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THE NORMAL HUMAN ECCRINE AND APOCRINE GLANDS*

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Preliminary to a study of benign appendageal tumors of the skin, the results of which will be published separately, the following study of normal apocrine and eccrine glands was undertaken. The use of the periodic acid-Schiff (PAS) and Alcian Blue (AB) stains separately on tissue sections and the clarification of the two color reactions of the PAS stain by Alcian Blue (1) has lead to a better histochemical characterization of some of the structures of normal human eccrine and apocrine glands.

MATERIALS AND METHODS

Specimens from four and five millimeter punch excisions of normal skin were fixed in 10% formalin solution for at least 24 hours and mounted in paraffin. Serial sections were stained with hematoxylin and eosin, periodic acid-Schiff, periodic acid-Schiff after diastase digestion, Alcian Blue, and Alcian Blue after diastase digestion.

Standard aqueous Schiff reagent was prepared by mixing: (a) basic fuschin 4.0 Gms., (b) sodium metabisulfite 7.6 Gms., (c) N hydrochloric acid 60 cc., and (d) distilled water 340 ml. The resulting solution was shaken with 2 Gms. of charcoal for two to three hours. If the first filtrate was not colorless more charcoal was added and the solution was again shaken and refiltered until a colorless filtrate was obtained. The solution was stored at 5°C.

Periodate solution was prepared by dissolving 0.69 Gms. of potassium metaperiodate in 100 ml. of 0.3% nitric acid with heat.

The procedure for staining the tissue sections was as follows: (a) paraffin sections were brought to water, (b) 10 minutes in periodate, (c) washed in three baths of water, total of 6 to 9 minutes, (d) 10 minutes in Schiff reagent in the refrigerator at 5°C., (e) two minutes in each of three rinses of sodium metabisulfite (1:20 dilutions of 10% solutions), (f) counterstained with saturated picric acid solution for about 15 seconds, (g) washed in water briefly, and (h) dehydrated and mounted in the standard manner. Diastase digestion was employed between steps (a) and (b) and consisted of incubation of sections in a freshly prepared 0.1% malt diastase solution (which had been filtered minutes before use) for 45 minutes, followed by washing in running tap water for another 45 minutes.

Alcian Blue solution was prepared by dissolving 0.1% Alcian Blue 8GS in 3% acetic acid to which was added a crystal of thymol after filtering. The final pH was 2.7 to 3.0.

The tissue sections were stained as follows: (a) paraffin sections to water, (b) 30 minutes in Alcian Blue solution, (c) washed for 15 minutes in running tap water, (d) counterstained with picric acid for about 15 seconds, and (e) washed in water briefly, and dehydrated and mounted in the standard manner. If diastase was employed it was introduced before step (b).

RESULTS AND DISCUSSION

I. The Eccrine Unit:

A. Glycogen: Glycogen is present in both ductal and secretory coil cells of eccrine glands. Secretory cells have glycogen throughout their cytoplasm but the amount varies, certain cells having little, while the majority of the cells have large amounts (2, 3). Lobitz showed that the glycogen in secretory cells is very labile, disappearing under the stimulus of heat (4). In duct cells most of the glycogen is found in the peripheral or basal cells, while the luminal cells show only a fine line of glycogen in the cytoplasm next to the lumen. The total amount varies in different parts of the duct, luminal cells closely associated with the secretory coil in the deeper part of the dermis having less glycogen than similar cells in the mid-dermis level (1).

B. Non-Glycogen Carbohydrates: Intracytoplasmic granules are found in the small dark cells of the eccrine secretory coil (5, 6, 7). These have been reported to be PAS +, diastase resistant, and metachromatic to toluidine blue (6, 7). They become orthochromatic after profuse sweating (6). In our material these granules are dif-

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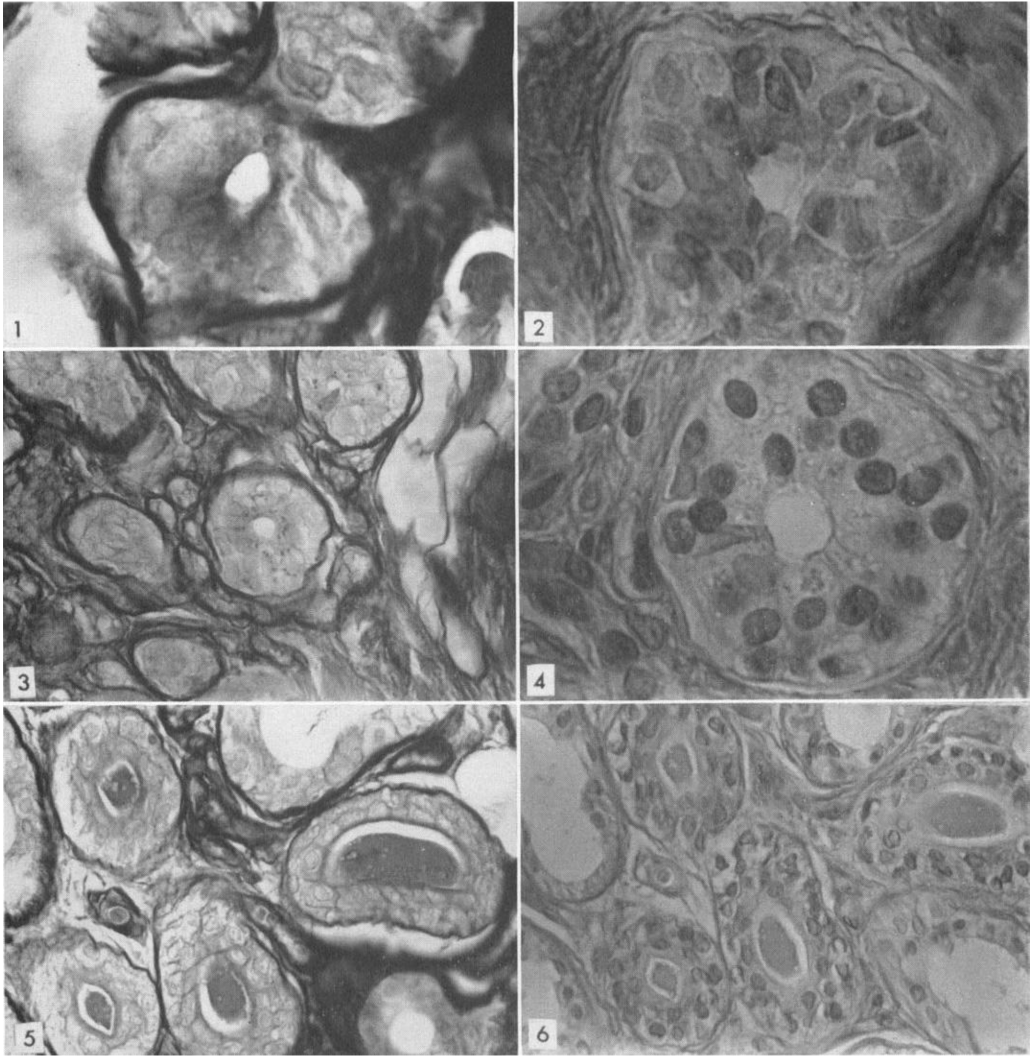

FIG. 1. Eccrine secretory coil; PAS after diastase digestion; 680 ×; PAS + D M intracytoplasmic granules of various sizes.

FIG. 2. Eccrine secretory coil; AB after diastase digestion; 680 ×; AB + intracytoplasmic granules

FIG. 3. Eccrine secretory coil; PAS after diastase digestion; 272 ×; PAS + D M large intracytoplasmic granules.

FIG. 4. Eccrine secretory coil; AB after diastase digestion; 680 ×; AB + large intracytoplasmic granules.

FIG. 5. Eccrine duct content; PAS after diastase digestion. 272 ×; PAS + D M luminal content.

FIG. 6. Eccrine duct content; AB after diastase digestion; 272 ×; AB + luminal content.

ficult to demonstrate. The majority of our specimens did not show them. When present, these granules vary in size from a fine haze of minute grains (Fig. 1, 2) to large granules, easily discernible (Fig. 3, 4). The variation in sizes did not allow segregation into groups; a continuous spectrum of sizes was present. Their position in the cytoplasm varied but usually they were present in the luminal apical portions of the cells but they were not necessarily limited to that location.

These granules stained PAS +, diastase resistant, magenta, (PAS + D M) (Fig. 1, 3) and AB + (Fig. 2, 4). These reactions correspond to the finding of Vialli (8) that Alcian Blue tends to follow the staining pattern of metachromatic dyes.

The lumens of eccrine ducts contained a PAS +

D M (Fig. 5) and AB + material (Fig. 6). Occasionally the luminal content were PAS negative and AB −, but this latter reaction was not common in our material.

II. The Apocrine Unit:

A. Glycogen: Glycogen is barely visible in the cytoplasm of apocrine secretory cells (3) and many authors (9, 10) have failed to observe it, but its presence is now well established. Apocrine duct cells are identical in their content of glycogen to those of the eccrine duct. The peripheral cells contain essentially all the glycogen.

B. Non-Glycogen Carbohydrates: Winkelmann and Montgomery (11), Winkelmann and Huttin (12), and Montagna (13) and co-workers (3) have demonstrated large PAS +, diastase resistant, non-metachromatic intracytoplasmic granules at the luminal end of apocrine secretory cells. In our specimens it was observed that these large granules, while most numerous at the luminal portion of the cells, may be seen in smaller numbers elsewhere in the cytoplasm. These granules are PAS +, diastase resistant, *red* (PAS + D R) (Fig. 7) and *fail* to stain with Alcian Blue (AB −) (Fig. 8). This is in contrast to the granules in the eccrine secretory cells, which are PAS + D M and AB +. In addition to the PAS + D R, AB − large granules, the apical luminal portion of the apocrine secretory cells has a haze of intracytoplasmic PAS + D M (Fig. 9), AB + material (Fig. 10). This material is distinctly different from the large granules.

The luminal contents of the apocrine ducts vary in their staining reaction. They may be either PAS + D M and AB +, or PAS + D R

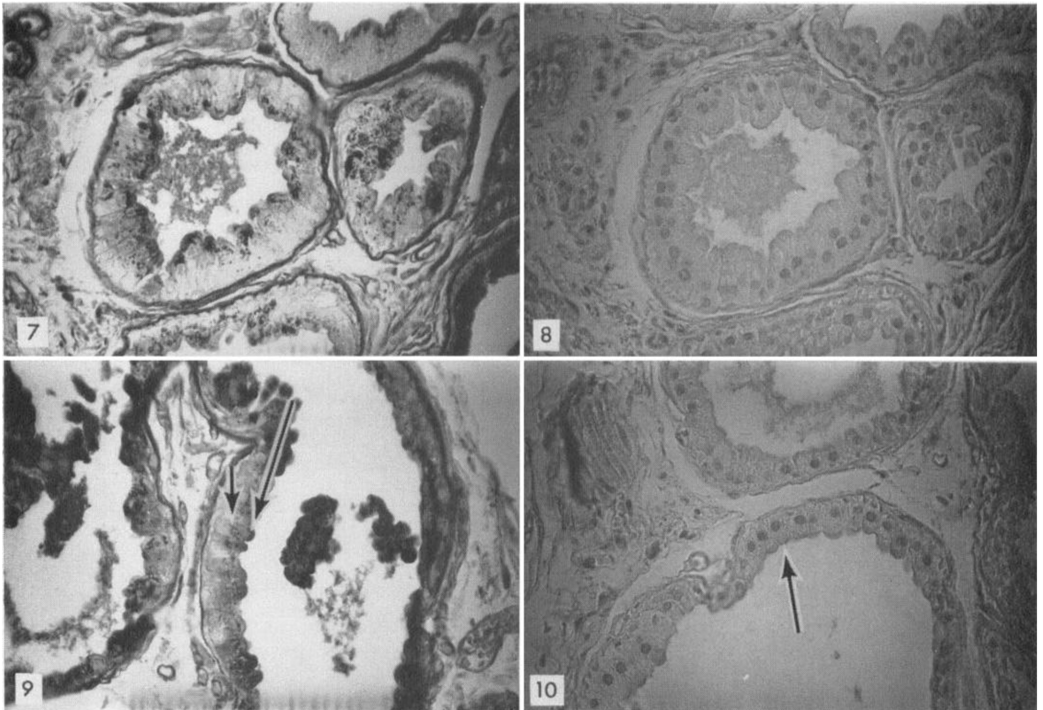

Fig. 7. Apocrine secretory coil; PAS after diastase digestion; 170 ×; PAS + D R intracytoplasmic granules.

Fig. 8. Apocrine secretory coil; AB after diastase digestion; 170 ×; intracytoplasmic granules do not take AB stain.

Fig. 9. Apocrine secretory coil; PAS after diastase digestion; 170 ×; long arrow indicated PAS + D M non particulate intracytoplasmic haze at luminal end of cell; short arrow indicates PAS + D R large intracytoplasmic granules.

Fig. 10. Apocrine secretory coil; AB after diastase digestion; 170 ×; arrow indicates AB + non particulate intracytoplasmic haze at luminal end of cell; note absence of staining (AB −) of the large PAS + D R granules.

and AB –, or even PAS – and AB –. This is in contrast to the eccrine duct contents which are almost invariably PAS + D M and AB + and only occasionally PAS – and AB –.

The histochemical PAS and AB reactions of the various cytoplasmic structures noted above are consistent. As noted recently, (1) those substances which react PAS + D magenta and AB + are probably complex acid mucopolysaccharides and acidic mucoproteins, therefore the PAS + D M and AB + granules of the eccrine secretory cells and the fine PAS + D M and AB + haze of the apical portion of the apocrine secretory cells represent compounds of this character. The PAS + D red and AB –, large granules of the apocrine secretory cells are either neutral mucopolysaccharides or neutral mucoproteins.

SUMMARY AND CONCLUSIONS

Eccrine and apocrine glands may be distinguished by their histochemical reactions to periodic acid-Schiff and Alcian Blue.

Eccrine secretory cells contain small secretion granules which stain magenta with periodic acid-Schiff and are Alcian Blue positive. Apocrine secretory cells, on the contrary, contain larger granules which are stained red by periodic acid-Schiff and are Alcian Blue negative. In addition the apical portion of the apocrine secretory cells shows a fine, apparently nonparticulate haze of periodic acid-Schiff + diastase resistant magenta Alcian Blue + material.

The luminal contents of eccrine glands stain magenta with periodic acid-Schiff and Alcian Blue + and occasionally are periodic acid-Schiff negative and Alcian Blue negative. The luminal content of the apocrine unit may be periodic acid-Schiff + diastase resistant red or magenta or periodic acid-Schiff negative and if periodic acid-Schiff + red it is Alcian Blue – and if periodic acid-Schiff + magenta it is Alcian Blue +.

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