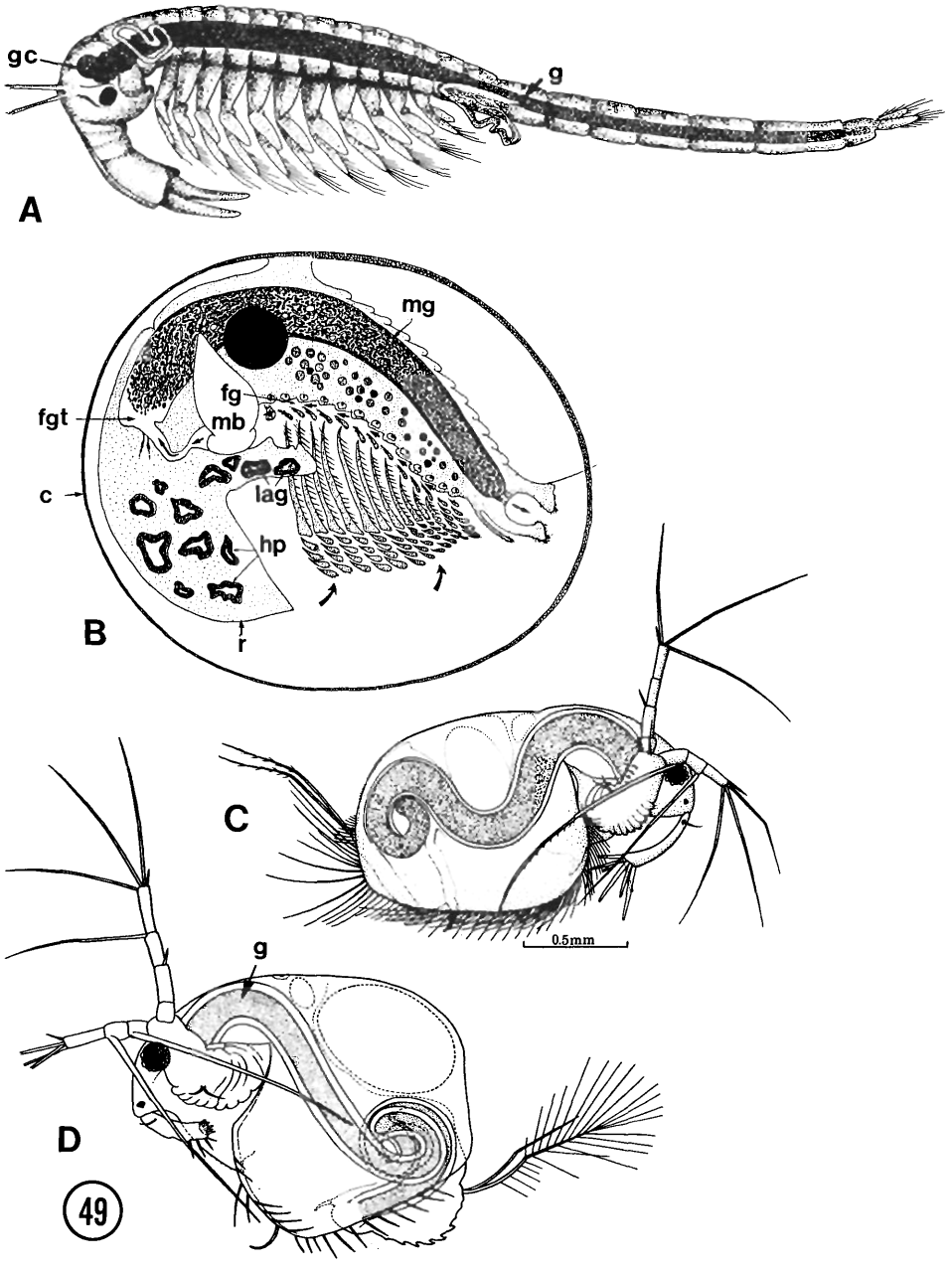


Fig. 49. General form of the alimentary canal in selected orders. **A:** *Branchinecta paludosa* (Anostraca). (After Sars, 1896.) **B:** *Lynceus gracilicornis* (Laevicaudata), with arrows showing general direction of food movement. (After Martin, 1989a.) **C,D:** *Acantholeberis curvirostris* **(C)** and *Streblocerus serricaudatus* **(D),** two anomopods with convoluted or looped guts. (Both after Fryer, 1974.) c, carapace; fg, food groove; fgt, foregut (placement slightly incorrect); g, gut; gc, gastric ceca; hp, hepatopancreas (= gastric ceca); lag, labral glands; mb, mandible; mg, midgut; r, rostrum.

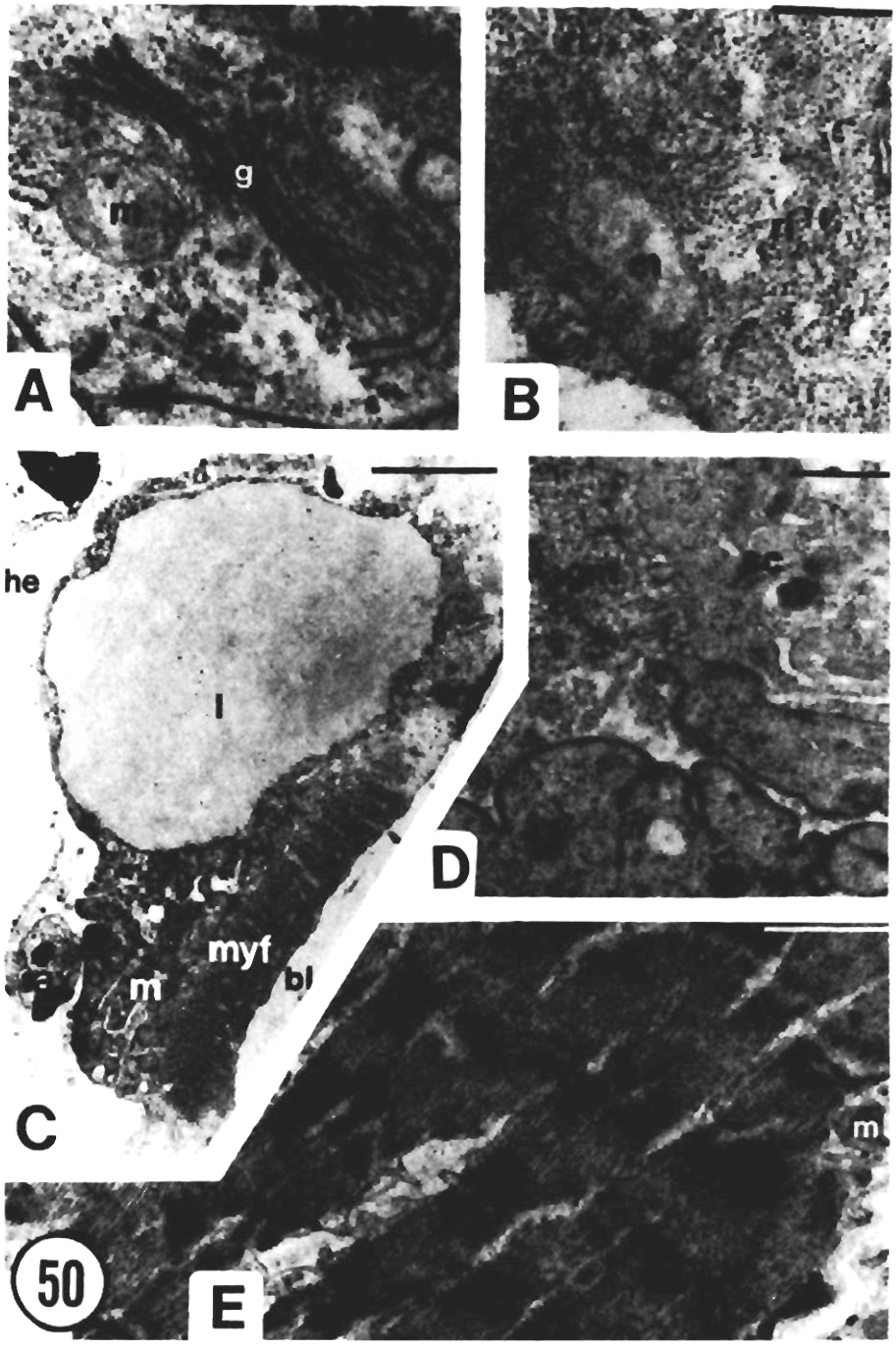


Fig. 50. Cells of the esophagus (foregut). (From Schrehardt, 1987b.) **A:** Apical portion of an esophagus cell with Golgi complex. Scale bar = 1 μm. **B:** Ribosome field of apical portion of esophagus cell. Scale bar = 1 μm. **C:** Transverse section through axon (ax) and adjacent circular muscle cell, separated from esophagus epithelium by basal lamina (bl). Note synaptic vesicles of the axon and the large lipid droplet of the muscle cell. Scale bar = 2 μm. **D:** Apical portion of secretory cell near junction of midgut and esophagus. Note secretion of granular matrix. Scale bar = 1 μm. **E:** Tangential section through dilator muscle of esophagus. Scale bar = 2 μm. ax, axon; g, Golgi apparatus; gm, granular matrix; he, hemocoel; l, lipid; m, mitochondrion; myf, myofilaments; ri, ribosomes; sc, secretory cell.

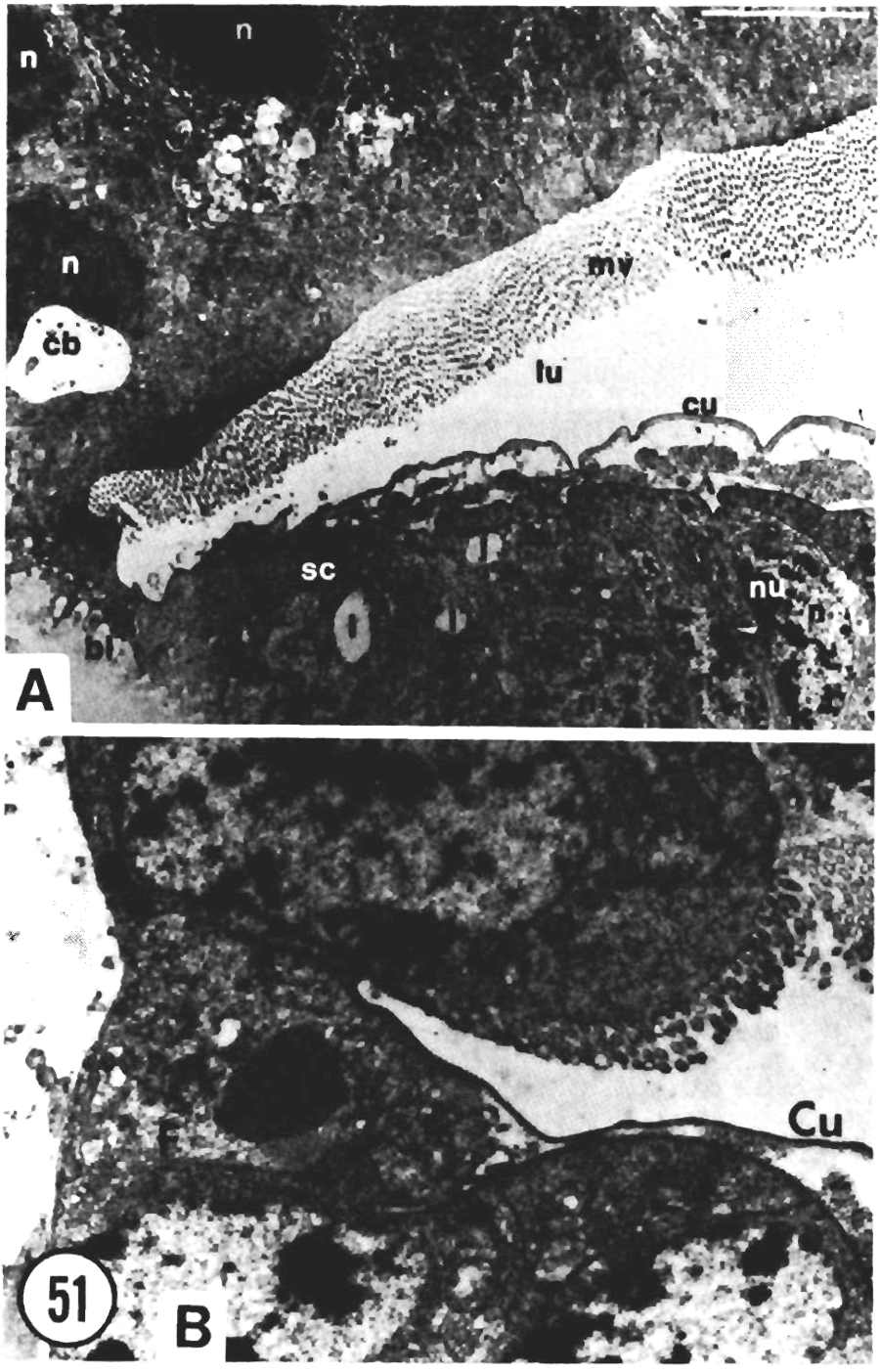


Fig. 51. Junction of esophagus with midgut. **A:** Transition to the midgut is evidenced by loss of foregut cell cuticle and microvillous border of midgut epithelium. (From Schrehardt, 1987b.) Scale bar = 5 μm. **B:** Junction between foregut and midgut epithelia showing long septate desmosome (arrow). (From Hootman and Conte, 1974.) ×8,400. bl, basal lamina; cb, cytoplasmic body; Cu, cu, cuticle; F, foregut; l, lipid; lu, lumen; mv, microvilli; n, nucleus; nu, nucleolus; sc, secretory cell.

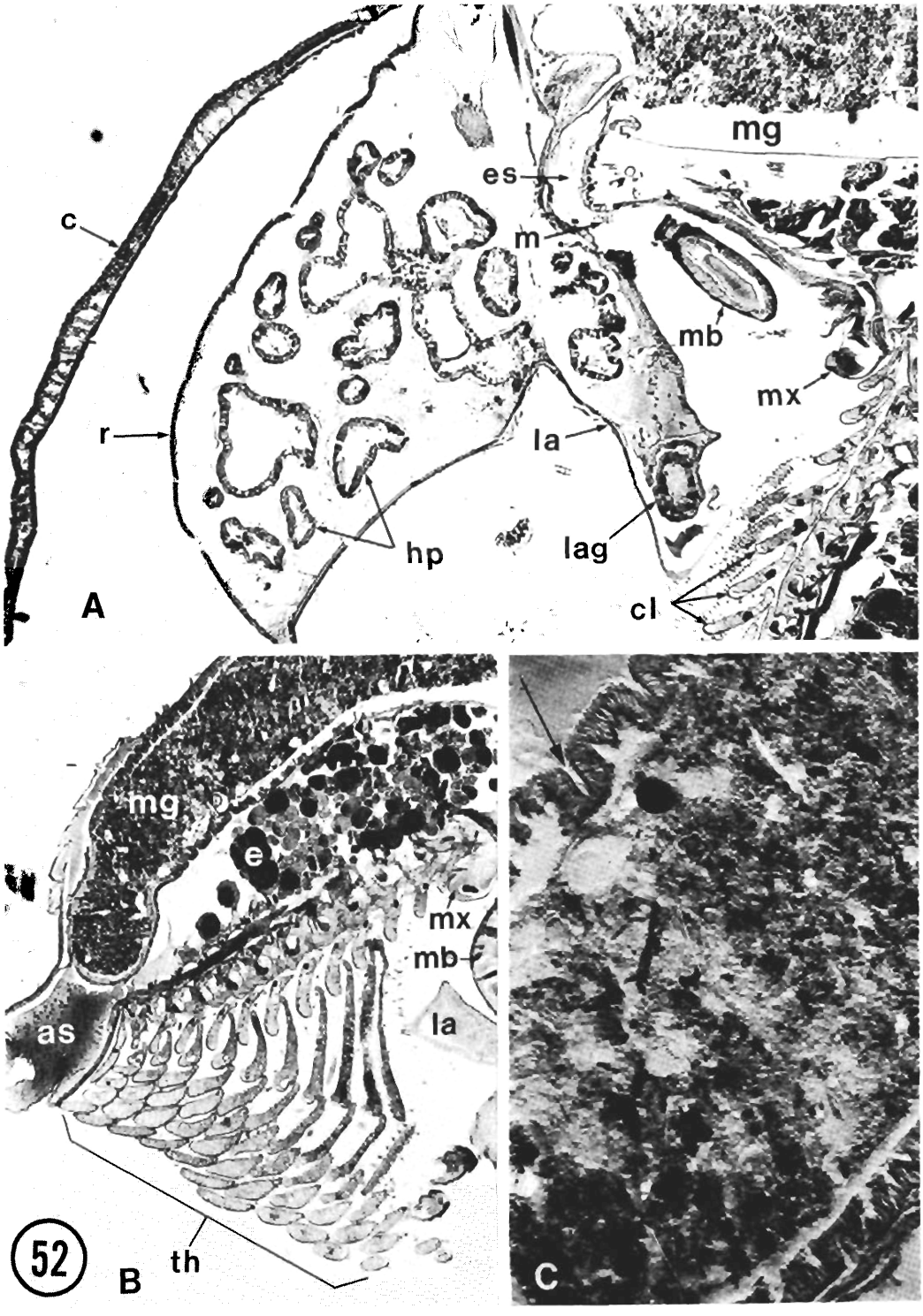


Fig. 52. Sagittal sections through gut of female *Lynceus gracilicornis* (Laevicaudata). (Modified after Martin, 1989a.) **A:** Head region showing mouth, esophagus, midgut, and extensive hepatopancreas (gastric ceca). **B:** Posterior region showing packed midgut, anal somite, and thoracopods (th). **C:** High magnification of midgut; arrow indicates convoluted lining of gut wall. Scale not given. as, anal somite; c, carapace cuticle; cl, coxal lobe of thoracopods; e, egg; es, esophagus; la, labrum; lag, labral glands; m, mouth; mb, mandible; mg, midgut; mx, maxilla; r, cuticle of rostrum.

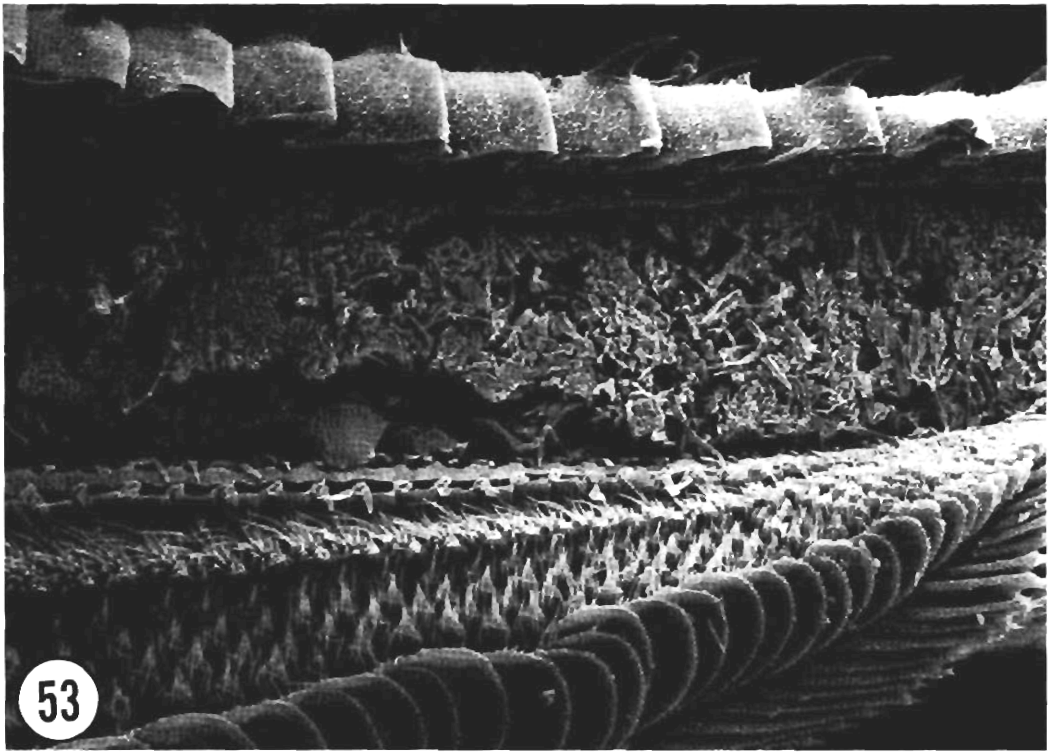


Fig. 53. Paraffin carved section through gut in posterior region of thorax of *Triops longicaudatus* (Notostraca). Visible are circular muscles, cuticle at top of food groove, and thoracic limb gnathobases. (Courtesy of B. Felgenhauer.)

for *Lynceus*). Cells of the circular muscles contain "typical striated myofibrils" and are "innervated by mostly multipolar neurons" (Schrehardt, 1987b). These muscle cells also contain mitochondria, cisternae of RER, small amounts of glycogen, and often large lipid globules. Longitudinal muscles are similar in their ultrastructure (Fig. 50E). Interestingly, Schrehardt (1987b) stated that both circular and dilator (longitudinal) muscles are covered by a basal lamina apparently continuous with the lamina underlying the epithelial cells. The esophagus in anostracans probably functions in simple transfer of food to the midgut, as there are no mechanical structures to facilitate further breakdown of food. However, in the Spinicaudata, Schlecht (1979) reported unusual "cuticular ledges" that may play a role in mechanically breaking down food prior to absorption across the midgut epithelium.

### Midgut

The midgut is endodermally derived and consequently is the only region of the gut unlined by cuticle. It is large, occupying nearly all of the body cavity in conchostracans (Fig. 52B) and notostracans (Fig. 53). Transition from the esophagus to the midgut is obvious (Fig. 51A,B), marked not only by the loss of overlying cuticle but by the conspicuous morphology of the midgut epithelial cells. Cells of the midgut, including those of the protruding gastric ceca, are all of one basic type in *Artemia,* although there may be regional specialization (Schrehardt, 1987b; Criel, 1991a). However, cells of the *Artemia* ceca were assigned to three different types by

Foster and Wolfe (1986), depending upon their electron density, and Günzl (1991) described three cell types in the midgut of an anomopod. Schlecht (1979) also found light and dark cells in the midgut of *Daphnia* and the spinicaudatan *Leptestheria,* attributing some of the differences to development (the lighter cells being younger), but noted that some of the light cells appeared mature and had two free cilia (Figs. 58, 59A,B) (*Leptestheria* only). This feature, midgut cells with apical cilia, is unique, an apparent synapomorphy of the "Conchostraca" (Rieder and Schlecht, 1978; Schlecht, 1979), although their presence has not been documented in the Laevicaudata (Martin, 1989a). The apical borders of the midgut epithelial cells are lined with numerous, long (up to 8 μm), cylindrical microvilli, which become progressively shorter from the front to the back of the midgut (Criel, 1991a) (Figs. 51, 54, 55A, 56A, 57). In the midgut of *Artemia,* the microvilli are arranged regularly, approximately 0.25 μm apart (center to center) in a "hexagonal" pattern (Schrehardt, 1987b), and apparently they are coated with a mucopolysaccharide material (Kikuchi, 1971; Schrehardt, 1987b). A regular arrangement of microvilli is also seen in midgut cells of conchostracans (Fig. 58C,D) (Schlecht, 1979). Each microvillus contains fine filaments extending from its center into the apical cytoplasm (Figs. 55A, 56A), the arrangement of which has been termed the terminal web (Schrehardt, 1987b; Criel, 1991a). The apical cell membrane between the microvilli is indented, forming small (up to 250 nm) pinocytotic pits (Schrehardt, 1987b). Nuclei of these cells (midgut and hepatopancreatic) are oval, averaging 8 μm in diameter, and most have two conspicuous nucleoli (Fig. 54). Small amounts of heterochromatin are seen scattered throughout the cytoplasm (Fig. 55A,B) and adhering to the inner nuclear membrane. Large lipid globules, more obvious in well-fed animals, can occupy so much of the cell that other cell components appear distorted (Schrehardt, 1987b). Beneath the apical web, the cytoplasm is rich in mitochondria and additionally contains numerous vesicular bodies, ribosomes, and glycogen particles (Schrehardt, 1987b) (Figs. 55A, 56A). Cisternae of the RER occur in the supranuclear region. Ribosomes can be seen on the outer nuclear membrane and free in the cytoplasm, the latter often aggregated into polysomes (Fig. 56A). Electron-dense granules up to 300 nm in diameter, of unknown function, occur in midgut cells near the esophageal junction. These are found close to the nucleus and are surrounded by many ribosomes. Golgi bodies consisting of three to five cisternae are also found in the supranuclear cytoplasm (Schrehardt, 1987b). Details of midgut cells in the anomopod *Alona affinis* differ in several respects (Günzl, 1991).

Absorption occurs across midgut epithelial cells, as indicated by the presence of microvilli and multivesicular bodies (Foster and Wolfe, 1986). In *Artemia,* the gut is the site of uptake of water, Na, Cl, K, Mg, and $SO_4$ (Conte, 1984; Holliday et al., 1990). All midgut cells contain a phagolysosomal cytoplasmic body (Fig. 56A), indicative of intracellular digestion (Criel, 1991a). Digestive enzymes are known at least in *Artemia* (Kikuchi and Shiraishi, 1969) and *Daphnia* (Hasler, 1935, 1937; Lampert, 1987). Günzl (1991) suggests functions of water and solute transport and excretion in *Alona.*

The midgut cell basal lamina has two layers, thus differing from that of the foregut and hindgut (Figs. 54, 55B, 56B). The basalmost layer (exposed to the hemocoel) is thin, finely granular, and does not extend into the infoldings of the cell membranes, whereas the inner layer is thicker, densely packed with granules and amorphous matrix material, and extends into infoldings of the basal cell membrane (Kikuchi, 1971; Schrehardt, 1987b; Criel, 1991a; Günzl, 1991).

The midgut is surrounded by circular muscle fibers at regular intervals but no longitudinal muscles (Hootman and Conte, 1974; Foster and Wolfe, 1986; Schrehardt, 1987b). The

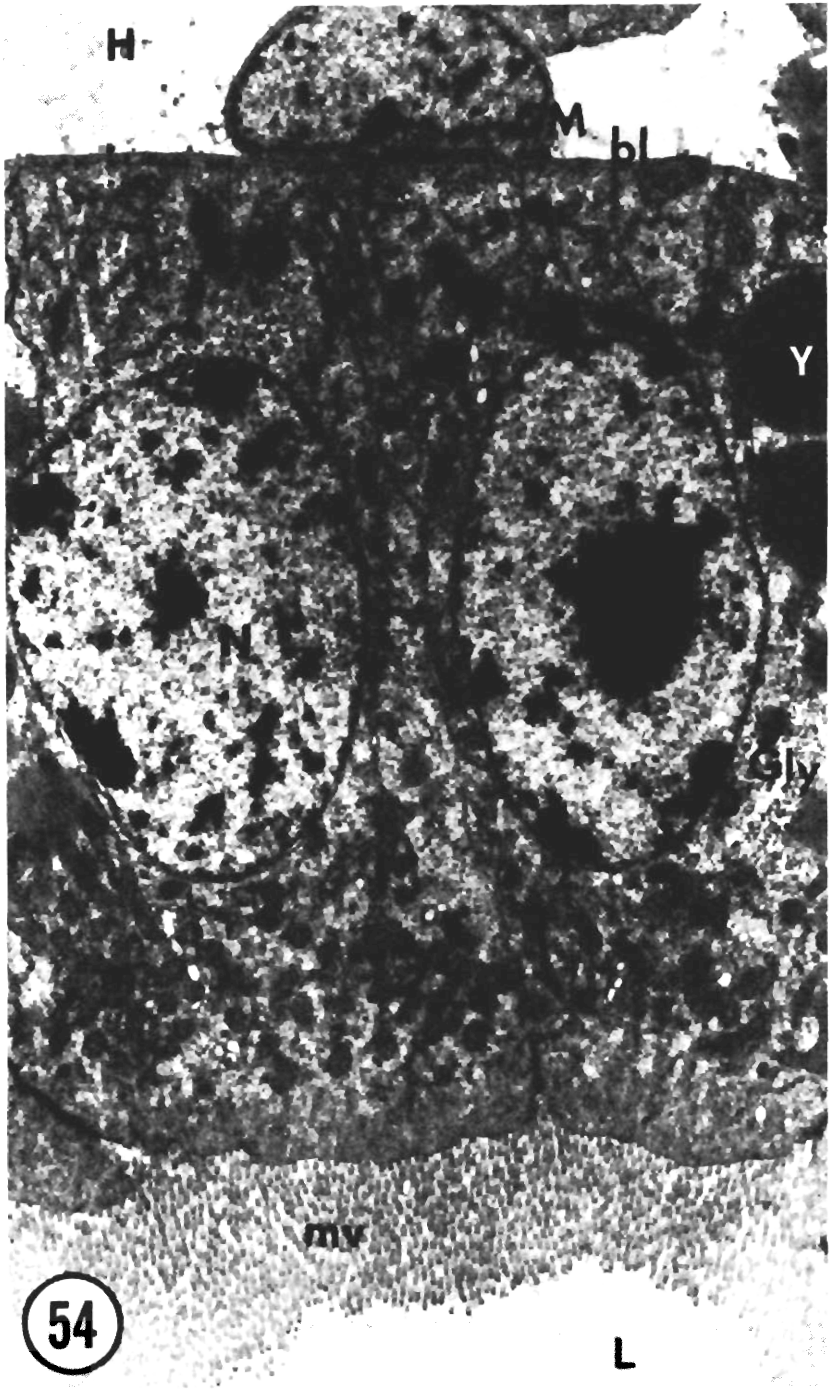


Fig. 54. Portions of four midgut cells in *Artemia*. (From Hootman and Conte, 1974.) bl, basal lamina; CM, circular muscle in cross section; Gly, glycogen fields; H, hemocoel; m, mitochondria; mv, microvilli; N, nucleus; Y, yolk droplets.