Transducing element of crustacean mechano-sensory hairs[1]

N. Kouyama, T. Shimozawa and M. Hisada

Zoological Instiute, Faculty of Science, Hokkaido University, Sapporo 060 (Japan), 31 July 1980

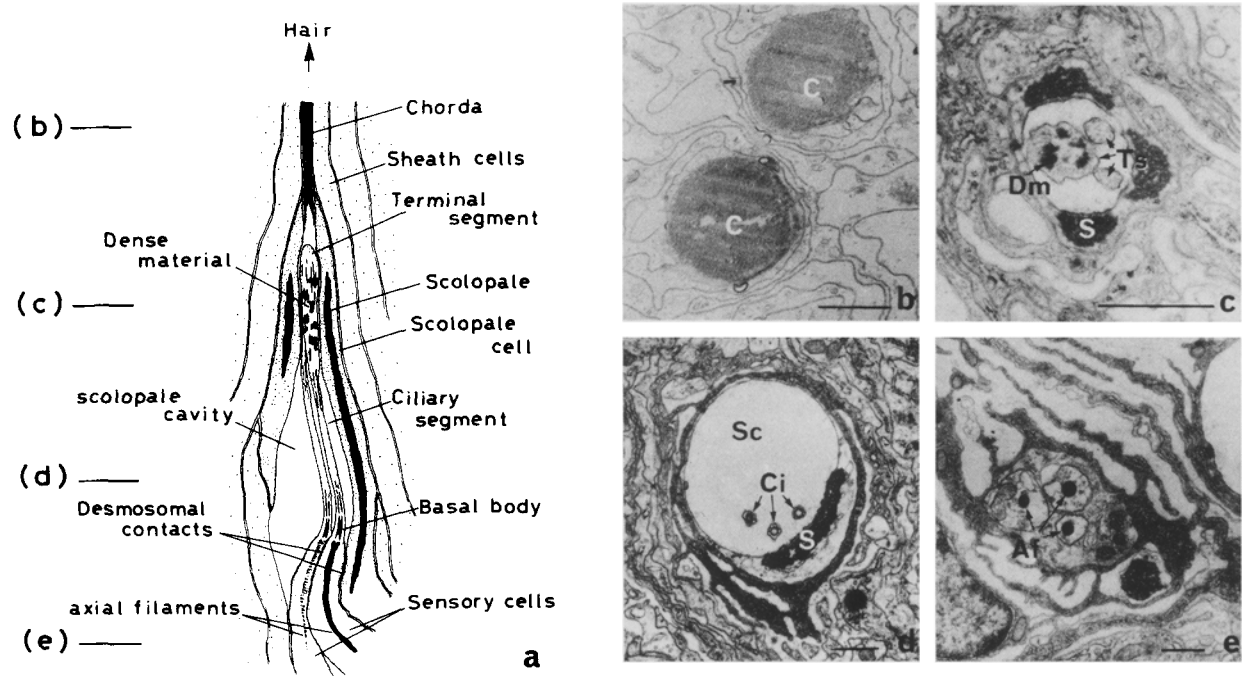
Summary. Scolopidia have previously been reported in the subcuticular chordotonal organs of arthropods, but not in their cuticular sensilla. For the first time, a type of scolopidium was observed in the mechanoreceptive hairs of decapod crustacea.

Crustacea and insects are the principal arthropods whose external surface is protected by sclerotized chitinous cuticle. Such animals therefore possess a cuticular structure as a sensory interface with the environment[2-5]. For instance, the antennular basal segment of the crayfish carries several groups of mechano-sensory hairs. A group of hairs at the dorso-anterior edge of the segment has been identified as the particular input of the semi-giant interneuron C4 in the ventral cord[6,7]. We studied the receptive structure of the hair group with both light microscopy (LM) and electron microscopy (EM) and found a scolopidial transducing element (scolopophore) in the receptor region of the hair. Previously, this structure has been reported only in subcuticular chordotonal organs, such as proprioceptive stretch receptors in insects and crustacea, or in the subgenial, tympanal and Johnston's organ of insects[8,9]. In this report the fine structure of this receptive element is described, and the comparative aspects are discussed.

Antennular basal segments of the crayfish *Procambarus clarkii* were fixed with a gentle injection into the mesenchyme 1.25% glutaraldehyde in 0.1 N cacodylate buffer with 2.5% sucrose added for osmolarity adjustment. The dissected preparation was postfixed in 1% osmic acid, dehydrated and embedded in epon. The sensory structure was reconstructed from the alternate sections for LM and EM. Ultra-thin sections were made at every 5–10 µm. In addition, the external hair structure and the mechanical linkage to the sensory sites were investigated with scanning EM[7].

Electronmicrographs and the schematic reconstruction of the receptive site are shown in the figure. The hair is mechanically linked to the sensory cells by an extracellular 'chorda'. This subcuticular chorda is an amorphous yet solid substance, probably chitinous, and is surrounded by several layers of sheath cells (figure, b). The chorda attaches to the anterior inner wall of the hollow hair shaft. As a result the external force deflecting the hair is transferred to the sensory site via the chorda in the form of tension change. Each chorda is generally 1–2 µm in diameter with a maximal length of up to 5 mm. The transducing element is a spindle shaped scolopidium of 40 µm in length and 4 µm in diameter. This scolopidium consists of 3 sensory cells, 1 scolopale cell and several sheath cells (figure, a). The scolopale cell contains electron dense fibrous material, i.e. scolopale in the cytoplasm[8] (figure, c and d). The cell forms a spindle shaped 'cavity' which contains sensory processes of the 3 sensory cells and the scolopale seems to provide the framework of this cavity (figure, a and d). 3 sensory cells are held tightly together by desmosomal contact at the proximal part of the processes. The scolopale cell also has desmosomal contact with the processes at the same level (figure, a). This seems to be an attachment anchoring the ciliary base to the framwork. An axial filament runs through the proximal process from the base of cilium to the pericaryon (figure, a and e).

Each sensory neuron is a bipolar cell whose soma is 10 µm in diameter and 15 µm in length. It has 1 sensory process protruding into the scolopale cavity. The middle part of the



a Schematic reconstruction of the scolopidial transducing element. The chorda extends upward to the hairs. *b–e* Cross-sectional electron micrographs of the scolopidium at the levels indicated in (a) with the same letter. Scale bars in photographs are 1 µm. C, chorda; Ts, terminal segment; S, scolopale; Dm, dense material; Sc, scolopale cavity; Ci, ciliary segment; Af, axial filament.

sensory process is a ciliary segment with a 9+0 type centriole organization. The terminal segment of the process is slightly dilated and rich in microtubules with scattered dense material and the distal end of the scolopale constricts a set of three dilatations of the terminal segment (figure, c). 3 sensory processes run along the scolopale side of the inner wall of the scolopale cavity so that the ciliary process bends along the curvature of the scolopale which itself warps to provide the cavity (figure, a and d). This ciliary bending seems to be characteristic of scolopidial transduction[8-12]. Tension applied to the scolopidium either straightens the bent cilium or activates the bending of the motile cilium[13]. Neural excitation follows in either case. When hair deflection produces tension in the chorda, the sensory afferent increases its firing frequency[7]. We also observed similar scolopidia in 2 other functionally identified mechanoreceptors: the telson hair[14] and the statocyst hair[15].

EM studies on these hairs have been reported elsewhere[16,17], but no reconstruction of the transducing element has been performed. The sensory structure described here closely resembles the sensory element of a proprioceptive chordotonal organ recorded in a shore crab limb[8]. Similar multi-sensory scolopidia have been also reported in the stretch receptors and in Johnston's organ of insects[9]. On the one hand, these scolopidial sense organs have so far been regarded as a chordotonal organ in which the scolopophore connects to the exoskeleton surface with a ligament under tension and there is no exoskeletal structure. On the other hand, the cuticular mechanoreceptors in insects, e.g. trichoid and campaniform sensilla, have no scolopidia as the sensory elements. Schmidt[11] however proposed that the sensory structures of these sensilla are homologous to those of the insect chordotonal organ.

Based upon the present discovery that the scolopidium in crustacea acts as the sensory element of a hair sensillum, it is likely that the sensory structures in both cuticular and subcuticular mechanoreceptors in arthropods have a homologous origin.

1 Supported by Grant-in Aid for Special Project Research on Mechanisms of Animal Behavior from Ministry of Education, Science and Culture. The authors thank Drs H.S. Hori, S. Nagata, and Mr K. Tanahashi for their help in EM.

2 M.S. Laverack, Comp. Biochem. Physiol. *5*, 319 (1962).
3 R.G.J. Shelton and M.S. Laverack, Comp. Biochem. Physiol. *25*, 1049 (1968).
4 K. Schmidt, Z. Zellfosch. *96*, 478 (1969).
5 W. Gnatzy and K. Schmidt, Z. Zellforsch. *122*, 190 (1971).
6 C.A.G. Wiersma, J. comp. Neurol. *110*, 421 (1958).
7 N. Kouyama and T. Shimozawa, in preparation.
8 M. Whitear, Phil. Trans. R. Soc. Lond. B *245*, 291 (1962).
9 D. Young, Phil. Trans. R. Soc. Lond. B *256*, 401 (1970).
10 K. Schmidt, Z. Zellforsch. *99*, 357 (1969).
11 K. Schmidt, Verh. Dtsch. Zool. Ges. *66*, 15 (1973).
12 K. Michel, Z. Morph. Tiere *77*, 285 (1974).
13 D.T. Moran, F.J. Varela and J.C. Rowley III, Proc. natl Acad. Sci. USA *74*, 793 (1977).
14 D. Kennedy, Physiologist *14*, 5 (1971).
15 M. Takahata and M. Hisada, J. comp. Physiol. *130*, 201 (1979).
16 K. Wiese, J. Neurophysiol. *39*, 816 (1976).
17 H. Schöne and R.A. Steinbrecht, Nature *220*, 184 (1968).

Circadian rhythm of plasma corticosterone in vagotomized rats

S. Itoh, G. Katsuura, R. Hirota and Y. Botan

Shionogi Research Laboratories, Fukushima-ku, Osaka 553 (Japan), 3 April 1980

Summary. In vagotomized rats, 2 weeks after surgery, the amplitude of the circadian rhythm of plasma corticosterone was extremely low, indicating that gastrointestinal activity may be in part involved in the hypothalamo-hypophyseal circadian rhythmicity.

Previous observations indicated that the circadian rhythm of plasma corticosterone is not only entrained by the light-dark cycle but also influenced by eating and drinking[1-5]. Following a 14-day restricted feeding schedule the peak of plasma corticosterone was observed just prior to feeding time and the elevated level declined promptly after food presentation[6]. Such alteration of the rhythm was assumed to be produced by a central mechanism. However, there is a possibility that gastrointestinal activity might be involved in the change of rhythm pattern of the plasma corticosterone level. If so, visceral stimulation may be in part transported to the hypothalamo-hypophyseal system via the vagus. The present investigation was therefore carried out to investigate participation of the vagus in the circadian rhythm of plasma corticosterone using vagotomized rats.

Methods. Female Wistar rats, 8 weeks of age, were used. They were housed at a constant temperature of 25±2 °C with a regulated photoperiod of 12 h, lights on at 07.00 h, and rat biscuit (Oriental Yeast Co.) was given with water ad libitum.

Subdiaphragmatic vagotomy was done under nembutal anesthesia. Through an upper abdominal midline incision the esophagus was isolated from its surrounding tissue near the esophago-gastric junction and the vagal nerve trunks were removed. In sham-operated rats the vagal nerves were kept intact after the isolation. The operated rats were used for experiments 14 days after surgery. The animals were sacrificed by decapitation and trunk blood was collected, 7 times during the day, from 06.00 h to 24.00 h, as seen in the figure. Plasma corticosterone was determined by the method of Zenker and Bernstein[7] with minor modifications. The stomachs of the vagotomized rats showed atonic distension, and vagotomy was verified anatomically using a dissecting microscope.

For data analysis Student's t-test was used. Moreover, a 24-h cosine curve was fitted by the method of least squares to obtain the mesor, amplitude and acrophase, employing an electronic computer[8].

Results. In vagotomized rats the peak value of plasma corticosterone was 45.6±4.74 (SEM) μg/dl at 19.00 h and the trough 20.3±4.06 μg/dl at 12.00 h, while in sham-

Circadian rhythm of plasma corticosterone in vagotomized and sham-operated rats

	Sham-operated	Vagotomized
Mesor (μg/dl)	38.3	33.5
Amplitude (μg/dl)	28.4	9.5
Acrophase (h)	21.28	20.30