Unusual Ultrastructural Variants in the Ferret Parietal Cell

The parietal cells of most mammalian species thus far studied have appeared morphologically similar[1-4]. The ultrastructural appearance of non-mammalian species[5,6] is generally similar to that observed in mammals except for the intracellular canalicular system, and absence of zymogen granules in mammalian parietal cells. Detailed accounts of the morphology of the ferret parietal cell are reported elsewhere[3,7]. However, several noteworthy, uncommon cellular organelles and inclusions have been discovered during extensive study of this cell type in the normal ferret, and are described here for the first time in this carnivorous species.

Materials and methods. Adult female, laboratory ferrets (Marshall Research Animals, Inc., North Rose, New York) were used in the present study. Observations reported herein were made in normal or, in the case of description of crystalloids, in drug-treated ferrets. Drug treatments consisted of administration of caffeine (12.5 mg/kg, i.p.), or acetylsalicylic acid (10 mg/kg, p.o.), and samples were taken 15 min after drug administration. The ferrets were maintained on a mixture of canned dog food (Ken-L-Ration) mixed with 25% mink ration (Purina), but were fasted (water ad libitum) overnight prior to sacrifice. Tissue samples were carefully removed from the anterior wall of the gastric fundus from ferrets lightly anesthetized with ether. The specimens were fixed overnight in 5% glutaraldehyde (phosphate buffered, pH 7.2), post-fixed in osmium by MILLONIG's[8] method, and embedded in Epon 812. Sections were double stained with uranyl acetate by WATSON's[9] technique, and lead oxide by KARNOVSKY's[10] procedure, and examined with a Jeolco Jem-7 Electron Microscope at 80 kV.

Observations. Crystalloids. In rare instances crystalloids were observed in the cytoplasm of ferret parietal cells following caffeine or acetylsalicylic acid administration, and appear either in aggregate (Figure 1) or isolated (Figure 2) form. Such crystalloids were not bounded by limiting membranes, nor were they preferentially located in any specific region of the cytoplasm. The subunits of the aggregate or the singular deposits were small, ranging from 0.4 μ to 1.0 μ, and appeared close to, but not associated with the mitochondria. When cross-sectioned (Figure 1), the spaces between the crystalline lattice approximated 122 Å, and the particle size was approximately 83 Å. In longitudinal section (Figure 2) the spacing was approximately 110 Å, and the particulate portion of the crystal was 62 Å.

Giant mitochondria. In general, the mitochondria of ferret parietal cells occupy a great proportion of the intracellular space, and contain a dense matrix and closely

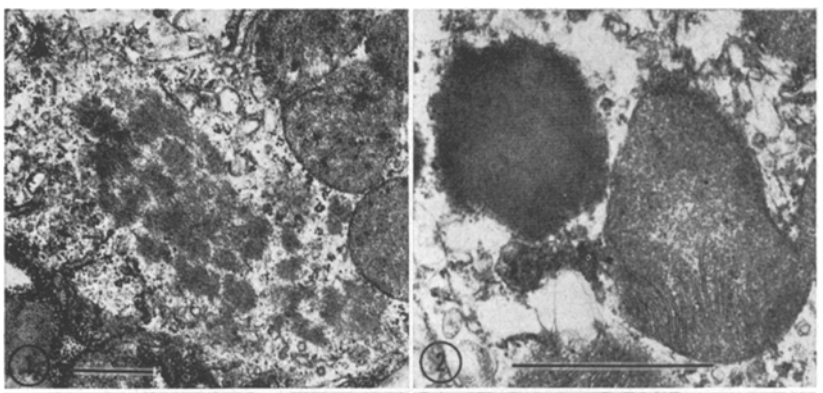

Fig. 1. Section through parietal cell showing aggregate of cytoplasmic crystalloids near mitochondria. Note varying orientation of crystalline lattices. This parietal cell was in an animal treated with caffeine (12.5 mg/kg, i.p.). × 13,200.

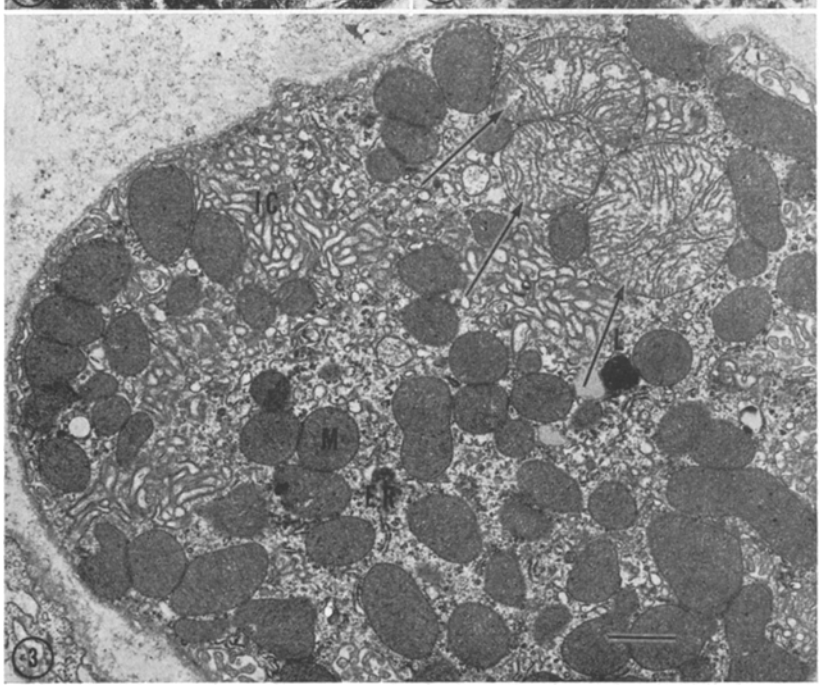

Fig. 2. Higher magnification of a single crystalline inclusion next to mitochondrion, showing parallel-line lattice with 110 Å spacing. This parietal cell was in an animal treated with acetylsalicylic acid (10 mg/kg, p.o.). × 34,000.

Fig. 3. Section through parietal cell showing 3 giant mitochondria (arrows) in cell containing numerous, normal-sized mitochondria. ic, intracellular canaliculi; m, normal mitochondria; l, lysosomes; er, rough endoplasmic reticulum. × 11,700.

packed membrane system. Giant mitochondria were observed, however, in rare instances. As shown in Figure 3, giant mitochondria can appear within an otherwise normal cell containing normal mitochondria. Such giant mitochondria may have diameters 2 or 3 times in excess of normal mitochondria, and lack the dense mitochondrial matrix characteristic of this cell. Although it appears that the number of crystae are not increased and that the internal membrane system remains intact in giant mitochondria, a degree of degenerative vacuolization is apparent.

Discussion. Crystalline structures are not commonly observed in parietal cells of any species. The only previous report of cytoplasmic crystalloids in parietal cells was that of WINBORN and BOCKMAN[4]. These investigators observed intramitochondrial crystals in hamster parietal cells, inclusions which appeared to be degenerative by-products of the mitochondria, and which were contained within a sectional area of mitochondrial size. In addition, HELANDER[11] recently reported the appearance of crystalline structures in the secretory product of parietal cells of weanling rats, a material which was present in the intracellular canaliculi and lumen of the fundic glands. Although the intracellular crystalline material observed in the case of the ferret may be the same as that previously reported[4], we have no evidence of it arising from mitochondria. Intramitochondrial crystalloids have also been reported in other cell types[12–14]. Crystal containing cells of the mouse large and small intestine have been reported[15], but such cell types are presumably of migratory origin[16]. The crystalline material observed in the ferret parietal cells is probably distinct from the oxyphilic substance noted many years ago by ZIMMERMANN[17], and considered as a precursor of hydrochloric acid. Indeed, it is currently held that hydrochloric acid is not stored, and is formed within the membrane system of the intracellular canaliculus.

Giant mitochondria have not been reported previously in parietal cells. The normal integrity of the parietal cell illustrated in Figure 3, as well as the conventional appearance of most of the mitochondria, suggests a pleomorphism of mitochondria in this cell. Such enlargement of mitochondria, observed here in an untreated ferret, has not previously been noted in our laboratory in ferrets treated by a variety of ulcerogenic drugs[18].

Résumé. On décrit deux rares variations structurelles: cristalloïdes cytoplasmiques intracellulaires et mitochondries géantes, observées dans les cellules marginals de la muqueuse gastrique du furet. Les cristalloïdes cytoplasmiques sont probablement en rapport avec la dégénération mitochondrielle, et les mitochondries géantes sont un caractère aberrant.

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Effect of Acriflavin on the Mitochondria of the Rat

Studies of the last few years have demonstrated that the antitrypanocidal drug acriflavin provokes alterations of the mitochondria in yeast[1,2] and of the kinetoplast and mitochondria in members of the family Trypanosomatidae[3–6]. To our knowledge no studies have yet been performed on the effect of this drug on the fine structure of mammalian cells. This fact induced us to find out if acriflavin could provoke in a higher species mitochondrial modifications similar to those reported in unicellular organisms.

Thirty male rats of the Wistar strain were injected i.p. with 10 mg/kg body wt. of neutral acriflavine (British Drug Houses Ltd.). The dose was chosen after a preliminary test of toxicity. Groups of 5 animals each were killed at 1, 2, 4, 6, 12 and 24 h after the injection of acriflavin. As controls 2 non-injected animals were used for each group. Samples of liver, heart and kidney were fixed in 1% osmium tetroxide in phosphate buffer and processed for the electron-microscopic study. Thin sections were stained with uranium acetate-lead citrate, with lead citrate alone or with potassium permanganate.

In the animals belonging to the 4 and 6 h groups, the fine structure of the parenchymal cells of the liver, kidney and myocardium did not show alterations and appeared normal according to currently accepted morphologic criteria. The only modification found consisted in the appearance of clear areas within the mitochondrial matrix. These electron-lucid zones were approximately circular in shape and measured 210 nm in mean diameter. Within the clear areas thick fibers from which emerged thin fibrils were frequently seen. The fibers stained heavily with uranium. In alternate sections stained with lead alone, or with potassium permanganate, the fibers were hardly apparent. Generally only one electron-lucid zone was seen per mitochondrion, but occasionally mitochondria with 2 clear areas were observed. The general morphology and the affinity for the uranium stain of the intramitochondrial fibers was similar to those reported in other plant and