

PHAGOCYTOSIS BY OUTER ROOT SHEATH CELLS OF THE MOUSE VIBRISSAE (SINUS HAIRS)*

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ABSTRACT

Phagocytic cells are present at all levels of the outer root sheath of mouse vibrissa (sinus hair) follicles. Occasionally these cells are seen in the process of engulfing other cells; more often phagocytic cells are seen which contain one or more vacuoles whose contents are in various stages of digestion. Most of the phagocytosed cells contain many tonofilaments and a reduced amount of cytoplasm. A few of the vacuoles contain no filaments. As one vibrissa after another grows from the follicle without an intervening rest period, the outer root sheath changes in size and shape to accommodate both the club hair and actively growing vibrissa. Phagocytosis appears to play a role in the continuous reshaping of the outer root sheath.

Epithelial cells have been shown to be capable of phagocytosis in response to injury or trauma. While some of the injured cells die, other cells in the vicinity have been shown to ingest fibrin [1, 2], serous exudate [1], erythrocytes [3, 4], portions of other cells [5], and injected inert particles [3, 4]. In cortical cells of the hair matrix, phagocytosis occurs as a normal process as the cells ingest dendritic processes of melanocytes [6]. We wish to report in this study that phagocytosis also occurs as a normal process in the outer root sheath of mouse vibrissa follicles.

MATERIALS AND METHODS

Vibrissa follicles were excised from the upper lip of male and female young adult mice. Some of the animals were albino and others were black and white mice of unknown strain.

For light microscopy, tissue was fixed in 10% formalin, washed in tap water, dehydrated in ethanol, embedded in paraffin, and sectioned at 8 μ . The sections were stained with Holmes silver stain and luxol fast blue counterstain [7].

For electron microscopy, the tissue was fixed in 4% glutaraldehyde in sodium cacodylate buffer, postfixed in 2% osmium tetroxide, dehydrated in methanol, and embedded in Epon. Thick sections ($1\frac{1}{2}$ μ) were stained with toluidine blue and studied with the light microscope. Thin sections (about 500 Å) were stained with uranyl acetate and lead citrate and studied with an RCA EMU3F electron microscope at 50 KV.

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OBSERVATIONS

Light Microscopy

The structure of the vibrissa (sinus hair) follicle in mice, in general, corresponds to the description given by Melaragno and Montagna [8]. Each follicle is surrounded by a blood sinus which, in turn, is surrounded by a connective tissue capsule. The lower part of the sinus (the cavernous sinus) is traversed by delicate connective tissue trabeculae; the upper part (the ring sinus) contains no trabeculae. A collar-like structure (the ringwulst) extends from the follicle into the ring sinus.

In contrast to the regular hair follicles of the coat of the mouse, the sinus hair follicles appear to remain in an actively growing state without going through a periodic resting state (telogen). Soon after a vibrissa stops growing and becomes a club hair, a new hair begins to grow from the bulb of the follicle. As the club hair moves upward in the follicle, it is displaced to one side, causing an enlargement in that part of the outer root sheath. While mitotic figures of the hair and its inner root sheath are confined to the bulb of the hair, mitotic figures are seen at all levels in the outermost layer of cells in the outer root sheath.

Phagocytic vacuoles are often present in the outer root sheath (Fig. 1), but are easily overlooked because the contents of the vacuoles usually do not stain. The vacuoles were not noted in Melaragno and Montagna's work.

Electron Microscopy

Phagocytic cells may be present in all levels of the mouse vibrissa outer root sheath, from the region just above the bulb to the region just below the epidermis. These cells were found in each animal studied. Dehydrated-looking cells are often seen in the outer root sheath. Occasionally a phagocytic cell is seen which was fixed while in the process of engulfing one or more of these cells. In Figure 2, two cells are being engulfed by a phagocytic cell. The desmosome within the forming

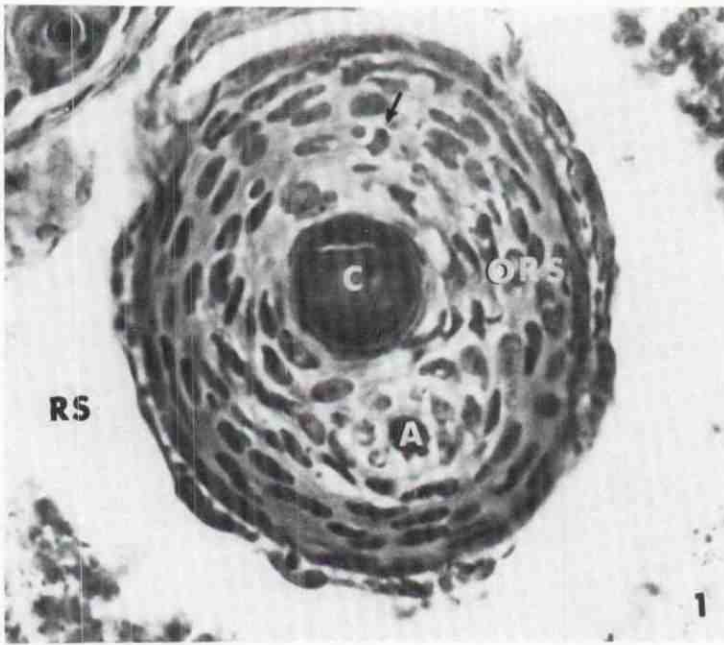


FIG. 1: Cross section of a vibrissa follicle with surrounding ring sinus (RS). Follicle contains a club hair (C) and the tip of an actively growing hair (A). Outer root sheath (ORS); phagocytic cell (arrow). Holmes silver stain with luxol fast blue counterstain. ($\times 57$)

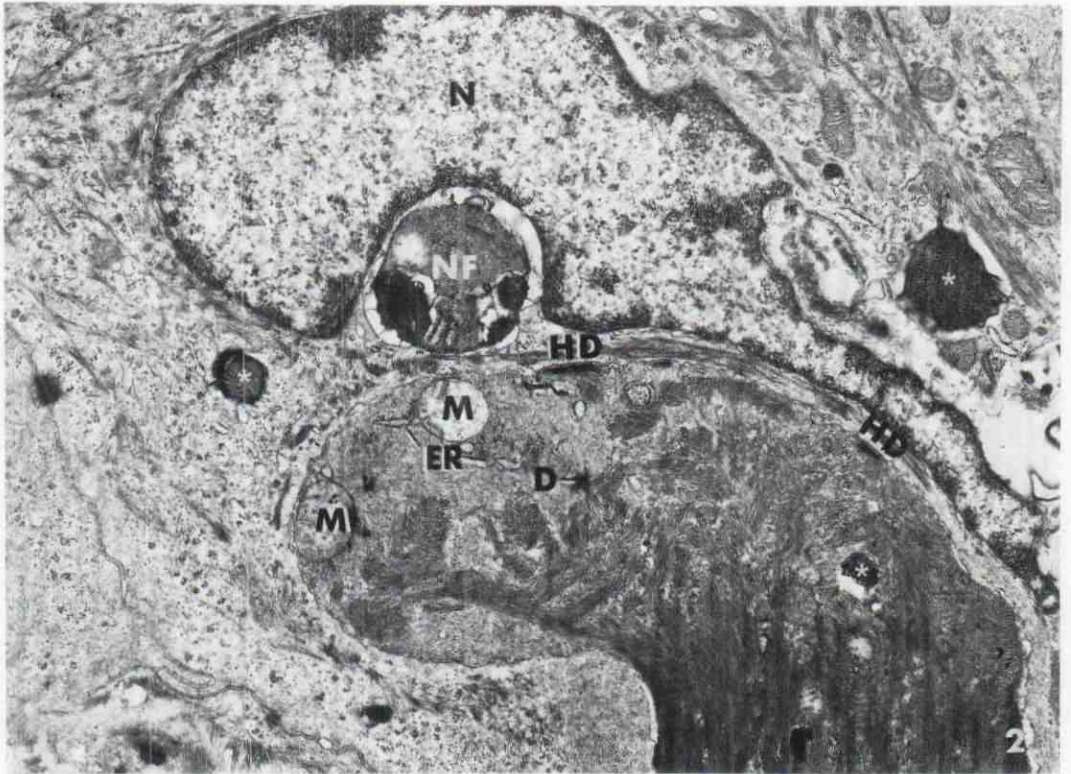


FIG. 2: Phagocytic cell fixed during engulfment of two dehydrated-looking cells which contain many filaments. Some remnants of desmosomes are seen on the surface of the vacuole (HD) and within the cell mass (D). Also seen are mitochondria (M) and granular endoplasmic reticulum (ER). A smaller vacuole of the nonfibrillar type (NF) deeply indents the nucleus. Smaller dense bodies (*) may be remnants of phagocytic vacuole contents. Nucleus of phagocytic cell (N). ($\times 12,000$)

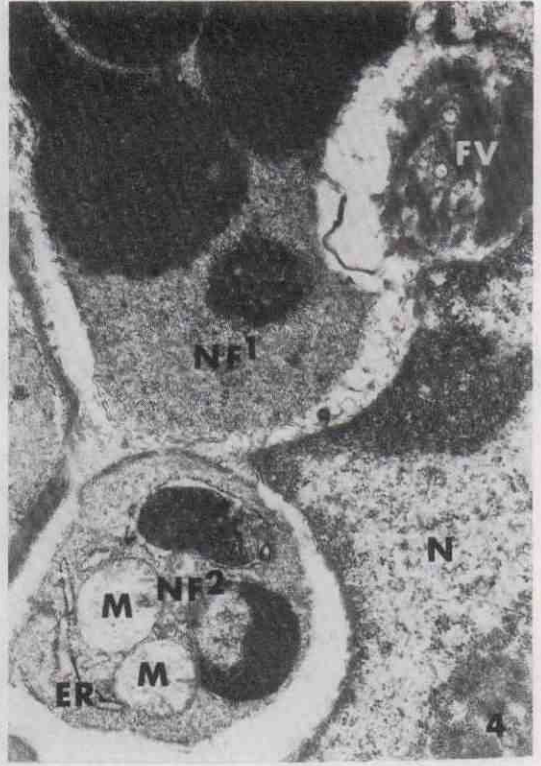
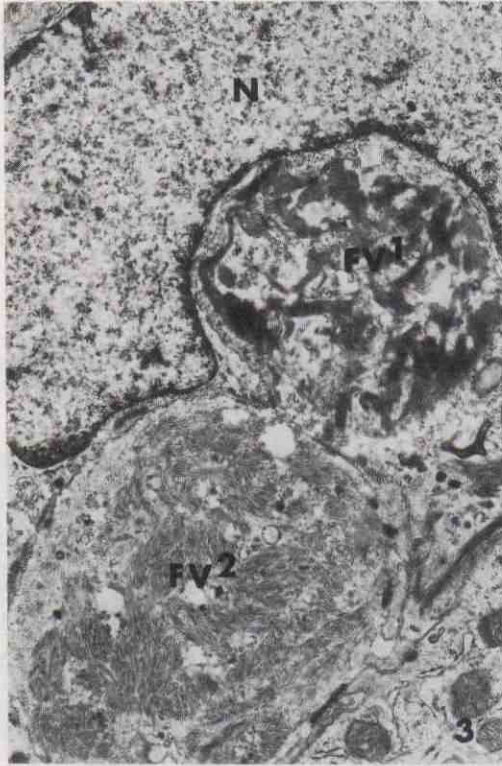


FIG. 3: Portion of a phagocytic cell containing two fibrillar-type vacuoles (FV¹ and FV²). Upper vacuole appears to be in a more advanced stage of digestion. Nucleus of phagocytic cell (N). ($\times 10,000$)

FIG. 4: Portion of a phagocytic cell containing one fibrillar vacuole (FV) and two nonfibrillar vacuoles (NF¹ and NF²). Note mitochondria (M), granular endoplasmic reticulum (ER), and darkly staining nuclear remnants. Nucleus of phagocytic cell (N). ($\times 15,000$).

vacuole shows that the cell mass being engulfed consists of two cells. Half-desmosomes at the surface of the cell mass denote the remains of their former attachment to adjacent cells. More commonly seen are phagocytic cells containing vacuoles with their contents in various stages of digestion (Figs. 2-4). Small electron-dense clumps of material in the cytoplasm (Fig. 2) may represent the final stages in the digestion of vacuoles.

Phagocytic vacuoles containing recognizable cell parts are of two types. The most common type contains many tonofilaments and a reduced amount of cytoplasm (Figs. 2-4). A few mitochondria and some rough-surfaced endoplasmic reticulum may be seen (Fig. 2). Vacuoles in Figures 3 and 4 show subsequent stages in the digestion of the contents of this type of vacuole.

The second type of vacuole contains few if any filaments (Figs. 2, 4). Some nuclear remnants, along with some mitochondria and rough endoplasmic reticulum, are seen. A relatively small number of Langerhans cells and Merkle cells, neither of which have filaments, are present in the outer root sheath. It may be that remnants of these cells are present in the second type of vacuole.

DISCUSSION

While several investigators have shown that epithelial cells can become phagocytic in response to injury or trauma, the present study shows that phagocytic cells are normally present in the outer root sheath cells of mouse vibrissa follicles. The

early cortical cells of hair which ingest melanosome-containing processes of melanocytes have been described earlier as being phagocytic [6].

Montagna noted the presence of necrotic cells and mitotic figures in the outer root sheath of human hair follicles and stated that cell death and cell division have a direct relation to each other [9]. He did not note the fate of the necrotic cells, although he mentioned that some of the outer root sheath cells were shed into the pilary canal after keratinization. The necrotic cells he described may correspond to the dehydrated-looking cells in the outer root sheath of mouse vibrissa follicles.

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