CORPUS INTRA CRISTAM: A DENSE BODY WITHIN MITOCHONDRIA OF CELLS IN HYPERPLASTIC MOUSE EPIDERMIS

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This communication reports the occurrence of dense bodies within the cristae[1] of many mitochondria in the stratum germinativum and stratum granulosum of hyperplastic mouse epidermis. We have seen a variety of forms which suggest that these intramitochondrial bodies may be related to the so called keratohyaline granules of skin seen in the light microscope. A similar finding has been reported previously from electron microscope studies on experimentally induced squamous metaplasia of mouse cornea (13).

[1] The transverse lamellae of mitochondria appear in thin sections as two parallel dense lines which enclose a less dense area; they were designated *cristae mitochondriales* by Palade in 1952 (11).

MATERIALS AND METHODS

Ear epidermis of adult male Swiss mice was made hyperplastic by painting twice weekly with 5 per cent croton oil in mineral oil for 5 weeks. Specimens of such epidermis obtained from four animals were fixed either in buffered 1 per cent osmium tetroxide at pH 7.2–7.4 (17) or in similarly buffered 2 per cent potassium permanganate (6), and embedded in Epon by the method of Luft (7). Thin sections were stained with lead hydroxide (15) and examined in an RCA EMU 3E microscope.

OBSERVATIONS

Intracristal bodies may be seen occasionally in mitochondria of cells of the stratum germinativum. They appear as dense circular bodies which lie between the two dense lines of the cristae which contain them (Fig. 1). These dense lines of the cristae do not pursue their normal parallel course, but bulge around the dense body. There is usually a clear zone or halo about the corpus separating it from the dense lines of the cristae.

In the stratum granulosum the intramitochondrial bodies are of varying sizes and shapes, but in the deeper layers they are circular in profile. Most of the smaller corpora appear in mitochondria at the periphery of nuclei. More than one corpus may be seen within a single mitochondrion. The larger bodies often displace other cristae within the same mitochondrion (Fig. 2). We propose that this intracristal structure be called the *corpus intra cristam* to distinguish it from other bodies which have been described in mitochondria.

Because of the sequence of forms which can be seen in Figs. 1 to 6, we believe that the corpus intra cristam may contribute to the formation of keratohyaline granules We have observed that the corpora are sometimes completely surrounded by a membrane similar to the mitochondrial membrane, and sometimes are only partly enclosed (Fig. 4). In the stratum granulosum, many of the corpora lie free in the cytoplasm and appear larger than mitochondria, and oval or irregular rather than round. In osmium-fixed specimens, many of these larger granules have RNP particles at their surface and appear contiguous with bundles of tonofilaments (Fig. 6). This complex structure (2) may be the "keratohyaline granule" of the light microscopist.

DISCUSSION

There are several reports of experimentally produced alterations in mitochondrial structure. Swelling occurs as a non-specific change in normal cells after the blood supply is curtailed, and appears to be reversible if perfusion with oxygenated blood ensues (8). Thyroid hormone exerts a similar effect which is also reversible (4), and glutathione in homogenates also causes mitochondrial swelling (5).

Particulate matter within the matrix of mitochondria was first described by Sjöstrand and Rhodin in their observations on normal renal tubular epithelium (14). Similar dense particles have been seen in the matrix of mitochondria from many cell types. Weiss suggested that these granules represent intramitochondrial aggregates of cations (16). More recently ferritin (1) and silica granules (12) have been described within mitochondria, but none of these substances have been visualized within the cristae mitochondriales.

Granules which appear similar to those shown in the present communication were described within mitochondria by Nilsson in cuboidal epithelial cells of mouse uterus under estrogen stimulation (10). While they are surrounded by a single membrane, it is not clear whether they originated within the cristae in this instance.

The present observations and those on mitochondria in the stratum granulosum of experimentally produced squamous metaplasia of mouse cornea (13) suggest that there is a progressive loss of structural integrity of the mitochondria as the dense material accumulates within the cristae. Menefee has described an inverse relationship between the presence of mitochondria and that of keratohyaline granules in mouse epidermis (9).

It is interesting that in 1910 Favré and Regaud postulated a sequence of events, from light microscope observations, in which they stated that mitochondria were transformed into keratohyaline granules, and the process of keratinization began in the basal layer of the epidermis (3).

Because of our failure to demonstrate the corpus intra cristam in normal mouse skin, we hesitate to suggest that keratohyaline granules may be formed only by the growth of such a body. No chemical analyses of this body were attempted.

SUMMARY

The corpus intra cristam is a structure observed within the cristae mitochondriales of cells of hyperplastic epidermis. A similar structure has been observed in cells undergoing squamous metaplasia. As it enlarges it seems to destroy the surrounding mitochondrion. Once free in cytoplasm, it may act as a center of agglomeration of tonofilaments and RNP granules to form the so called "keratohyaline granule." Its contribution to the phenomenon of keratinization is not known.

Note added in proof: It has been brought to our attention that Setälä and others have observed similar intramitochondrial changes in mouse epider-

mis treated with carcinogens and considered them to be a specific change due to the carcinogen. Setälä, K., Merenmies, L., Niskanen, E. E., Nyholm, M., and Stjernvall, L., Mechanism of experimental tumorigenesis. VI. Ultrastructural alterations in mouse epidermis caused by locally applied carcinogen and dipole-type tumor promoter, *J. Nat. Cancer Inst.*, 1960, **25**, 1155.

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Figure 1

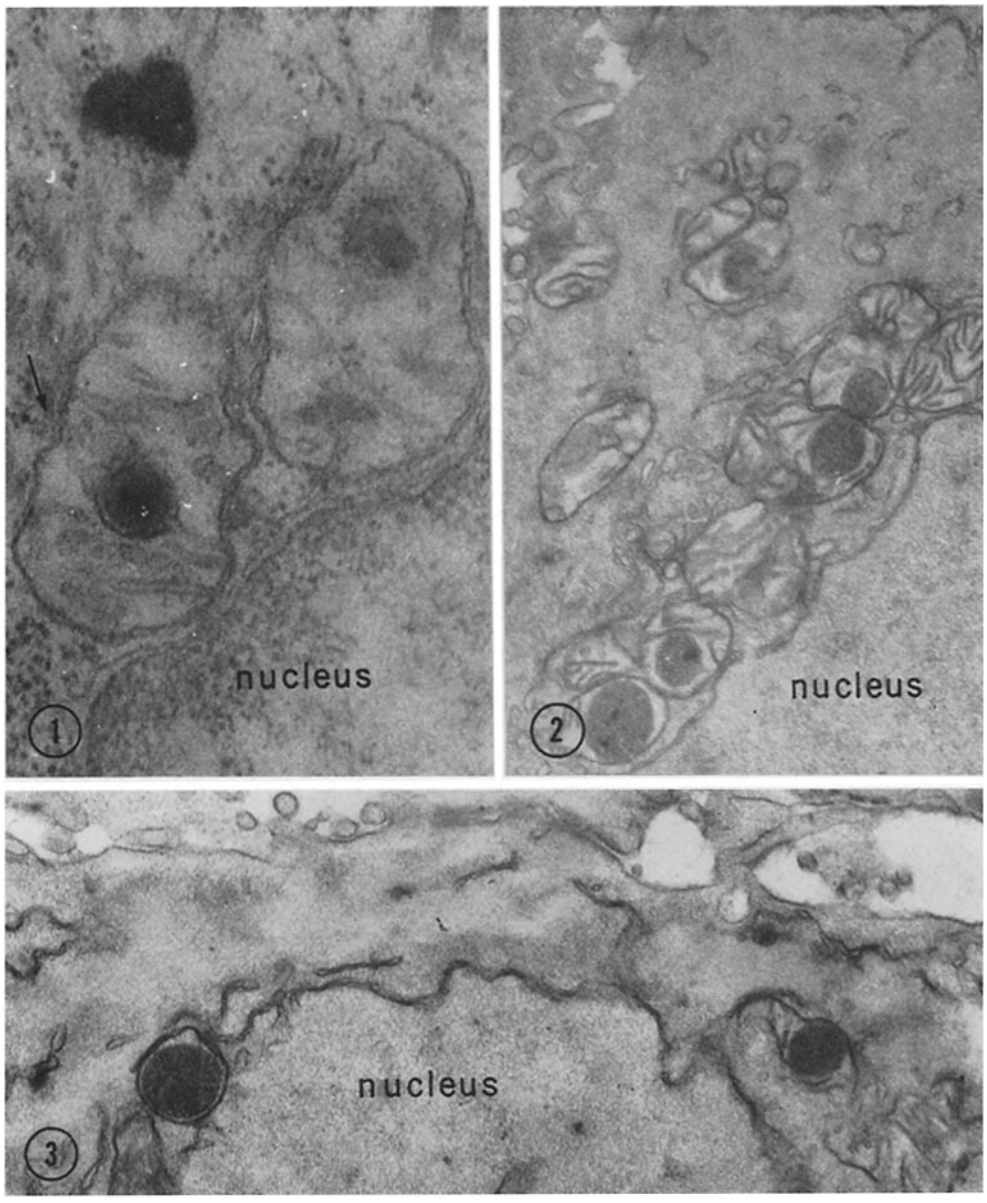
This electron micrograph of osmium-fixed tissue shows two mitochondria close to the nucleus. At the arrow is a dense body within one mitochondrion. This body we have designated a *corpus intra cristam* because it lies within the substance of the transverse lamella of a mitochondrion rather than in the matrix of the intercristal spaces. × 52,000.

Figure 2

This electron micrograph of permanganate-fixed tissue shows several corpora intra cristam in mitochondria at the edge of a nucleus. In several mitochondria the corpora are large enough to alter the external configuration of the mitochondrion and appear to compress other cristae within the mitochondrion. × 24,000.

Figure 3

This electron micrograph of permanganate-fixed tissue shows a corpus intra cristam filling the mitochondrion at the left and a smaller one on the right of the nucleus. × 26,000.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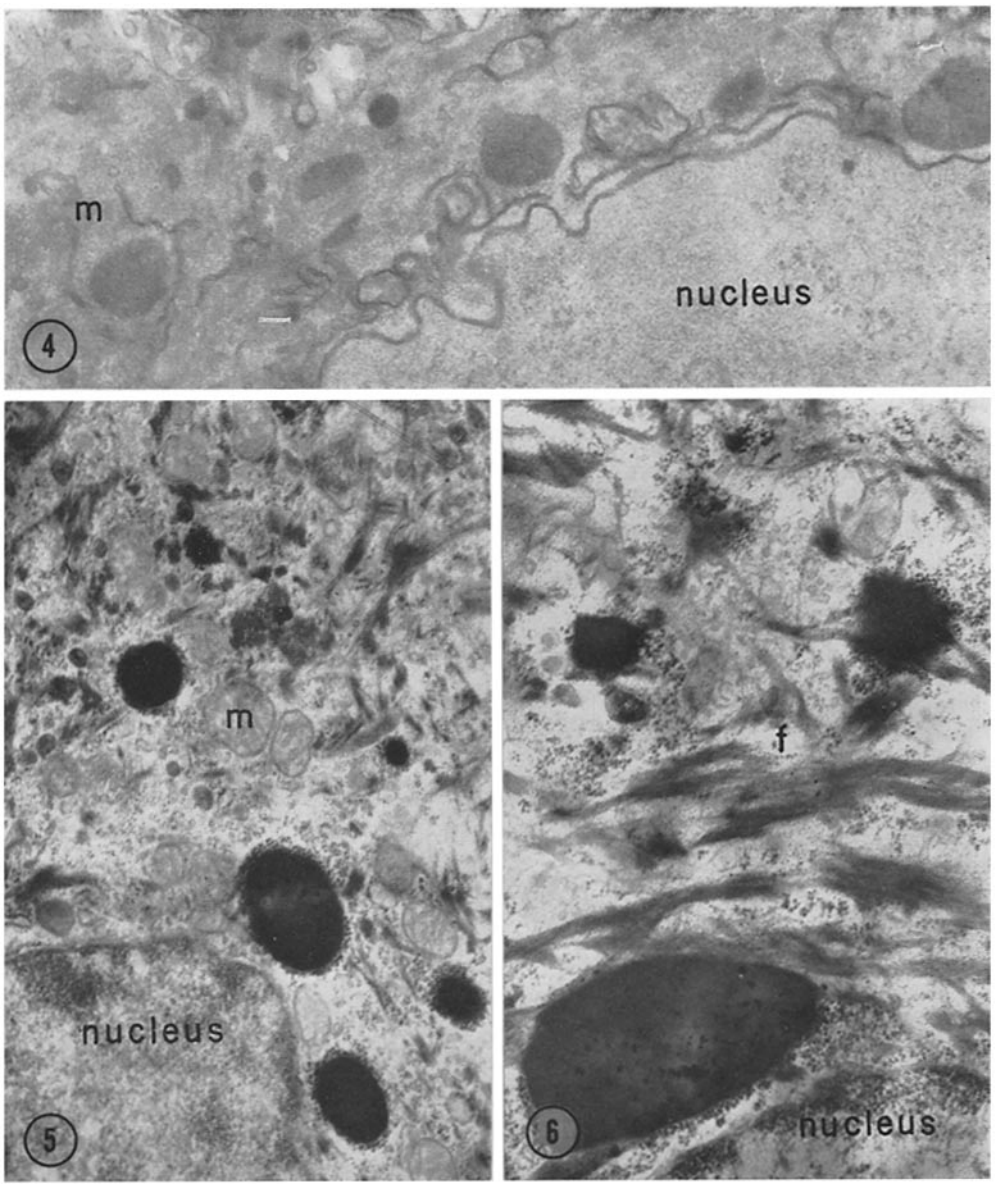

FIGURE 4

This electron micrograph shows a corpus intra cristam (at the left, under *m*) without enclosing membranes. Two other corpora are near the nucleus in the upper part of the picture. × 19,000.

FIGURE 5

This electron micrograph of osmium tetroxide–fixed tissue may be compared with Fig. 4. Here the small dense bodies are surrounded by RNP particles. These corpora closely resemble the large keratohyaline granule in the next picture. Several normal appearing mitochondria (*m*) are also present. × 22,000.

FIGURE 6

This electron micrograph of osmium tetroxide–fixed tissue shows a large keratohyaline granule at the bottom of the picture and two smaller granules in the upper part. Bundles of tonofilaments at *f* course through the cytoplasm. × 26,000.

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