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Fine structure of a sensory organ in the arista of *Drosophila melanogaster* and some other dipterans

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Summary. The arista, a characteristic appendage of dipteran antennae, consists of 2 short segments at the base and a long distal shaft. A small sensory ganglion, from which arises the aristal nerve, is located proximally in the shaft. The fine structure of the aristal sensory organ was studied in detail in the fruitfly (*Drosophila*) and for comparison in the housefly (*Musca*) and the blowfly (*Calliphora*). In *Drosophila*, the aristal sense organ consists of 3 identical sensilla that terminate in the hemolymph space of the aristal shaft, and not in an external cuticular apparatus. Each sensillum comprises 2 bipolar neurons and 2 sheath cells; a third sheath cell envelops the somata of all six neurons of the ganglion. The neurons have long slender dendrites with the usual subdivision into an inner and an outer segment. One of the outer segments is highly lamellated and bears small particles (BOSS-structures) on the outside of its cell membrane; the other outer segment is unbranched and has a small diameter. The fine structure of the first dendrite is strongly reminiscent of thermoreceptors known from the antennae of other insects. These thermoreceptors are often coupled with hygroreceptors; however, we can only speculate whether the second dendrite of the aristal organ also has this function. Our present results argue against mechanoreceptive functions, as formerly postulated. The aristal sense organs in *Musca* and *Calliphora* are similar to those in *Drosophila*, but contain more sensilla (12 in *Musca*, 18 in *Calliphora*).

Key words: Antennae – Sensory cells – Thermoreceptors – *Drosophila melanogaster* – *Musca domestica* – *Calliphora erythrocephala* (Insecta)

Dipteran flies have a typical rod-like appendage on their antennae, the arista. It is believed that the arista corresponds to the reduced distal segments of the common form of insect antenna. Since the arista arises from the third antennal segment (funiculus), the 3 subdivisions of the arista should represent antennal segments 4, 5, 6 (Ferris 1965; Postlethwait and Schneiderman 1971). In the fruitfly *Drosophila melanogaster*, a small nerve had been observed entering the arista; it was assumed to be sensory (Stocker and Lawrence 1981). However, nothing was known about the exact location, structure, and possible function of the presumed sensory organ. Previous investigations of the antennal pathway have demonstrated that the aristal nerve projects into the antennal lobes, but not to the adjacent mechanosensory center of the brain (Stocker et al. 1983, Lienhard and Stocker 1987). Since the antennal lobes predominantly receive afferents from antennal chemoreceptors, it seems more likely that the aristal nerve carries information from chemo- or hygroreceptors rather than from mechanoreceptors. A detailed fine structural study was undertaken as a first step to solving this problem.

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Materials and methods

Males of *Drosophila melanogaster* (strain Sevelen) were used. For light microscopy, the proboscis was removed from CO_2-anesthetized flies and the entire head fixed in 4% paraformaldehyde. After several hours, the heads were transferred into a 25% sucrose solution and then into carboxymethyl-cellulose before freezing in melting nitrogen (Buchner et al. 1986). Sections (10 µm thick) were cut on a cryomicrotome and usually inspected, without staining, in a phase contrast (or interference contrast) microscope. Some antennae were embedded in Epon, sectioned serially (8 µm thick) with a steel knife, and stained with toluidine blue.

For transmission electron microscopy, 2 methods were employed: chemical fixation and cryofixation. Chemical fixation started with a 5% cacodylate-buffered glutaraldehyde solution (5% sucrose added) for 6 h and was followed by a 1% OsO_4-fixation (same buffer) for 2 h. In order to facilitate penetration of the fixative, the distal half of each arista was cut using iridectomy scissors. After dehydration in an ethanol series and infiltration with Epon, the antennae were carefully removed from the heads and oriented in flat-embedding molds before polymerization at 65° C. For cryofixation, freshly decapitated heads were pinned on thin tungsten wire (0.1 mm) and rapidly injected (2–3 m/s) into liquid propane (−180° C). They were then transferred to liquid nitrogen and later freeze-substituted in acetone containing 2% OsO_4 for 3 days (−80° C). After slowly warming to room temperature (overnight), entire heads or isolated antennae were flat-embedded in Epon (Steinbrecht 1985). Thin sections were cut with a diamond knife, picked up on Formvar-coated slot grids and double-stained with uranyl acetate and lead citrate. Sections were inspected in a Philips 300 or Zeiss 10A electron microscope (fitted with

a goniometer stage) at 80 or 100 kV. Two cryo-fixed aristae were cut in serial cross-sections and photographs were taken systematically at intervals of approximately 1 μm, thereby allowing the reconstruction of entire aristal sense organs. A dozen chemically fixed aristae were sectioned in transverse and longitudinal planes for studying selected points of interest. Aristae of larger dipterans (*Musca domestica*, *Calliphora erythrocephala*) were similarly prepared for electron microscopy, but only chemical fixation was used.

For scanning electron microscopy, several *Drosophila* heads were fixed in 4% formaldehyde and 5% glutaraldehyde before dehydration in alcohol and acetone. After critical point drying, they were coated with gold and examined in a Hitachi S-700 SEM at 25 kV.

Results

External morphology of the arista in Drosophila

The aristae emerge postero-laterally from the third antennal segment (funiculus), close to the joint with the second segment (pedicellus; Figs. 1, 2). The aristal shaft points strongly laterally (20–30° off the horizontal axis toward anterior) and slightly downward. The entire arista is about 300 μm long and 10–20 μm in diameter (Fig. 2). Each arista consists of three parts: the base begins with a small ring (5 μm high, 20 μm in diameter; Fig. 3), the following segment forms a stout cylinder (30–35 μm long, 20 μm in diameter), and the long aristal shaft represents the last segment

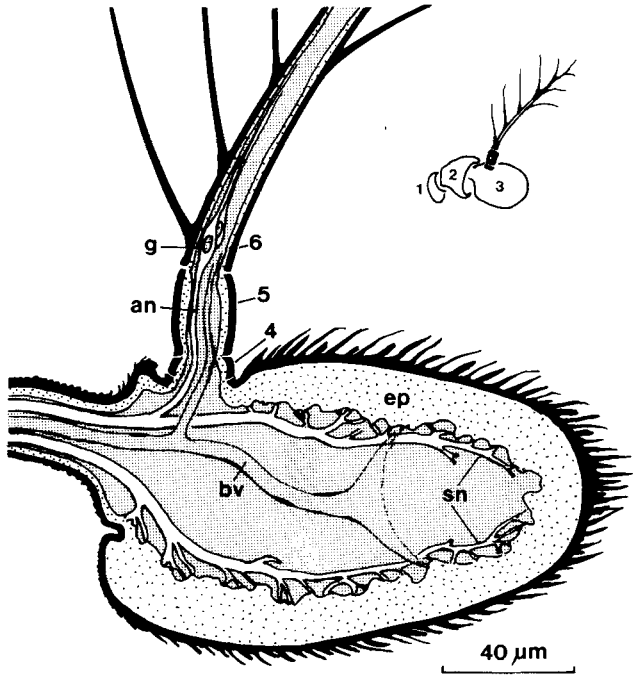


Fig. 1. Diagrammatic longitudinal section of the third antennal segment (funiculus, *3*) in *Drosophila*. The arista is located postero-laterally and consists of 3 parts that are connected by joint membranes: an annular basal segment (*4*), a stout cylinder (*5*), and a plumose terminal shaft (*6*). At the base of the shaft lies a small sensory ganglion (*g*), from which the aristal nerve (*an*) arises; it joins one of the two sensory nerves (*sn*) coming from the sensory epithelium (*ep*). The blood vessel (*bv*) supplying the funiculus ends like a funnel, but gives off a branch that extends into the aristal lumen

(10 μm in diameter) and gradually tapers toward the tip. This shaft is plume-shaped (Figs. 2, 3), with six long (100 μm) cuticular processes dorsally and 3 processes ventrally; additionally, short spines project from both sides of the aristal shaft. These processes and spines do not correspond to bristles but apparently represent simple trichomes (Peyer and Hadorn 1965).

All 3 aristal segments are connected by joint membranes (Fig. 3). These specialized cuticular areas stain strongly with certain dyes (e.g., methylene blue; fluorescent with Lucifer Yellow); in electron micrographs, they appear lighter than the surrounding cuticle. Although these joint membranes certainly allow for movement between the segments, the arista as a whole is relatively rigidly connected to the funiculus. Slight blowing at the arista through a micropipette always moves the funiculus with respect to the pedicellus, without affecting the aristal segments themselves. Indeed, a very gentle head-on air stream is sufficient to deflect the arista further laterally, thereby rotating the entire funiculus. Only with stronger air currents does one elicit a bending of the arista at the base.

Histology

The cuticle of the arista is very thin, measuring only 1 μm or less. The underlying epidermal cells form a conspicuous epithelium (6–7 μm) in the two basal aristal segments, but only a thin lining (0.1–0.2 μm) inside the shaft (Figs. 5, 8). The aristal lumen is filled with hemolymph and communicates with the large lumen of the funiculus. A side branch of the antennal blood vessel enters at the base of the arista (Fig. 4) and becomes attached to the epidermal cells; it can be followed well into the shaft, where it seems to end openly.

A fine nerve branch, the aristal nerve, can be seen running in parallel to the small blood vessel. It arises from an inconspicuous sensory ganglion, that is situated at the base of the aristal shaft (Fig. 4). This ganglion belongs to an aristal sense organ, which will be described in the following section.

Fine structure

The aristal sense organ consists of 3 sensilla, each of which comprises 2 neurons and 2 sheath cells. An additional sheath cell (neuroglial cell, s3; Fig. 23) is shared by all 3 sensilla. The neuronal somata, together with their cover of 3 sheath cells, form a small elongated ganglion.

All 3 sensilla are structurally identical, but they do not lie exactly in register; this means that their dendrites end at different heights within the arista shaft. It is a special feature of these sensilla that neither the perikarya, nor the dendritic endings, are integrated into (or attached to) the epidermis. Instead, all sensilla lie freely in the hemolymph space, where they also terminate. In the following, we will focus on the cellular organization of the aristal sensilla.

(1) Sensory cells. The somata of the 2 bipolar neurons are about 10 μm long and 3 μm wide; the long and slender dendrites extend about 60 μm distally (Fig. 23). Both dendrites have a typical ciliary region at the same level (about 10 μm from the tip), which marks the transition from the inner (IS) to the outer dendritic segment (OS). One of the OS is small (0.25 μm diameter) and cylindrical proximally,

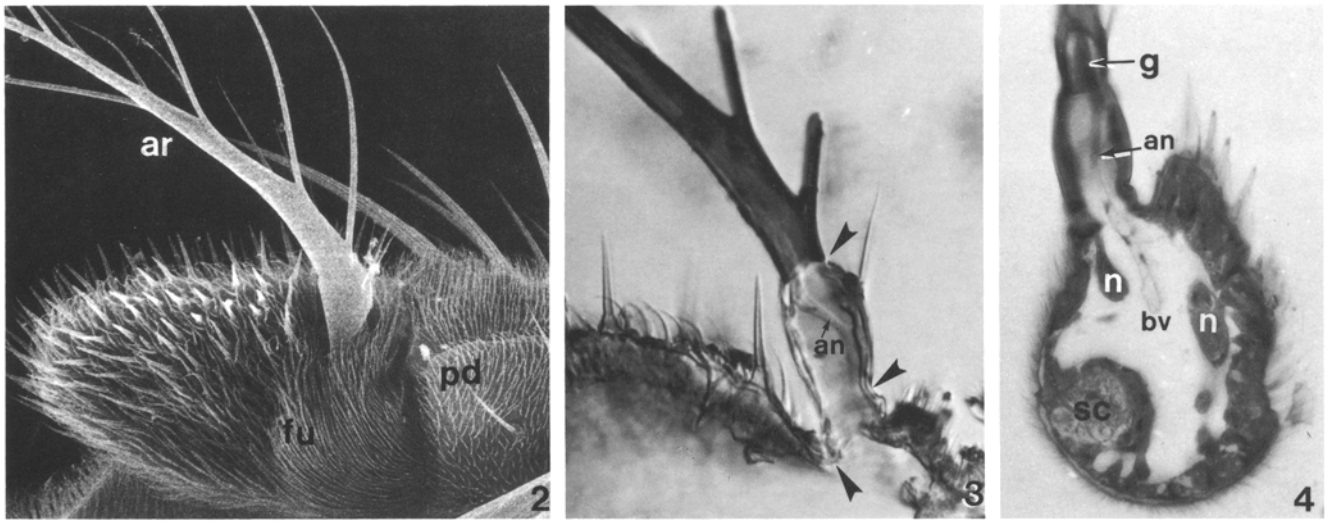


Fig. 2. Posterior aspect of antenna showing the pedicellus (*pd*) and the funiculus (*fu*) with the plumose arista (*ar*). The pedicellus bears a few large mechanosensitive hairs, whereas the funiculus is covered with hundreds of chemoreceptive sensilla. × 370 (SEM)

Fig. 3. Longitudinal section of the aristal insertion showing the aristal nerve (*an*) and the segments with their articulating membranes (*arrow heads*). This unstained section demonstrates that only the plumose aristal shafts is dark-pigmented. × 550 (interference contrast)

Fig. 4. Cross-section of the funiculus at the level of the aristal insertion. A small sensory ganglion (*g*) is located at the base of the aristal shaft. The aristal nerve (*an*) can be seen running toward one of 2 small nerves (*n*) inside the funiculus. A delicate blood vessel (*bv*) extends into the arista; *sc* sacculus, a presumed sense organ. × 450

but flattened distally; the other OS is larger (1 μm diameter) and highly lamellated.

(a) Outer dendritic segment (OS). Both OS are encased by an electron-dense dendritic sheath and a thin layer of one sheath cell distally (Figs. 5, 6), or 2 sheath cells more proximally (Fig. 10).

Lamellated OS. The plasma membrane of the bigger dendrite (d1) forms whorls of very thin lamellae (10–30 nm; Figs. 6, 7). Their arrangements is rather complex and can be compared with the pages of a paperback book, in which the pages (lamellae) are rolled in a spiral fashion. Often 2 lamellae originating from the dendritic stem form a right-handed and a left-handed spiral, respectively, which interdigitate (Figs. 7, 9). It seems that the lamellation results from a fusion of intracellular vesicles (Fig. 24), which are aligned in a spiral course. In some sections rows of such vesicles in various degrees of fusion can be seen spiraling toward the dendritic surface, where they open to the extracellular space of the so-called inner receptor lymph cavity (Fig. 9; see below).

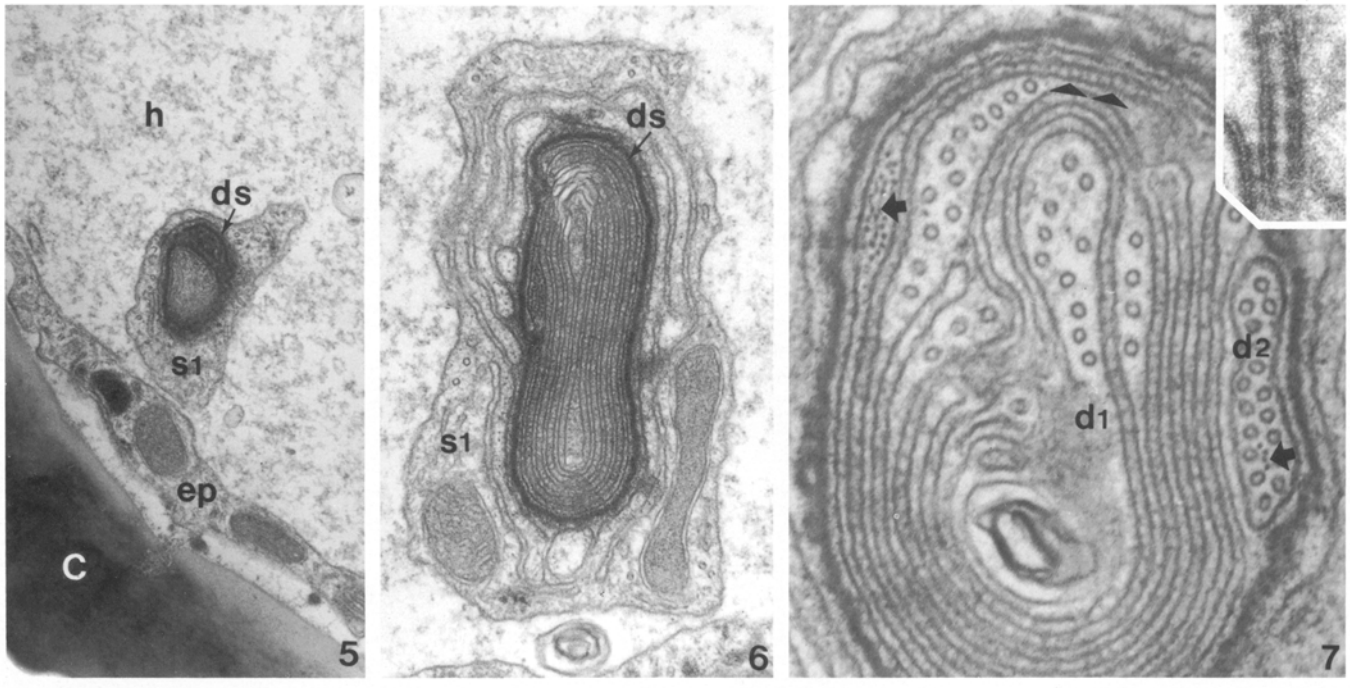
The extent of lamellation is variable but is most pronounced near the tip of the OS (Figs. 6, 7). In this distal region, the lamellae are almost devoid of cell organelles, whereas at more proximal levels, they may contain a row of microtubules and sometimes clusters of intermediate filaments (Fig. 7). The interspace between 2 neighboring lamellae appears regular and electron-dense. Periodically arranged particles can be noticed protruding from the plasma membrane into the extracellular space (Fig. 7). Cross sections of the dendrites must be tilted by 20–30° in order to make these particles visible; thus their orientation appears to be oblique with respect to the longitudinal axis of the dendrites. These particles closely resemble the small knobs (BOSS-structures) covering the OS membrane of a lamellated insect thermoreceptor (Steinbrecht 1989).

The second dendrite (d2) is slightly shorter than the lamellated d1, but its diameter is less than one third of the former. Near its distal end, it is laterally compressed and contains 1–2 rows of microtubules and also some intermediate filaments (Fig. 7); proximally, d2 assumes a more cylindrical shape with a diameter of 0.2–0.3 μm. Both OS are encased by a distinct irregularly shaped dendritic sheath and 2 enveloping cells (s1, s2). The space inside the dendritic sheath is termed the inner receptor lymph cavity. Most of it is occupied by the 2 OS, except near the ciliary region, where this extracellular space becomes more prominent (Fig. 10).

At the level of the ciliary regions both dendrites measure about 0.3 μm in diameter (Fig. 11). This extremely short region exhibits the typical $9 \times 2 + 0$ pattern of microtubules, which arises from a centriole structure.

b) Inner dendritic segment (IS). The initial part of the IS increases immediately in size and contains many clear vesicles, mitochondria, microtubules, and ribosomes. Again, one can distinguish a larger (1.5 μm diameter) and a smaller profile (0.8 μm diameter; Figs. 12–14). From serial sections, it is evident that the smaller IS-profile corresponds to the smaller OS and the bigger IS-profile to the larger lamellated OS. Both IS retain their diameter over their entire length (about 50 μm) before gradually expanding into their somata.

(c) Somata. The somata are elongated and of small diameter (2–3 μm). The ovoid nucleus appears light due to the finely dispersed chromatin. The cytoplasm forms only a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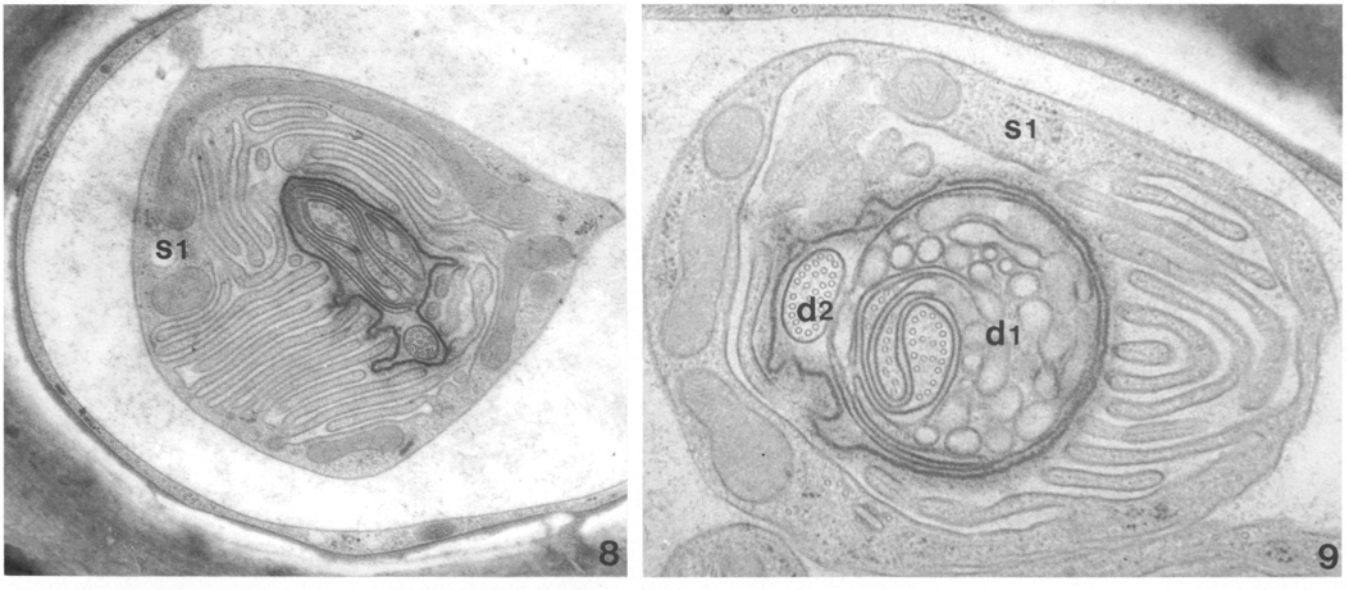
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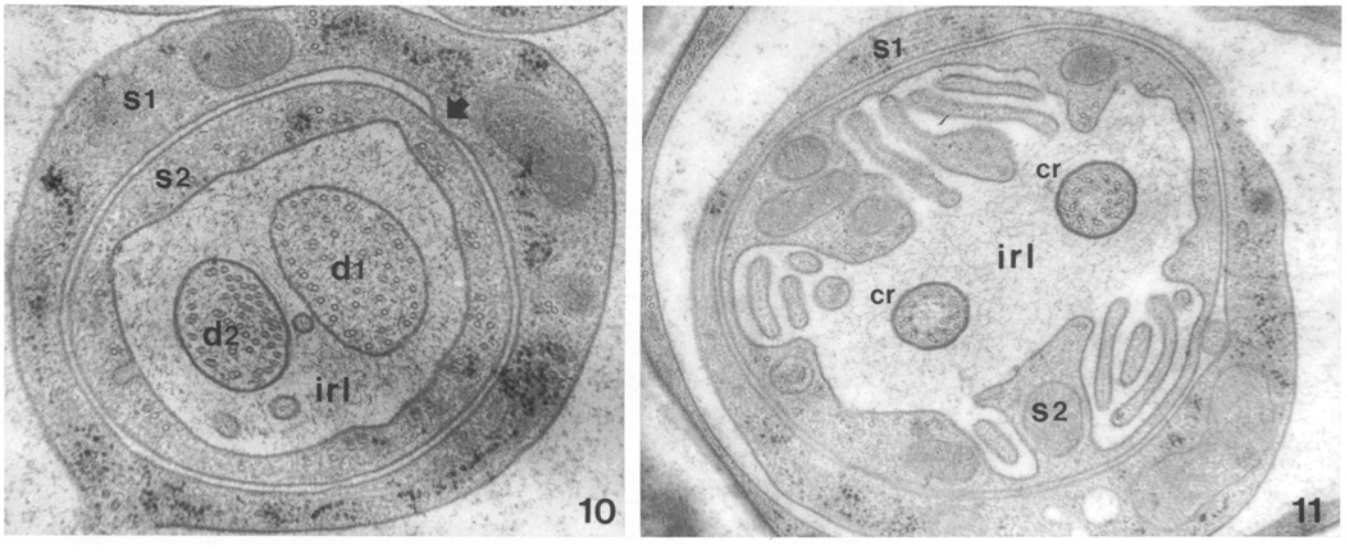
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thin layer around the nucleus and is also electron-lucent (Figs. 15, 16). Typical cell organelles are small dark mitochondria, Golgi elements, many free ribosomes and short portions of rough endoplasmic reticulum.

(d) Axons. The axons emanating from the 6 sensory cells give rise to the aristal nerve, together with a third sheath cell (s3) and an intervening glial cell (s4; Figs. 16, 17). An extensive extracellular space exists between the axons and the thin processes of the glial cell. The axons themselves are very delicate, measuring only 0.2–0.4 μm in diameter. Further proximally, near the base of the arista, the aristal nerve makes contact with the luminal surface of the epithelial cells. After entering the funiculus, it merges with one of the 2 sensory nerves exiting from this segment (Fig. 1).

(2) Sheath cells. It has already been mentioned that several types of sheath (or enveloping) cells (s1–s4) are associated with the aristal sense organ (Fig. 23). Each neuron has its own sheath cell 1 and 2, whereas sheath cell 3 is shared by all 6 neuronal somata. A fourth enveloping cell (s4) is probably a glial cell of the aristal nerve; it extends up to the axon hillocks of the sensory cells. All sheath cells appear relatively darker (cytoplasm and nuclei) than the sensory cells.

Sheath cell 1 (s1). This cell covers the OS and ends shortly below the ciliary region. Distally, it forms only a thin layer around the dendrites (Fig. 6), but more proximally it becomes separated from the dendritic sheath, thus creating a large extracellular space. Extensive microplicae originate from the periphery and project into this space (Figs. 8, 9). These plicae bear characteristic particles ("portasomes", Harvey 1980) on the inside of their plasma membrane and often exhibit fine filaments (possibly actin) in the center of each plica. More proximally, i.e., close to the ciliary regions, the microplicae become gradually reduced and the dendritic sheath ends. At the same level, the second sheath cell (s2) appears, enclosing the proximal parts of the OS. S1 no longer encircles the OS but withdraws to one side of the dendrites (Figs. 12, 13); this is also the site where the nucleus of s1 is located. Some proximal extensions of s1 detach completely from the sensilla and terminate on or near the epithelium beneath the cuticle of the aristal shaft.

Sheath cell 2 (s2). Distally, this cell penetrates for a short distance between the dendritic sheath and sheath cell 1. Since batches of dendritic sheath material can be seen adhering to the surface of the cell membrane of s2, it seems that this cell produces the dendritic sheath. Thus, s2 most likely represents the so-called thecogen cell (or dendritic sheath cell). Most of the IS is covered by s2 only. Like sheath cell 1, s2 exhibits a distinct extracellular space and elaborate microplicae, which gradually disappear more proximally. There is no connection between the extracellular space enclosed by s1 and that formed by s2; the latter space is the inner receptor lymph cavity (Figs. 10, 11).

Sheath cell 3 (s3). Shortly before the IS reach their neuronal somata, a third sheath cell (s3) squeezes between the dendrites and s2 (Fig. 14). This cell encases not only the initial IS but also the somata and the axon hillocks of all 6 sensory neurons. It only forms a thin cover and never exhibits microplicae. From its location, it seems reasonable to classify it as a neuroglial cell.

Sheath cell 4 (s4). In the middle of the sensory ganglion, fine processes of a further sheath cell (s4) appear, which loosely surround somata and axon hillocks (Fig. 16). This cell probably represents a glial cell of the aristal nerve.

(3) Cell contacts. The following cell junctions can be observed in the aristal sense organ: (1) desmosomes, (2) septate junctions, and (3) close contacts.

Desmosomes are typically seen between the distal IS and longitudinal ridges of the surrounding s2 (Fig. 12). The dense membrane apposition in s2 is backed by a few microtubules that are connected to a thin electron-dense platelet on the desmosomal side. Occasionally, desmosomes may occur between the IS of d1 and d2. Some cell processes of s2 also make contact with the epithelial surface via desmosomes, thereby providing some attachment of the dendrites within the aristal lumen.

Septate junctions are common between the IS and s2, as soon as the IS become completely engulfed by this sheath cell. The same type of junction was also noted between sheath cells, e.g., between s1 and s2. It should be mentioned that typical septae are only seen after chemical fixation; in cryo-fixed specimens, the intercellular cleft is filled with

Figs. 5–11. Consecutive cross-sections (distal to proximal) of aristal sensilla (**Figs. 5–7** chemofixation, **Figs. 8–11** cryofixation)

Fig. 5. The very tip of an aristal sensillum is covered by a dendritic sheath (*ds*) and a sheath cell (*s1*). It lies freely in the hemolymph (*h*) of the aristal shaft, but may be connected to the epidermis (*ep*) via extensions of *s1*. *C* cuticle. ×32000

Fig. 6. The tip region contains a lamellated dendritic outer segment inside a dendritic sheath (*ds*), which in turn is surrounded by several lamellae of *s1*. ×45000

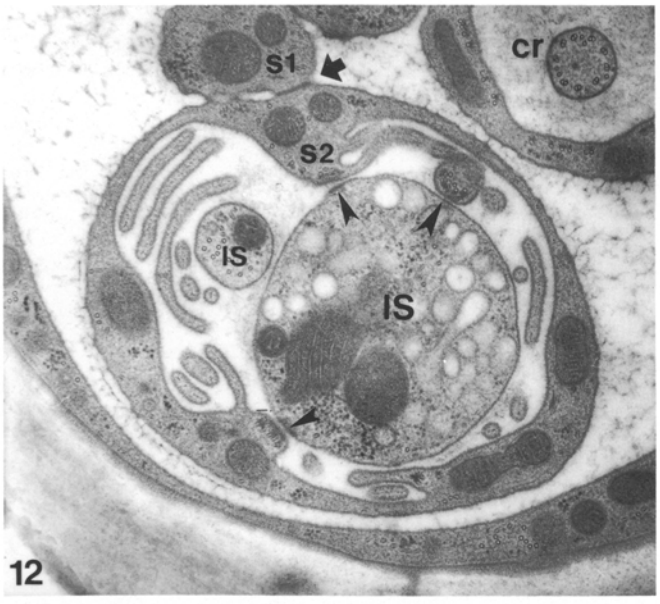
Fig. 7. Higher magnification of the tip region at a slightly more proximal level: most of the space inside the dendritic sheath is occupied by the lamellated dendrite (*d1*), but a smaller dendrite (*d2*) is visible at the periphery. Note the microtubules and microfilaments (*arrows*) in both dendrites. The thin lamellae of d1 are devoid of cell organelles but bear a regular array of particles on the outside of their plasma membrane (*arrowheads*). ×97000. Inset: These membrane particles (BOSS-structures) become better visible after 20–30° tilting of the section. ×160000

Fig. 8. Sheath cell 1 (*s1*) shows extensive microplicae in its distal portion. ×23000

Fig. 9. The lamellation of *d1* is apparently the result of fused intracellular vesicles; *d2* becomes more cylindrical and remains of small diameter. ×38000

Fig. 10. Slightly above the ciliary region the dendritic sheath and the lamellation of *d1* end. A second sheath cell (*s2*) appears that encloses the inner receptor lymph cavity (*irl*). Note close membrane contacts between *s1* and *s2* (*arrow*). ×45000

Fig. 11. The 2 dendrites have the same small diameter (0.3 μm) at the level of their ciliary region (*cr*). Microplicae of *s2* project into the inner receptor lymph cavity (*irl*). ×34000



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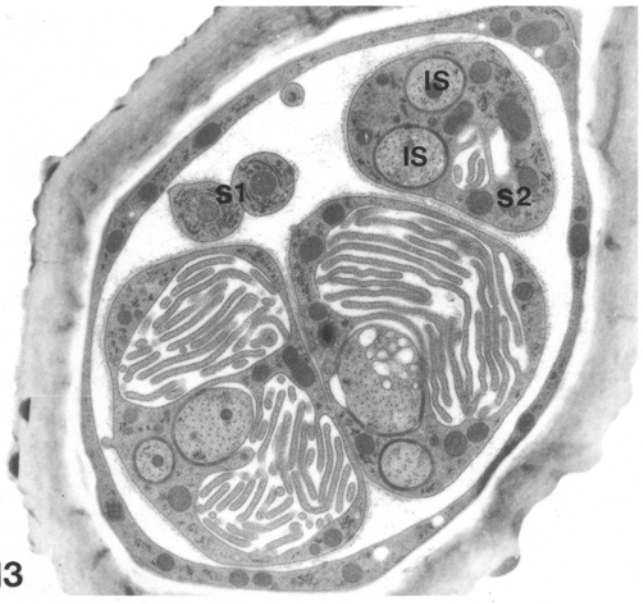

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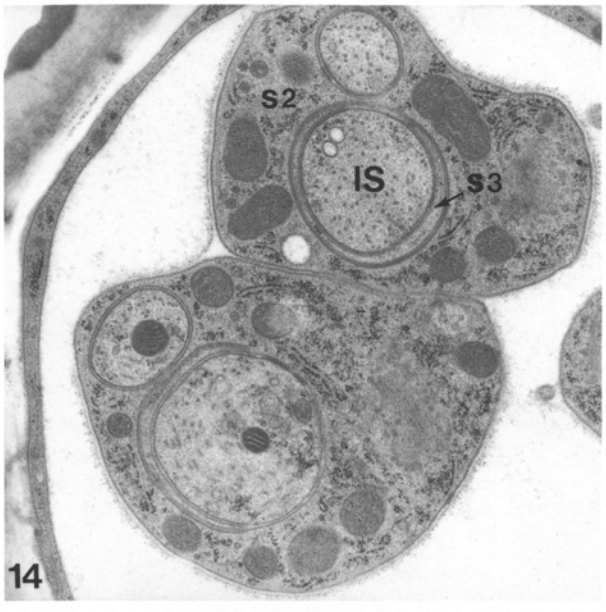

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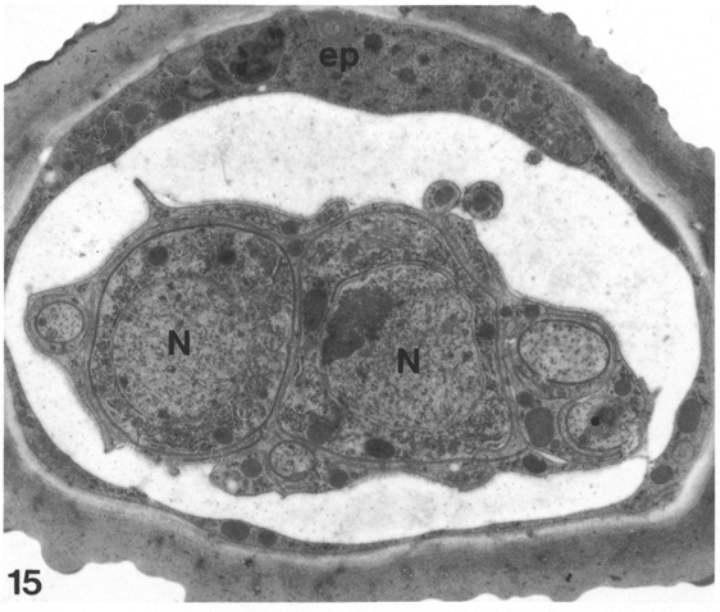

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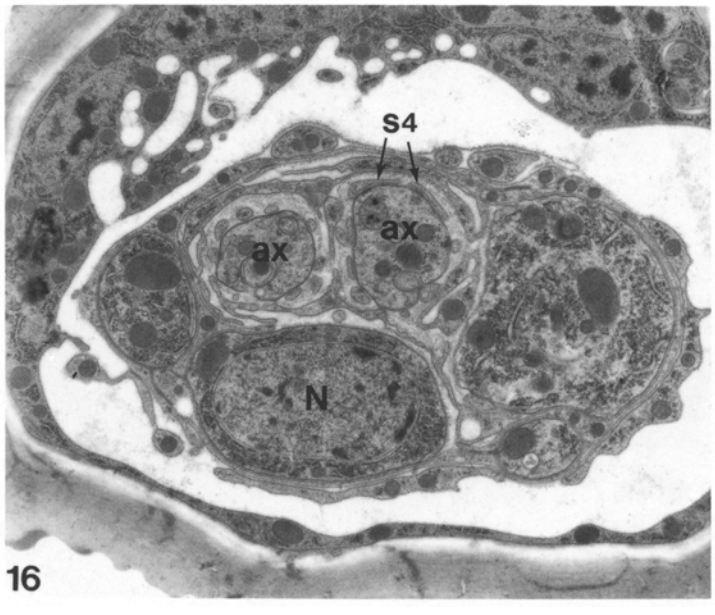

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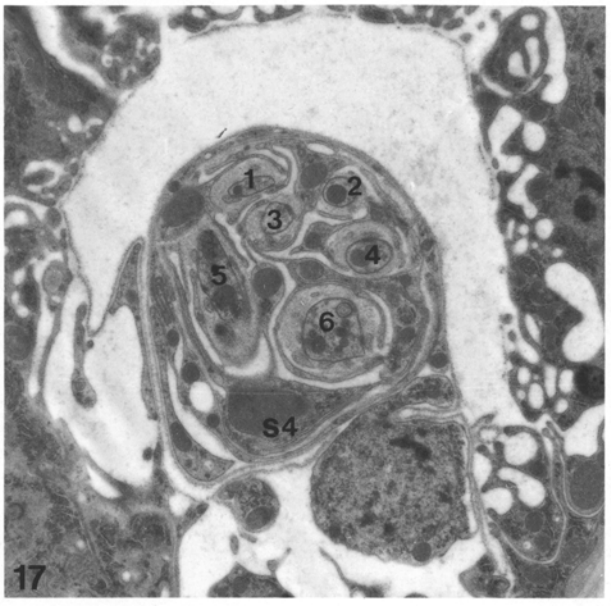

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an electron-dense material that is evenly distributed and that rarely forms discernible septae.

Close contacts appearing like gap junctions but extending over longer distances are very rare between the dendrites (OS and IS), but are usual between sheath cells, e.g., s1 and s2 (Figs. 10, 12) or between two adjacent s2 cells.

The aristal sense organ in other dipterans

After having established the presence of a specific sensory organ in the arista of *Drosophila*, it seemed of interest whether the same type of sensilla exists in other dipteran flies. A few large representatives (*Calliphora erythrocephala, Musca domestica*) were chosen for comparison. Due to the much greater size of their aristae, only specific regions were selected for examination. The 2 aspects that received particular attention were (1) the number of axons in the aristal nerve, and (2) the fine structure of the dendritic outer segments.

In *Calliphora*, the aristal nerve and the accompanying blood vessel are easily recognizable under the light microscope (Fig. 18). The bipolar neurons do not form a discrete ganglion as in *Drosophila* but extend over a long distance distad into the aristal shaft. Cross-sections of the aristal nerve exhibit 36 axons in *Calliphora* and 24 in *Musca* (Figs. 19, 20). This means that larger aristae also have a higher number of aristal sensilla. Furthermore, the larger aristal nerves are always provided with a tracheole, in contrast to *Drosophila* were the 6 axons of the aristal nerve receive no tracheal supply.

With respect to their dendritic OS, the aristal sensilla in both *Calliphora* and *Musca* show similar features to those observed in *Drosophila*, i.e. 2 profiles inside a dense dendritic sheath (Figs. 21, 22). One of the OS is typically lamellated, whereas the other is straight and unbranched, yet of the same diameter as the first. The surrounding sheath cells exhibit extensive microplicae with portasomes (Fig. 21, inset). In all, the basic design of the aristal sense organ in *Calliphora* and *Musca* resembles that found in *Drosophila*, but appears more complex, because of the larger number and diffuse arrangement of aristal sensilla.

Discussion

Initially, we expected to find some kind of mechanoreceptor, e.g. a chordotonal organ, in the arista of *Drosophila* (Stocker and Lawrence 1981). However, backfill experiments of the aristal nerve showed that the axons do not project to the mechanosensory center in the CNS. Instead, they end in specific glomeruli (VP2, VP3) in the antennal lobes, which receive mostly chemosensory afferents (Stocker et al. 1983). The present fine structural study of the aristal sense organ offers a new interpretation: neurons with highly lamellated dendrites are indicative of a thermoreceptive function (for review see Altner and Loftus 1985; Altner and Prillinger 1980; Steinbrecht 1984). Lamellated outer dendritic segments have been described for antennal sensilla of beetles (Corbière-Tichané 1971, 1974, 1977), dipterans (Chu-Wang et al. 1975; McIver 1973; McIver and Siemicki 1976, 1979), the stick insect (Altner et al. 1978), the bed bug (Steinbrecht and Müller 1976), and for silk moths (Haug 1985; Steinbrecht 1989). Most of them have been shown to be thermo/hygroreceptors by means of electrophysiological recordings (Altner and Loftus 1985; Steinbrecht 1989) and there is now good evidence that the lamellated dendrite is responsible for thermoreception (B. Zimmermann and H. Altner, in prep.). The lamellated dendrites of dipterans (this study) and those described in the silkmoth correspond even in fine structural details, i.e. the particle-studded membranes of the lamellae (BOSS-structures, Steinbrecht 1989) are present in both cases. Since these particles have not been found in any other receptor membranes, they might be modality-specific for thermoreception.

There is an interesting relationship between the lamellae and intracellular vesicles, that has not been reported before. Many micrographs suggest that several vesicles fuse to form a lamella (Fig. 9). Indeed, an analysis of serial sections has revealed that vesicles are clustered on both ends of a given lamella and several vesicles could be traced to be confluent with the lamella (Fig. 24).

So far, lamellated dendrites have only been observed in 3 neurons of the *Drosophila* antennae, but not in any of its other appendages. One might argue that they have simply been overlooked, but this seems unlikely, since the lamellae are quite conspicuous structures. If they were present on the funiculus, even in very low numbers, then one would expect to see projections from the funiculus to glomeruli VP2 and VP3 in the antennal lobes. However, backfill experiments from the funiculus have never shown any terminations in glomeruli VP2 and VP3 (Stocker et al. 1983). Perhaps it is sufficient to have just a few thermoreceptors, provided that they are located at strategic points. The well-exposed, thin-walled (1 μm) arista would certainly fulfill that condition. Lamellated dendrites may also occur in chemoreceptive sensilla, e.g., in CO_2-receptors of butterfly labial palps (Lee et al. 1985; Bogner et al. 1986). In

Figs. 12–17. Consecutive cross-sections of aristal sensilla (from the IS to the axon level). Cryofixation

Fig. 12. Just below the ciliary region (*cr*) the 2 dendritic inner segments (*IS*) again assume different calibers. Note the larger number of vesicles inside the larger IS and desmosomal contacts (*arrow heads*) between *IS* and *s2*. Close membrane contacts are seen between sheath cells 1 and 2 (*arrow*). × 29500

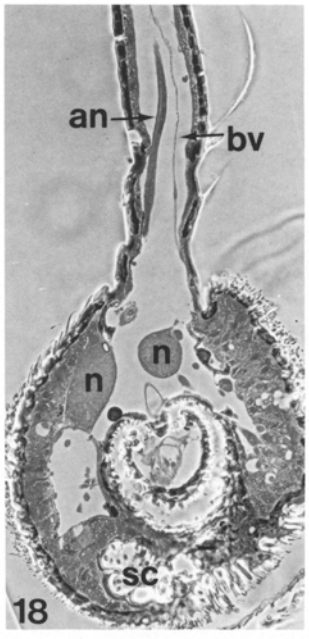
Fig. 13. Further proximally, the *IS* become first surrounded by extensive microplicae of *s2* (two lower sensilla) and are later engulfed by *s2* (uppermost sensillum). Only remnants of sheath cell 1 (*s1*) are present at this level. × 12000

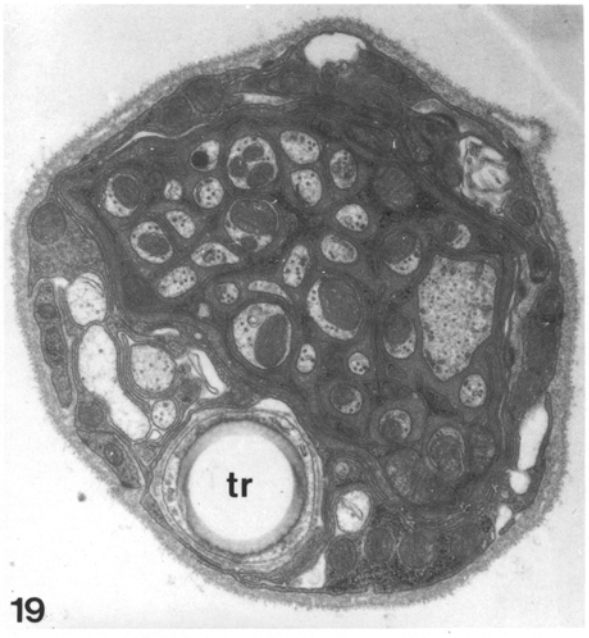
Fig. 14. Shortly after both *IS* have become enclosed by *s2*, a third sheath cell (*s3*) appears, which wraps around the *IS* and somata of all neurons. × 18000

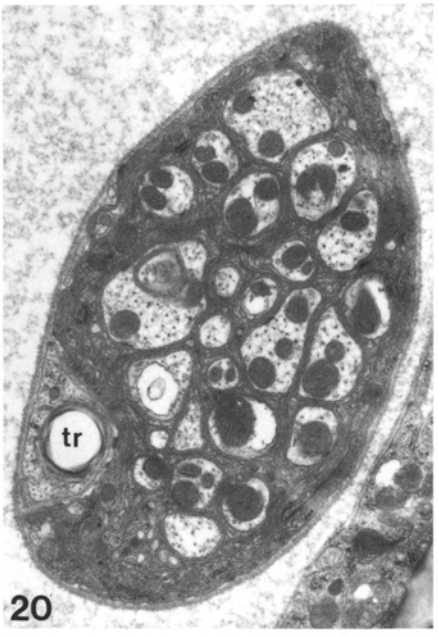
Fig. 15. The nuclei (*N*) of the bipolar neurons lie at the base of the aristal shaft. The epidermis (*ep*) lining the arista increases in thickness when entering the cylindrical aristal segment. × 11000

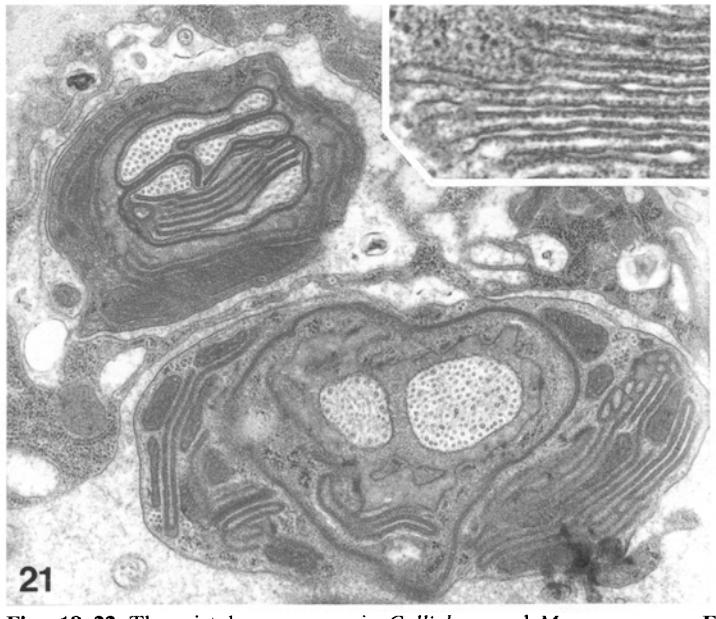
Fig. 16. Two sensory cells sectioned at their axonal level. Each axon (*ax*) is surrounded by fine processes of a fourth sheath cell (*s4*). The thicker epidermis shows a typical basal labyrinth (cf. Fig. 19). × 12500

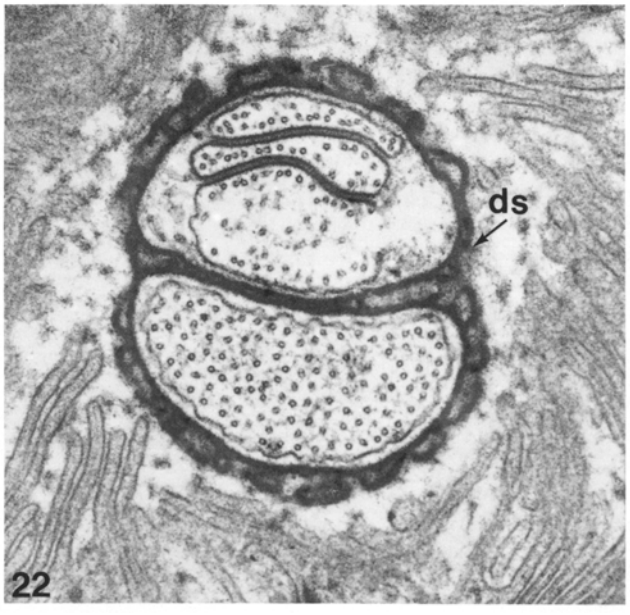
Fig. 17. The aristal nerve consists of 6 small axons (1–6), which are loosely surrounded by long extensions of s4. × 10000











Figs. 18–22. The aristal sense organ in *Calliphora* and *Musca*

Fig. 18. *Calliphora.* Cross-section of the proximal funiculus, showing the arista in longitudinal section. The aristal nerve (*an*) and a blood vessel (*bv*) can be seen entering the aristal shaft. The small aristal nerve will join one of the 2 larger nerves (*n*) in the dorsal part of the funiculus; *sc* sacculus. × 200 (phase contrast)

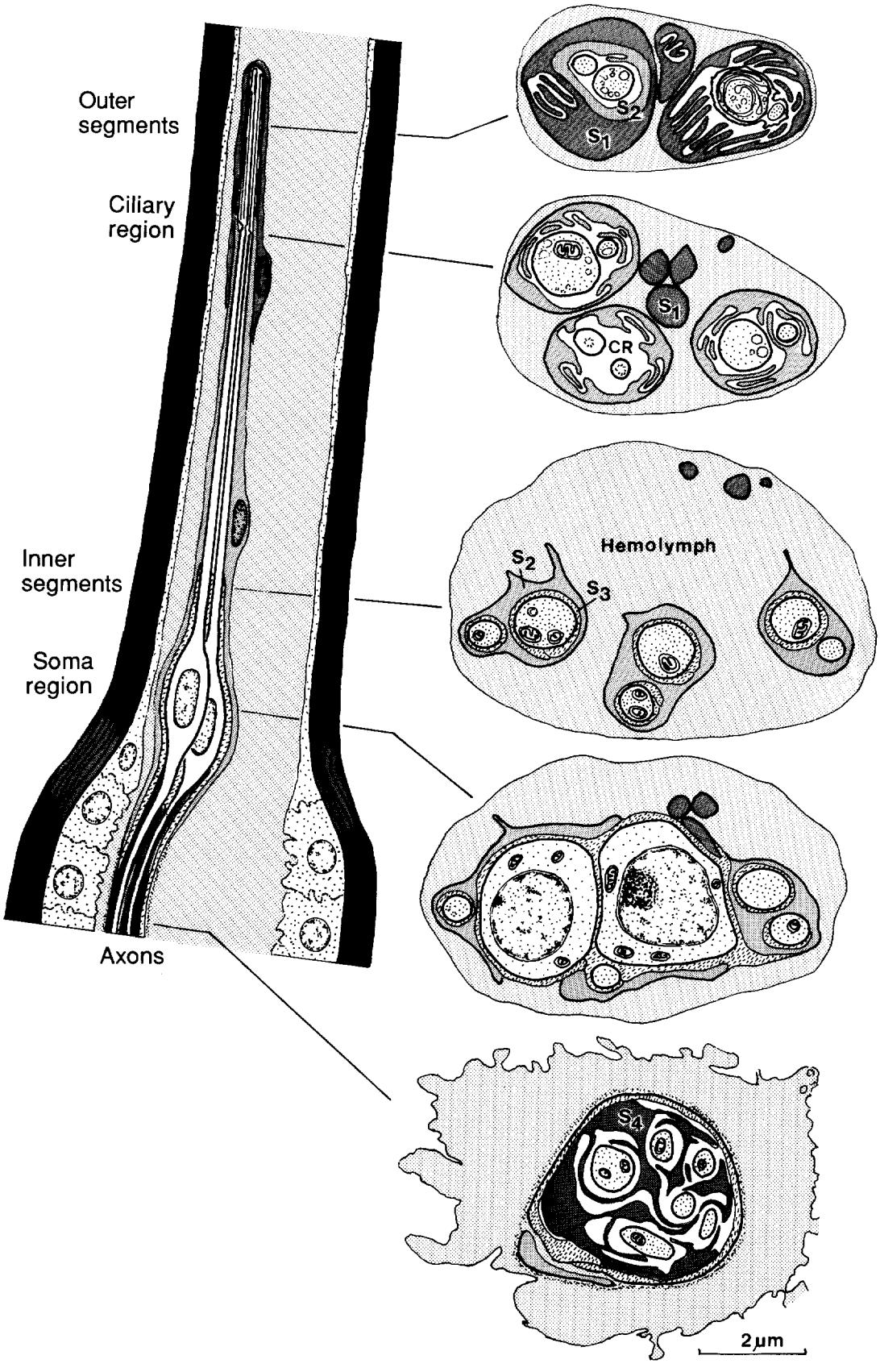
Fig. 19. *Calliphora.* Cross-section of the aristal nerve. About 36 small axons can be counted within the glial meshwork. A tracheole (*tr*) is present at the periphery of the aristal nerve. × 17000

Fig. 20. *Musca.* Cross-section of the aristal nerve. About 24 axons, glial lamellae, and a small tracheole (*tr*) can be recognized. × 12000

Fig. 21. *Calliphora.* Cross-section of 2 sensilla through their outer segments. Distally (upper sensillum), a dendritic sheath encloses a lamellated and a non-lamellated OS; more proximally (lower sensillum), the dendritic sheath has disappeared and 2 sheath cells become visible. × 20000 *Inset:* The microplicae of the sheath cells bear typical membrane particles (portasomes) on their cytoplasmic leaflet. × 62000

Fig. 22. *Musca.* Cross-section of the OS of 1 aristal sensillum. The 2 dendrites are of equal size but only the upper one shows some lamellation. Note that the dendritic sheath (*ds*) encloses both dendrites separately. × 38000

Fig. 23. Summarizing diagram of the aristal sensory organ in *Drosophila.* On the *left*, a longitudinal section of 1 aristal sensillum is shown. The soma region lies at the border of the cylindrical aristal segment and the distal shaft. The dendrites extend into the hemolymph space of the shaft; they are subdivided by a ciliary region into inner and outer segments. On the *right*, cross-sections of the arista are represented, showing the arrangement of the sensory cells and 4 sheath cells (*s1–s4*). *CR* ciliary region


Outer segments
Ciliary region
Inner segments
Soma region
Axons
S2
S1
S1
CR
Hemolymph
S2
S3
S4
2µm

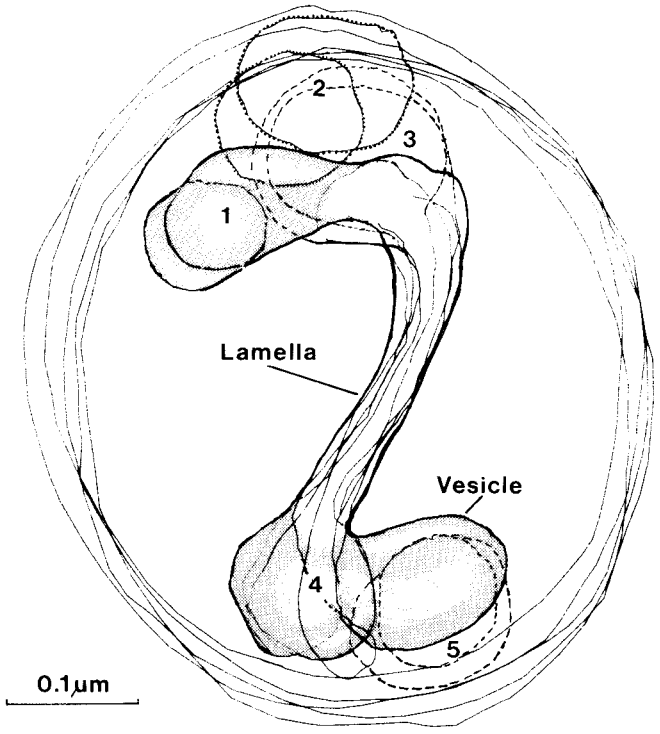


Fig. 24. Relationship between dendritic lamella and intracellular vesicles: several vesicles gather around each end of a lamella; vesicles 1 and 4 are stippled to show their continuity with the lamella. (Superposition of 5 serial cross sections of a dendritic outer segment)

these cases, however, the dendrites protrude into a porous cuticular peg, which is typical of insect olfactory sensilla.

Although there is good reason to interpret the lamellated dendrite d1 as a thermoreceptor, the function of the small, non-lamellated dendrite d2 remains uncertain. There are no structural specializations that would indicate mechanoreception (tubular bodies, special attachment to cuticle) or chemoreception (pores in the cuticle). In analogy to the thermo/hygroreceptors mentioned above, one could postulate hygroreception for d2. Yet hygroreceptive dendrites are usually tightly enclosed by cuticular pegs (Altner et al. 1983; Yokohari 1981, 1983) and this is not the case in the aristal sense organ.

The aristal sensilla are quite unusual when compared to most other types of insect sensilla: they lie freely in the hemolymph of the arista, without any direct association with epidermis or cuticle. Only in one case did we observe a sensillum whose dendritic tips (OS) terminated between the epithelium and the cuticle. However, since this was also the only arista that contained 4 sensilla instead of the usual 3, this was probably an exception. It would be interesting to see how the connection between sensilla and epithelium – which must be present early in the ontogeny of the arista – is lost during later developmental stages. From light microscopical observations (anti-HRP-staining) of aristae in the early pupal stage (24 h), we can state that the bipolar neurons are already detached from the epithelium; however, the precise localization of the OS could not be resolved in these preparations (M. Lienhard, pers. comm.). Somewhat similar "internal receptors", which have lost their connection to outer cuticular structures have been described in the antenna of a mosquito (McIver and Siemicki 1976) and of Collembola (Altner and Thies 1973, 1984).

In general, insect sensilla have 3 sheath cells: (1) a thecogen cell, which secretes the dendritic sheath, (2) a trichogen cell, which forms the hair shaft, and (3) a tormogen cell, which builds the hair base. Since the aristal sense organ is highly derived (no hair formation, no attachment to cuticle), it is difficult to correlate its 3 (or 4) sheath cells with the three enveloping cells named above. Only sheath cell 2 can be identified as the thecogen cell, because of the dendritic sheath material that adheres to its membrane. Sheath cell 1 could be the equivalent of the trichogen or tormogen cell, but a clear correlation is hypothetical since there exists neither a hair shaft nor a hair base. Sheath cell 3, which mainly envelops the somata of all 6 neurons, probably corresponds to the neuroglial cell in other insect sensilla (Keil and Steinbrecht 1984; Kuhbandner 1984).

The occurrence of membrane contacts between neuronal and sheath cells and their possible functional significance in thermo/hygroreceptive sensilla has been discussed recently (Steinbrecht 1989). Similar membrane contacts have been observed in the aristal sense organ, but we shall refrain here from any functional speculations, since physiological data on the arista are so far lacking. Future experiments shall first establish a physiological role of the arista in thermo-(hygro?) reception by testing the discrimination behavior of *Drosophila* with and without aristae in a temperature (humidity) gradient. Thereafter electrophysiological recordings will be necessary.

Note added to proof

Preliminary electrophysiological recordings from aristae of the blowfly *Protophormia* demonstrated the presence of a cold receptor (J. Gödde, pers. comm.)

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