Acta Dermatovener (Stockholm) 53: 22–30, 1973

HIDRADENOMA PAPILLIFERUM

An Electron Microscopic Study

Ken Hashimoto

From the Department of Medicine, Division of Dermatology, and University of Tennessee, College of Medicine, The Veterans Administration Hospital, Memphis, Tennessee, USA

Abstract. An electron microscopic examination of a typical specimen of hidradenoma papilliferum was performed. Glandular epithelium lining the lumina was composed of basally located myoepithelial cells and luminally located columnar secretory cells. The latter showed a striking similarity to the apocrine secretory cells in containing dense, multi-component secretory granules of enormous size, well developed Golgi apparatus, and swollen apical cytoplasm. The apical portion was segregated into the lumen by decapitation. A rosette form of glycogen particles and peculiar laminated fibrous structures were found.

Hidradenoma papilliferum is a rare tumor occurring in the female genitalia. A survey of a large series (65 specimens) of this tumor was made by Meeker et al. (8). They summarized their own findings as well as those of 21 others. This tumor is so rare that it is seldom brought to the attention of electron microscopists, also when the tumor is diagnosed histologically by the routine method, no more lesion is available for further study. I have obtained a clinically and histologically typical tumor fixed in 10% formalin buffered to pH 7.0 with phosphate buffer. In the following, the results of the first electron microscopic analysis of this tumor will be reported.

MATERIALS AND METHODS

A subcutaneous nodule was removed from a 48-year-old white female. The lesion was present asymptomatically for 6 months in the mid-portion of the left labium majus. A surgeon excised the lesion at the time of operation for patient's hemorrhoid. The excised specimen was first fixed for a short period in 10% neutral formalin adjusted with 1/10 M phosphate buffer to pH 7.0. The specimen was rinsed overnight in the same buffer, cut in 1mm³ pieces and fixed again in 5% glutaraldehyde in 1/10 M cacodylate buffer, pH 7.2 for 3 hours. The tissue blocks were rinsed in the same buffer overnight, post-fixed with 1% osmic acid in the same buffer and dehydrated with 50% through absolute alcohol and propylene oxide. During dehydration between 50% and 60% alcohol, specimens were stained with 1% uranyl acetate in 50% ethanol for 30 minutes. All tissue blocks were embedded in Araldite. Thin sections, 400–600 Å, were cut in a Porter-Blum MT-2 ultramicrotome, stained with 15% uranyl acetate in 50% methanol and Reynold's lead citrate (13), and observed in a Hitachi HU-11C and HU-12 electron microscopes at 100 kV and 125 kV.

For a comparative study the normal axillary apocrine gland of a 15-year-old Negro female was obtained by biopsy and similarly processed.

A part of the specimen was processed routinely and stained with hematoxylin and eosin.

RESULTS

Light microscopy. Papillary growth was encapsulated within an epithelial wall (Fig. 1 A). The wall cells were either keratinized through the formation of keratohyaline granules, or non-keratinized (Figs. 1 A, 1 B). Non-keratinized epithelium resembled the wall cells of steatocystoma multiplex (2, 3) and those of trichilemmal cyst (11). Papillary growth was located in the center of the wall and serial section failed to establish a continuity with the wall. It was assumed that the major portion of the growth was connected to the wall by a thin stalk which had been cut off above the level of the first section and therefore excluded from the present observation. At a high magnification, the epithelium lining most of the glandular lumina, and in many cases also that covering papillary projections, were resolved into two layers of dissimilar cells; the cuboidal basal

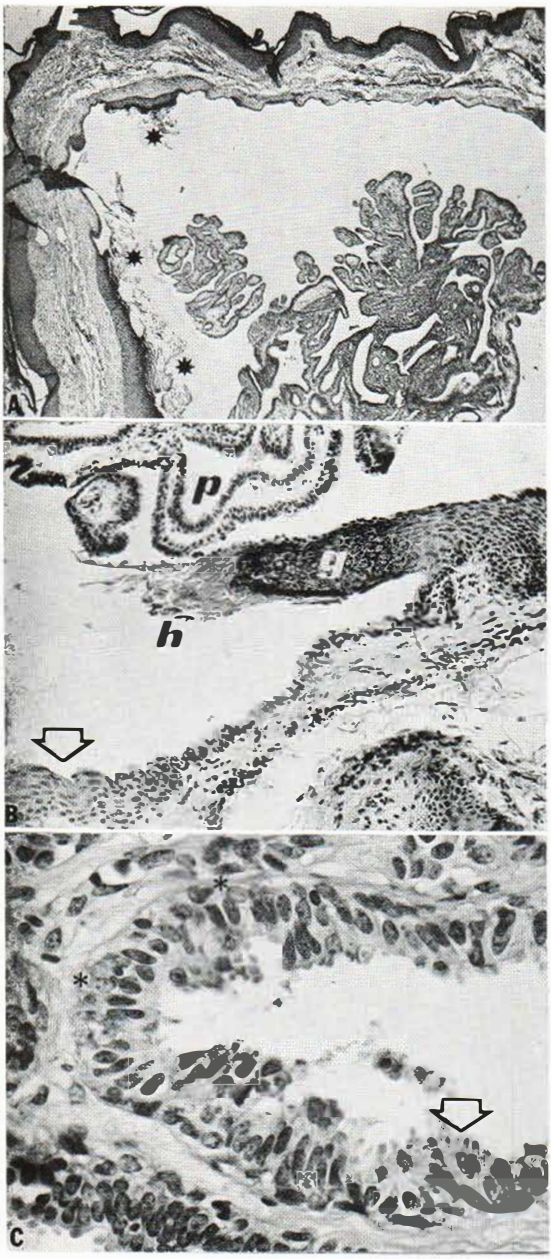

Fig. 1 (A) Papillary growth is encapsulated with an epithelial wall which shows several keratinized areas (*). *E*, epidermis. × 57.

(B) Keratinized wall projects into the lumen and touches papillary growth (*p*) of the tumor. In this projection, a thick granular layer (*g*) is underlying the horny layer (*h*). In the non-keratinizing part, scaly squamous cells cover the wall (*arrow*). × 93.

(C) Glandular epithelium is composed of basal cuboid cells (*) and luminal columnar cells. The apical portion of columnar cells often bulges (*arrow*). Secretion products are seen in the lumen. × 370.

cells and tall columnar luminal cells with rather clear apical swelling (Fig. 1 C). Apical cytoplasm was often "decapitated" and found in the lumen as fragments (Fig. 1 C). Some luminal cells appeared short and collapsed. Occasionally very cellular areas without lumina were found.

Electron microscopy. The cuboidal basal cells, when observed with the light microscope, were revealed to be myoepithelial cells with typical myofilaments showing dense areas (Figs. 2, 3). In favorably cut sections both columnar (Fig. 2) and short (Fig. 3) luminal cells were seen connected basally with these myoepithelial cells.

A typical columnar luminal cell showed a basally located oval nucleus with one or two nucleoli. Dense granules of various sizes (Figs. 2, 4) and numerous vesicles (Fig. 5) were found within an abundant apical cytoplasm. Sparsely distributed long villi grew from the luminal border. A large number of mitochondria, dilated cisternae of rough and smooth endoplasmic reticulum and well developed Golgi apparatus were observed (Figs. 4, 5). Narrowing of the neck portion of the swollen apical cytoplasm (Figs. 2, 4, 5) and disintegration or segregation of various amounts of apical cytoplasm (Fig. 2) were frequently seen. Dense granules were present in most of the columnar luminal cells (Figs. 2, 4, 5). They seemed to begin as small vesicles, gradually became denser as they enlarged and eventually coalesced to form very large compound granules which were delimited with their own membranes (Fig. 6 A). One component of these large granules was a round opaque substance compatible with lipids (Fig. 6 A). When compared with apocrine secretory granules (Fig. 6 B), a striking similarity was recognized. Aggregations of lamellar structures were found in many columnar luminal cells (Fig. 7). The lateral profile of individual lamella showed a unit membrane structure about 70 Å thick in outer diameter. *En face* profile was seen as a sheet of various shapes, sizes, and densities (Fig. 7). The whole aggregation was often surrounded with a membrane (Fig. 7). When compared with similar structures of the normal apocrine secretory cells (Fig. 8) the resemblance between them was again very striking. Glycogen particles in rosette forms, i.e. as α-particles (Fig. 5), were present in many luminal cells. In contrast to the columnar luminal cells, short luminal cells did not have large granules (Fig. 3). Other organelles present

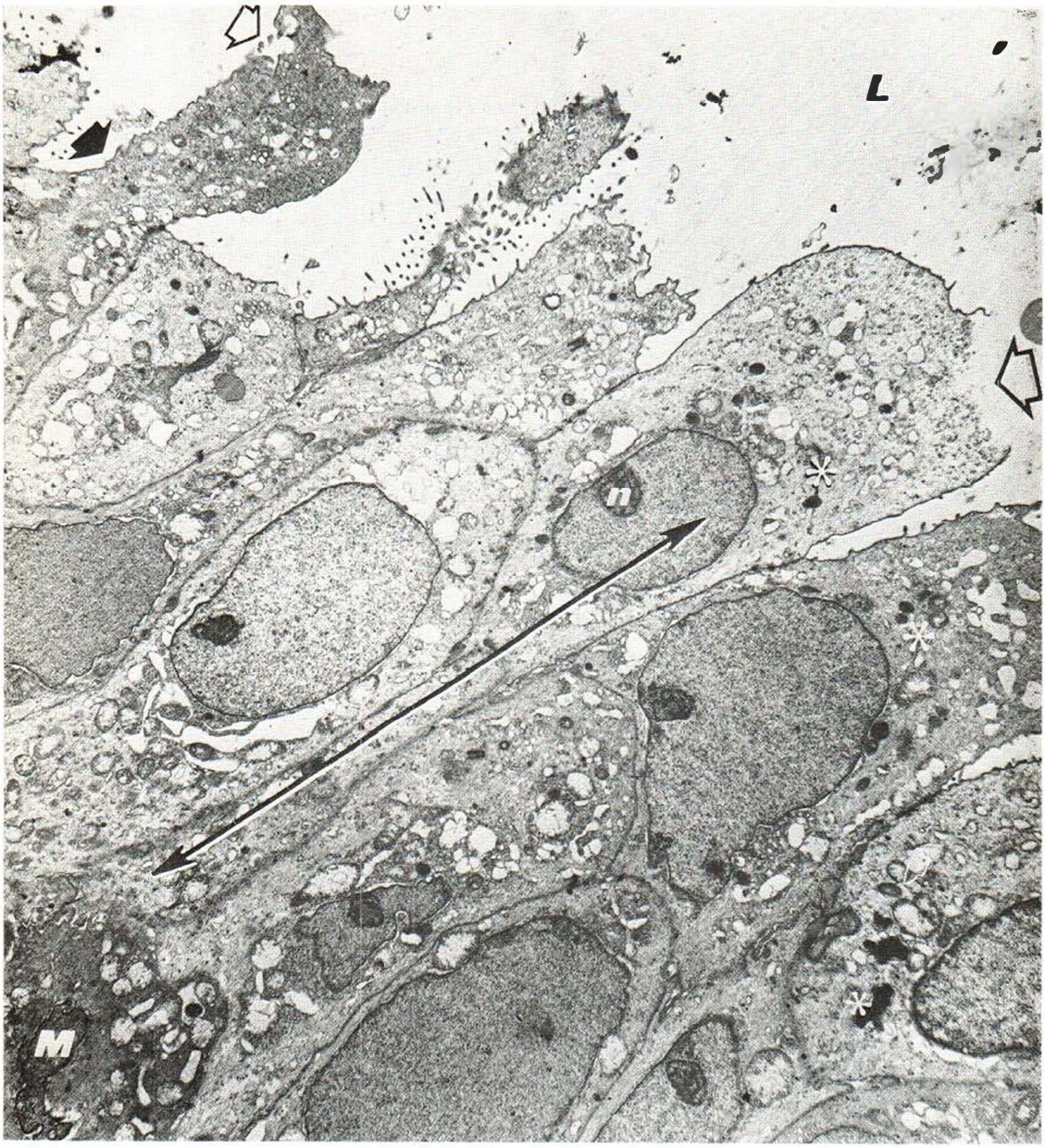

Fig. 2. Tall columnar cells extend into the lumen (*L*). A favorably sectioned cell (*long arrows*) shows attachment to a basally situated myoepithelial cell (*M*). Most of the columnar luminal cells contain dense secretory granules (*). Solid short arrow: narrowed neck of the apical cytoplasm. Hollow arrow: breakage of luminal plasma membrane and release of the contents. *n*, nucleolus. × 1 830.

in columnar cells were either absent or poorly developed. Since these short cells were also situated upon the myoepithelial cells and since they did not show the development of a periluminal meshwork of tonofilaments as apocrine (4) and eccrine (5) ductal epithelial cells always do, they were thought to represent the resting luminal cells or the columnar cells which had collapsed after discharging apically located secretory granules. Occasionally, myoepithelial cells were found among luminal cells (Fig. 4), apparently detached from their basal location. Typical ductal epithe-

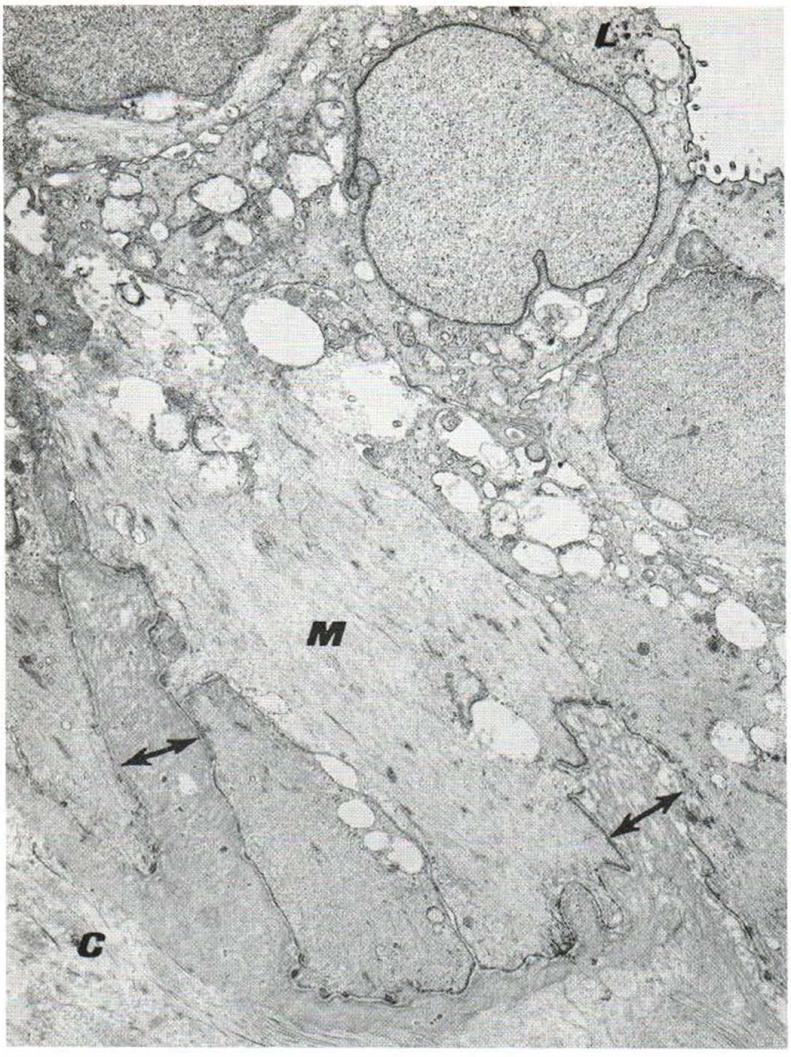

Fig. 3. Short luminal cells (*L*) do not contain secretory granules. Myoepithelial cells (*M*) surround the periphery of these cells. Ramified basal lamina and amorphous substance form a thick band (between arrows). *C*, collagen. × 7 800.

lium was not found in the present study. Had it been present, it would have been noticed because of several characteristics, such as the periluminal meshwork of tonofilaments and short but densely occurring luminal villi (4, 5). Surrounding the periphery of the glandular epithelium, the basal lamina often showed multiplication (Fig. 3). An admixture of fine filaments, anchoring fibrils, and amorphous substances filled the spaces between the ramified basal laminae. In some areas, the thickness of this admixture reached a few micra (Fig. 3).

DISCUSSION

According to the study of Meeker et al. (8), hidradenoma papilliferum occurs most commonly in the genital region of middle aged (30–49) white females; in this regard, the tumor examined in this study was typical. Histologically, tumors surrounded with an epithelial capsule were found, though not commonly, in their series (8). As demonstrated in the present study, this tumor resembles the secretory epithelium of the apocrine gland. It may be that the epithelial capsule, which undergoes keratinization through the formation of keratohyaline granules, represents an external hair sheath. A less keratinized portion, described above as resembling the wall of steatocystoma multiplex or trichilemmal cyst, may represent trichilemmal portion of the external hair sheath (11).

With the high resolving power of the electron microscope, myofilaments were observed in the

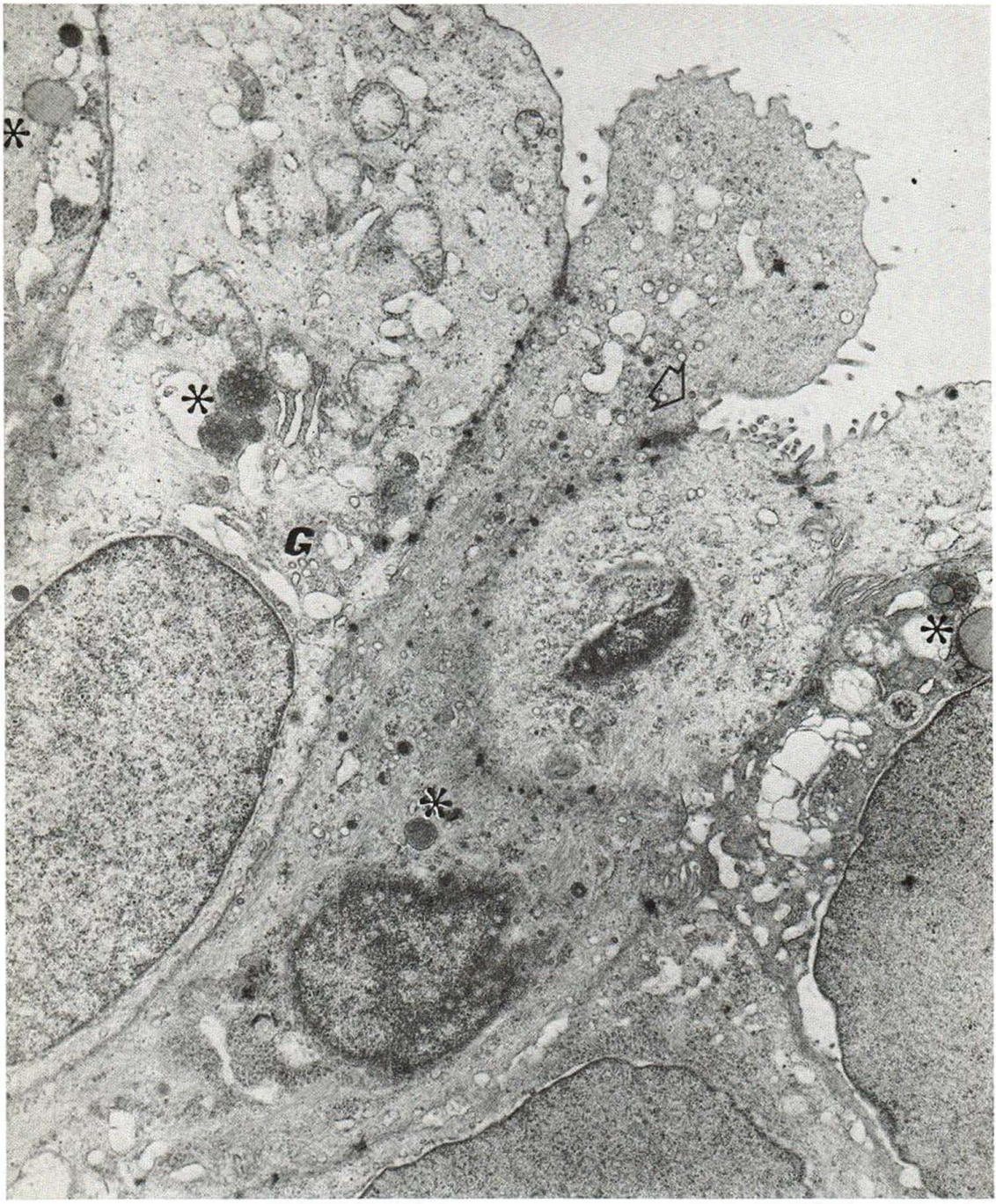

Fig. 4. High magnification of columnar luminal cells shows dense secretory granules in every cell (*). Arrow: narrowed neck of the apical cytoplasm. *G*, Golgi apparatus. × 7 500.

basally located cuboidal cells; this identified these cells as myoepithelial cells. Interestingly, the presence of myoepithelial cells was proposed by some (6, 10, 14) and denied by others (8) in previous light microscopic studies. The presence of myoepithelial cells and secretory granule-containing tall cells and the absence of ductal epithelium made it most likely that the main direction of differentia-

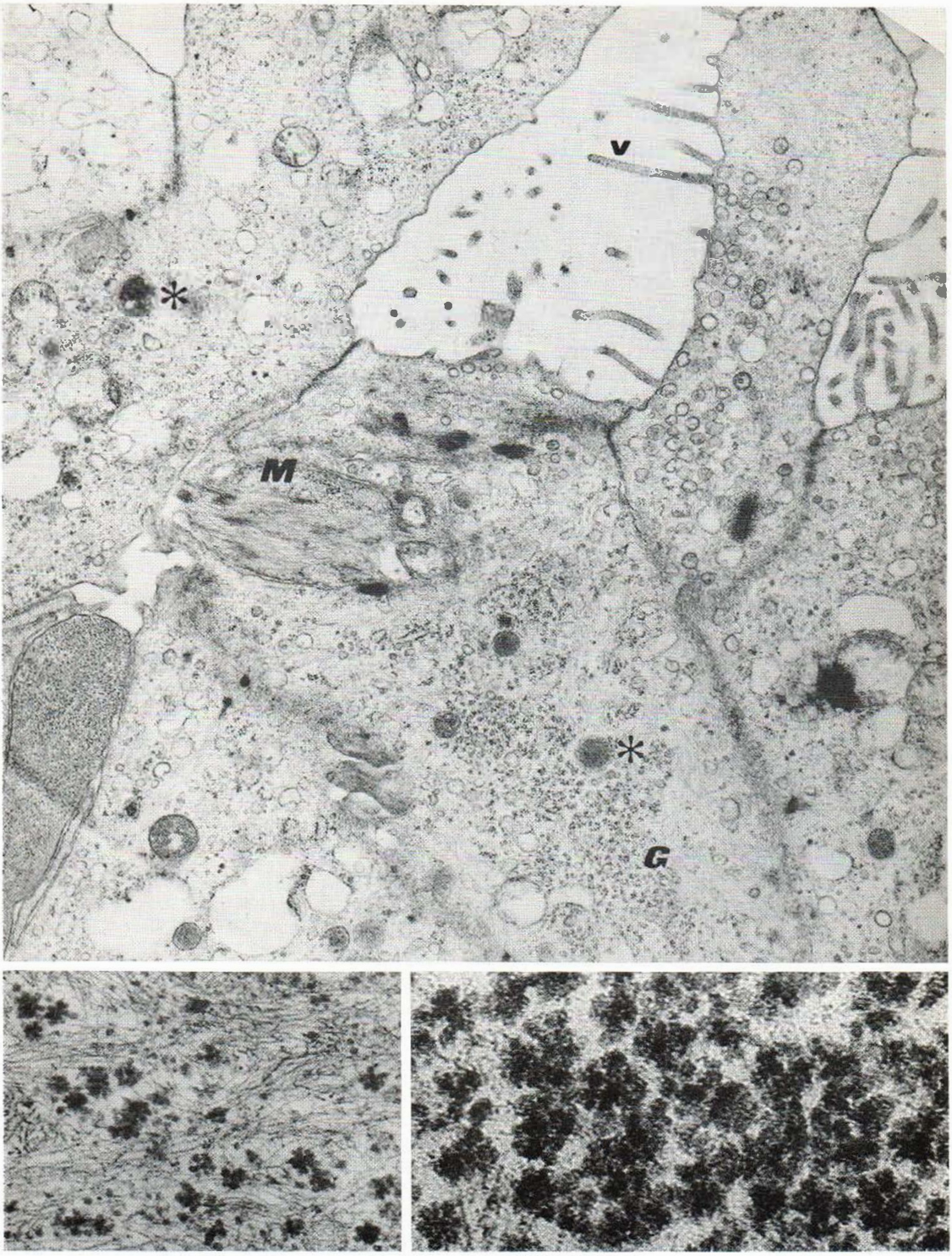

Fig. 5. Numerous vesicles and slender villi (*v*) are seen in the apical portion of columnar cells. A myoepithelial cell (*M*) is admixed with these cells. Rosette form of glycogen (*G*) is seen in one cell and enlarged in the lower pictures. *, secretory granules. Upper: × 13 800. Lower left: × 63 400. Lower right: × 124 000.

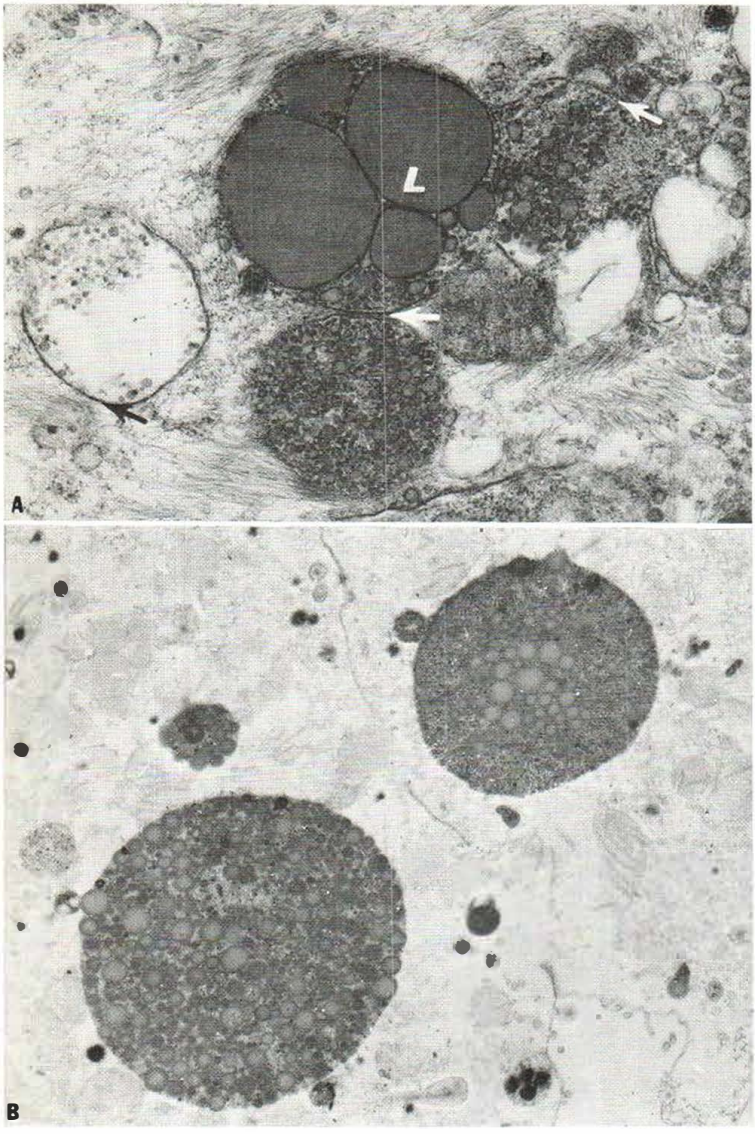

Fig. 6. (A) Coalescence of small and medium-sized granules produce larger granules. Many of the component granules are medium-opaque and lipid-like (*L*). Each granule is delimited with membrane (*arrows*). × 26 000.

(B) Large secretory granules of a normal apocrine gland are also composed of many lipid-like droplets and other multiple components. Although the magnification is different, these granules are very similar to those of the tumor cells as shown in Fig. 6 A. × 13 900.

tion of hidradenoma papilliferum is toward secretory epithelium. The presence of large secretory granules with lipid globules, lamellar structures and decapitation secretion (6) strongly suggest that the type of differentiation exhibited by this tumor is apocrine. This conclusion agrees with that of Meeker et al. (8) and also with the generally held view (7, 12). In the secretory segment of the eccrine gland, mucous cells or so-called dark cells contain dense granules. They are, however, located mainly along the luminal border and do not reach the basally located myoepithelial cells. Also, the dense granules neither coalesce nor attain the sizes of tumor cell granules. Furthermore, the majority of the cells in the eccrine secretory coil are granule-free "clear cells". Therefore, the presence of a large number of large granules in almost every cell, as seen in the tumor cells, is not characteristic of eccrine secretory cells.

Decapitation secretion or, more precisely, disintegration of apical cytoplasm and release of its content into the lumen, is typically apocrine. This mode of secretion, however, was observed in eccrine spiradenoma (3). Lamellar structures which were often surrounded with a delimiting membrane have not been described in the normal

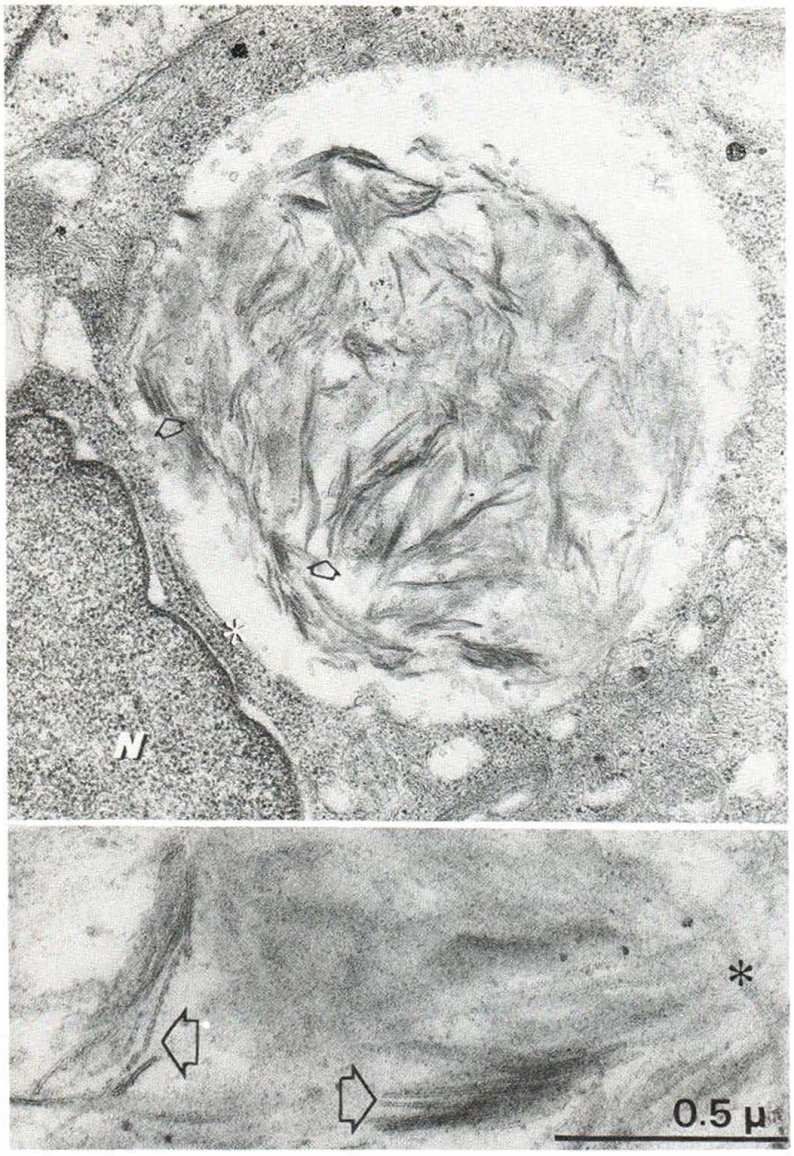

Fig. 7. An aggregation of lamellar structures is located in an tumor cell near the nucleus (*N*). The lateral view shows a unit membrane structure *(arrows)*, whereas *en face* profile reveals ill-defined sheets of various densities. The delimiting membrane is partially visible (*). Upper: × 25 600; lower: 64 000.

apocrine gland. This may be the precursor of the large laminated granules (6). It is different from the annulate lamellae (1) because the annulate lamellae are made of membrane appositions. Ramified basal laminae admixed with fine filaments and amorphous substances have been found to be the main constituents of the hyaline membrane and hyaline droplet of dermal cylindroma (3) and eccrine spiradenoma (3). The rosette form of glycogen, also known as α-particles, is not normally present in the skin and its appendages, although it can be induced by several hexosamines, notably galactosamines, as demonstrated by Nieland et al. (9). Apparently, some metabolic blockage peculiar to this tumor caused the accumulation of α-particles since no other appendage tumors examined by this author contained this type of glycogen.

ACKNOWLEDGEMENTS

The author is an awardee of Medical Investigatorship: The work reported here was supported in part by V.A. Part I, Designated Research Funds.

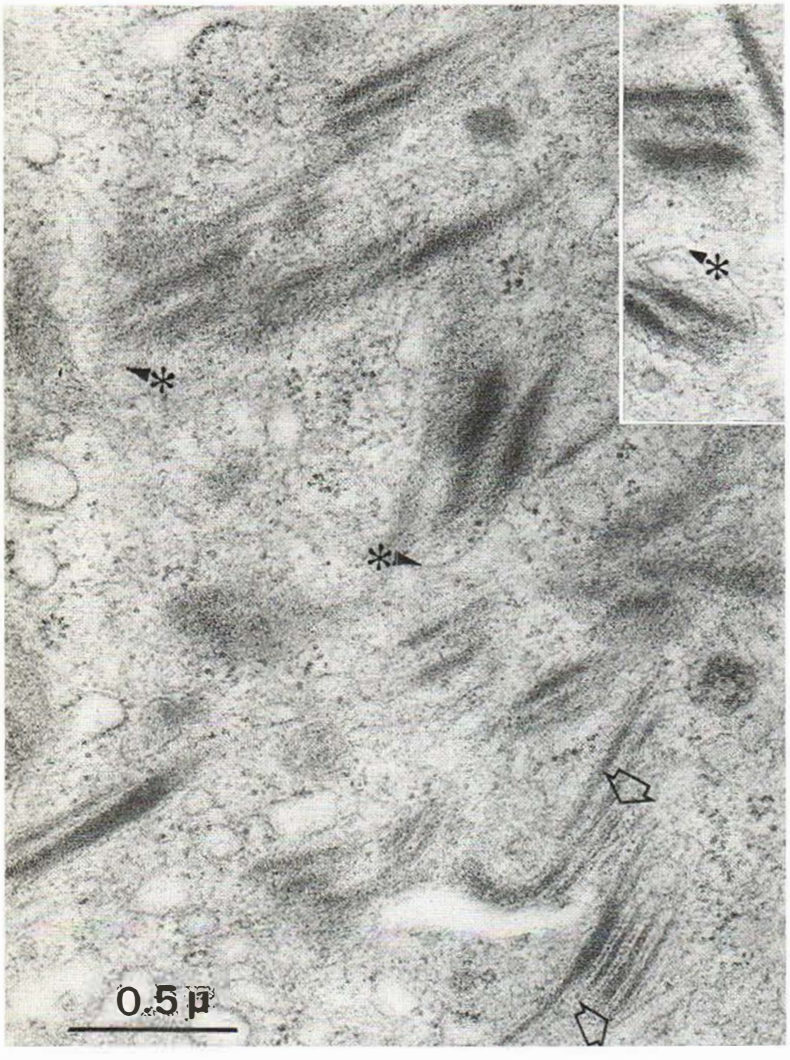

Fig. 8. Lamellar structures from normal apocrine secretory cells also reveal membrane profile in lateral view (*arrows*) and sheet-like dense areas in *en face* view. Some of these are membrane-surrounded (*). × 44 400.

REFERENCES

1. Gross, B. G.: Annulate lamellae in the axillary apocrine glands of adult man. J Ultrastruct Res *14:* 64, 1966.
2. Hashimoto, K., Fisher, B. K. & Lever, W. F.: Steatocystoma multiplex. Kasuistik und elektronenmikroskopische untersuchungen. Hautarzt *15:* 299, 1964.
3. Hashimoto, K. & Lever, W. F.: Appendage Tumors of the Skin. pp. 29–36; 55–83; 97–99, Thomas, Springfield, Ill., 1968.
4. Hashimoto, K., Gross, B. G. & Lever, W. F.: An electron microscopic study of the adult human apocrine duct. J Invest Derm *46:* 6, 1966.
5. — Electron microscopic study of the human adult eccrine gland. I. The duct. J Invest Derm *46:* 172, 1966.
6. — Electron microscopic study of apocrine secretion. J Invest Derm *46:* 378, 1966.
7. Lever, W. F.: Histopathology of the Skin, 4th ed., pp. 549–551. Lippincott, Philadelphia, 1967.
8. Meeker, J. H., Neubecker, R. D. & Helwig, E. B.: Hidradenoma papilliferum. Amer J Clin Path *37:* 182, 1962.
9. Nieland, M. L., Wanger, N. S. & Hambrick, G. W.: Keratinocyte glycogen: An ultrastructural study of the epidermal response to galactosamine and glucose in organ culture. J Invest Derm *56:* 113, 1971.
10. Pick, L.: Über Hidradenoma und Adenoma hidradenoides. Virchows Arch Path Anat *125:* 312, 1904.
11. Pinkus, H.: "Sebaceous Cysts" are trichilemmal cysts. Arch Derm (Chicago) *99:* 544, 1969.
12. Pinkus, H. & Mehregan, A. H.: A Guide to Dermatohistopathology, pp. 436–437. Appleton-Century-Crofts, New York, 1969.
13. Reynolds, E. S.: The use of lead citrate at high pH as an electron opaque stain in electron microscopy. J Cell Biol *17:* 208, 1963.

Received May 2, 1972

Ken Hashimoto, M.D.
Department of Medicine
Division of Dermatology
The Veterans Administration Hospital
Memphis, Tennessee, 38104
USA