

## EXTRACELLULAR MICROTUBULES IN THE APHID GUT

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As part of an investigation of a plant virus which multiplies within the cells of its aphid vector (O'Loughlin and Chambers, 1967), the fine structure of the aphid gut was studied. The alimentary canal of aphids commences with the food canal which is formed by the apposition of grooves in the two interlocking feeding stylets. The stylet food canal leads to the sucking or pharyngeal pump within the aphid head, and this carries food into the foregut. The foregut is a narrow tube which opens via the esophageal valve into the first part of the midgut, usually referred to as the "stomach." The second part of the midgut, the "intestine," follows a devious course and is the longest part of the alimentary canal. It finally widens to become the hindgut which is thin walled and highly distensible.

Specimens of *Hyperomyzus lactucae* (L.), reared on sowthistle (*Sonchus oleraceus* L.), were cut in half transversely in cold 3% glutaraldehyde and fixed for up to 14 hr, followed by postfixation in cold 1% osmium tetroxide in sodium cacodylate buffer pH 7.4. They were dehydrated in acetone and embedded in Araldite. Thin sections were stained with uranyl acetate followed by lead citrate.

The striated border of the intestine of the aphid *Myzus persicae* (Sulzer) has been reported to consist of typical microvilli (Forbes, 1964), but our observations, initially on *H. lactucae*, show that one can resolve the area surrounding each microvillus as made up of numerous extracellular microtubules that lie close to and parallel with the

microvilli (Figs. 1 and 2). The microtubules attain the same length as the microvilli, i.e. at least 1.5-2  $\mu\text{m}$ , and sometimes protrude farther into the gut lumen. The external diameter of the microtubules ranges from 14 to 16 nm, and the diameter of the less electron-opaque core is approximately 7 nm (Fig. 1). In longitudinal view, transverse striations with a periodicity averaging 6.4 nm can be observed (Fig. 4).

In the hindgut, microvilli occur less abundantly, but microtubules are consistently associated with the microvilli and frequently they are seen lying along the free surface of the hindgut cells (Fig. 3).

It is unlikely that these microtubules are formed directly by condensation from food material because they are present in embryos and in newly born aphid nymphs. Also, after adult aphids had been starved for 28 hr there was no obvious diminution in microtubule length or concentration. No microtubules were found to be associated with aphid stomach cells, which have a complex network of lamellae, or "microlabyrinth," at their free surface. This suggests that the microtubules are probably produced only by the gut epithelial cells to which they are attached, as are the surface coats of animal enteric microvilli (Fawcett, 1966). Microvilli of aphids differ in structure from those described for many other insects and for vertebrates. For example, the "brush border" of midgut cells in the flour moth *Ephesia* consists of microvilli which are very similar to those of vertebrate intestinal cells. Such types contain internal fibrils which extend through the length of the

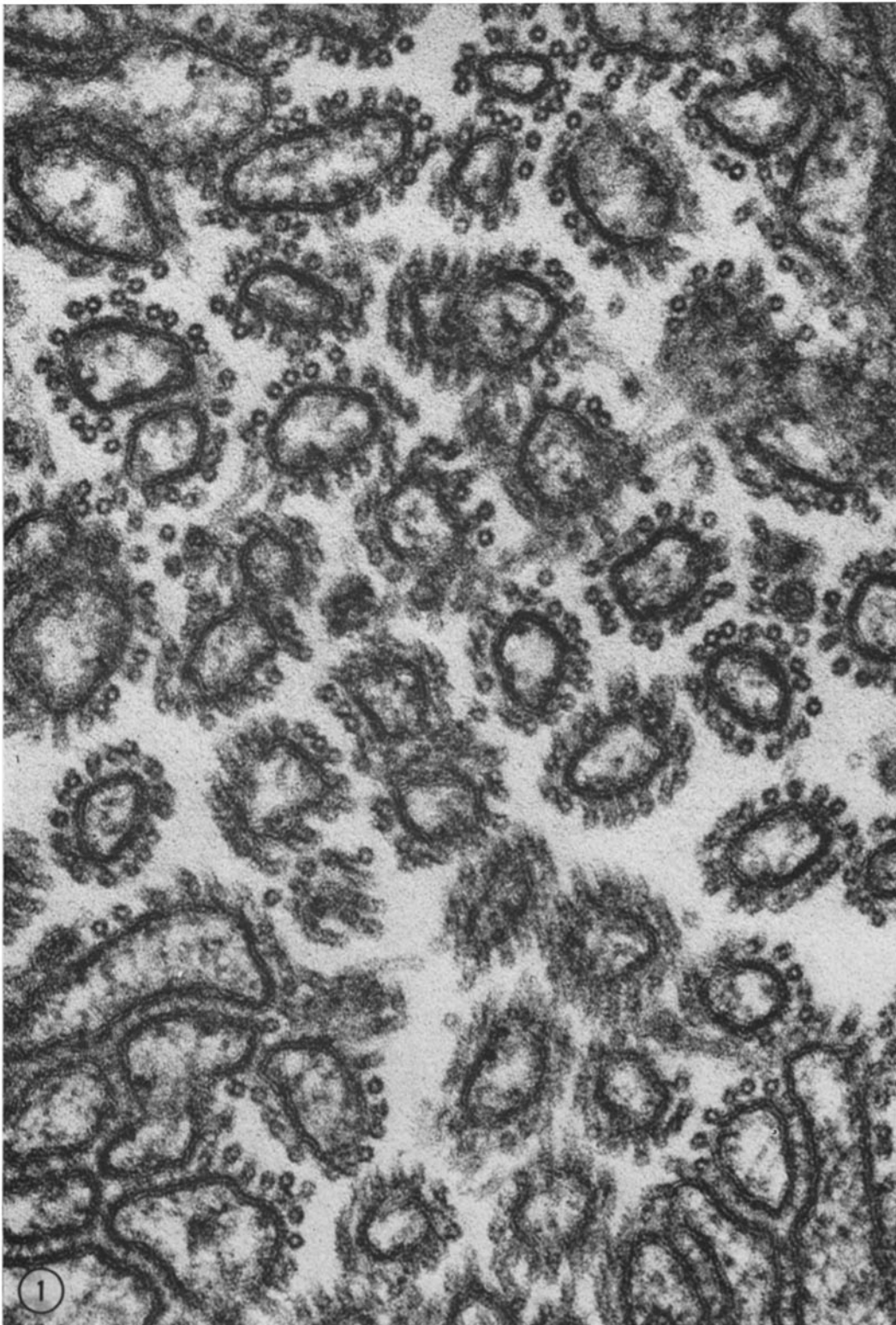


FIGURE 1 Transverse section through microvilli which project into the intestinal lumen of *H. lactucae*. Each microvillus is surrounded by microtubules.  $\times 195,000$ .

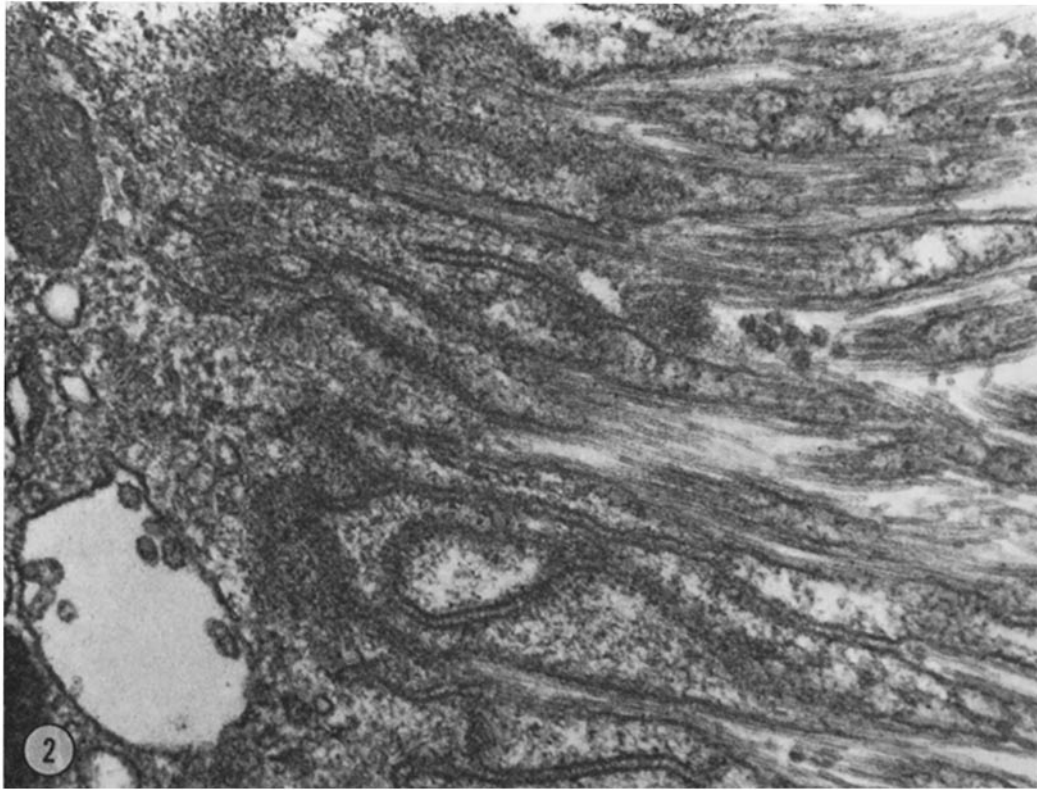


FIGURE 2 Oblique section of microvilli with associated microtubules at the free surface of an intestinal cell.  $\times 65,550$ .

microvilli and into the cell cytoplasm, and are covered by an extracellular coat or "glycocalyx" (Smith, 1968). However, in aphids we detected no internal fibrils in the microvilli, and in place of the usual glycocalyx are extracellular microtubules. It is of interest to note that the midgut of aphids is not lined by a peritrophic membrane which is present in most insects and is frequently interpreted as protecting the cell surfaces from direct contact with food material.

The function of these microtubules is unknown, but one may suspect that the vastly increased luminal surface area which they provide is associated with the efficient uptake of nutrients from the large volumes of liquid which constitutes the diet of these phloem-feeding insects. At present we have little evidence as to the true nature of these extracellular microtubules. (We have used the term "microtubule" in the broad sense.) It

seems likely that they are very different from the microtubules resolved as spindle fibers in a dividing cell and from the microtubules associated with the cytoplasm of many cell types. Further, these aphid extracellular microtubules are stable in cold fixatives, whereas many reports of intracellular microtubules suggest that they are cold labile.

Extracellular microtubules of the aphid gut are not confined to *H. lactucae* but are in all other aphid species we have so far examined—*Myzus persicae* (Sulzer), *Acyrtosiphon solani* (Kaltenbach), *Aphis craccivora* Koch, and *Macrosiphum euphorbiae* (Thomas).

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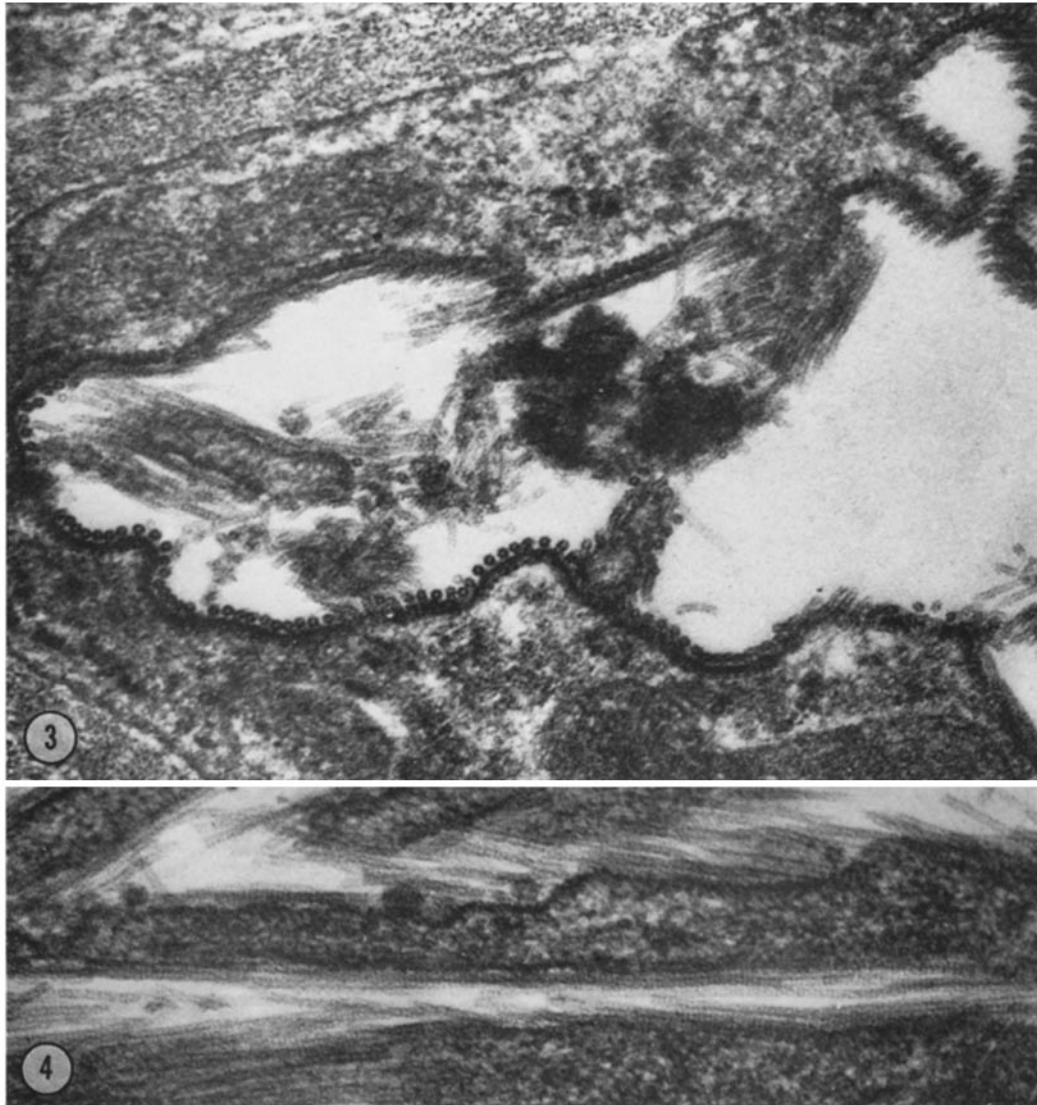


FIGURE 3 Microtubules associated with the luminal surface of a hindgut cell.  $\times 85,000$ .

FIGURE 4 Regular cross striations in microtubules of the intestine.  $\times 90,000$ .

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