Fine structure of the oesophageal and gastric glands of the red-legged pan frog *Kassina maculata* Dumeril

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Oesophageal and gastric glands of the anuran *Kassina maculata* Dumeril were studied using the electron microscope. The cells of the oesophageal glands contained abundant secretory granules, rough endoplasmic reticulum and mitochondria. The cells of the gastric glands were composed entirely of chief cells which contained abundant mitochondria, few secretory granules and rough endoplasmic reticulum. It is likely that in *K. maculata* the oesophageal glands produce more pepsinogen than the gastric glands.
S. Afr. J. Zool. 1982, 17: 28–31

Kliere van die slukderm en maag van die padda *Kassina maculata* Dumeril is deur middel van die elektronmikroskoop bestudeer. Die selle van kliere in die slukderm bevat baie sekretoriese granules, 'n growwe endoplasmiese retikulum en mitokondria. Selle van die maagkliere bestaan geheel en al uit hoofselle (chief cells) wat volop mitokondria, 'n klein hoeveelheid sekretoriese granules en 'n growwe endoplasmiese retikulum bevat. Dit is waarskynlik dat die slukdermkliere in *K. maculata* meer pepsinogeen produseer as die maagkliere.
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The occurrence and structure of oesophageal glands in some anurans from Tanzania have been studied previously (Hirji & Nikundiwe 1982). These and other observations (Jordan 1927; Reeder 1964; Porter 1972) suggest that the cells of the oesophageal glands of amphibians produce pepsinogen and are similar to the chief cells of the gastric glands. In order to clarify this relationship, the fine structure of the cells of the oesophageal and gastric gland was studied. The red-legged pan frog *Kassina maculata* which has well developed oesophageal glands (Hirji & Nikundiwe 1982) was selected for this study.

Materials and Methods

Small pieces of the oesophageal and gastric walls of a freshly killed *K. maculata* were fragmented with a sharp razor blade into small strips which were then fixed for 2 h in cold 2,5% gluteraldehyde solution buffered at pH 7,2 with 0,2 M sodium cacodylate. The tissues were rinsed in cold buffer and then postfixed for 2 h in cold 1% osmium tetroxide buffered at pH 7,2 with sodium cacodylate. After a brief rinse in the buffer, the tissues were dehydrated routinely and finally embedded in Durcapan ACM resin. Ultra-thin sections were cut with a Reichert OM U2 ultramicrotome, double stained on the grid with uranyl acetate and Reynold's lead citrate and viewed in a Carl Zeiss EM9-S2 electron microscope.

Results

Oesophageal glands

The secretory end pieces of the oesophageal glands were tubulo-acinar in shape. Each acinus was made up of four to five pyramidal cells with rounded bases and apices which enclosed a lumen.

Cell boundaries

At the free surface, the cells were studded with numerous microvilli. Laterally, the membranes of the adjacent cells were closely apposed and showed some interdigitation. The cell membrane rested on a thin basement membrane.

Endoplasmic reticulum

The rough endoplasmic reticulum (RER) was abundant and was laid down in parallel arrays of flattened cisternae (Figure 1). Various vesicular profiles of RER and free ribosomes were found amongst the cell organelles (Figures 2 & 3).

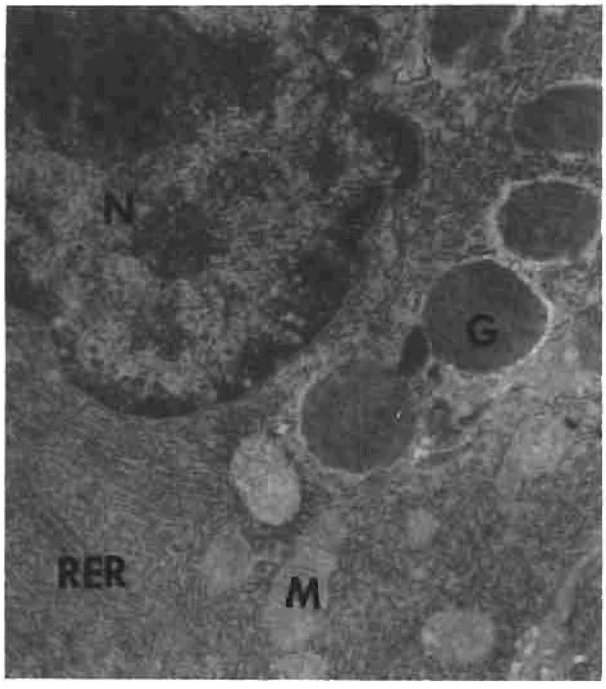

Figure 1 Maturing cell of the oesophageal glands showing general features. Note the extensive RER. N - nucleus, G - secretory granules, M - mitochondria. × 9500.

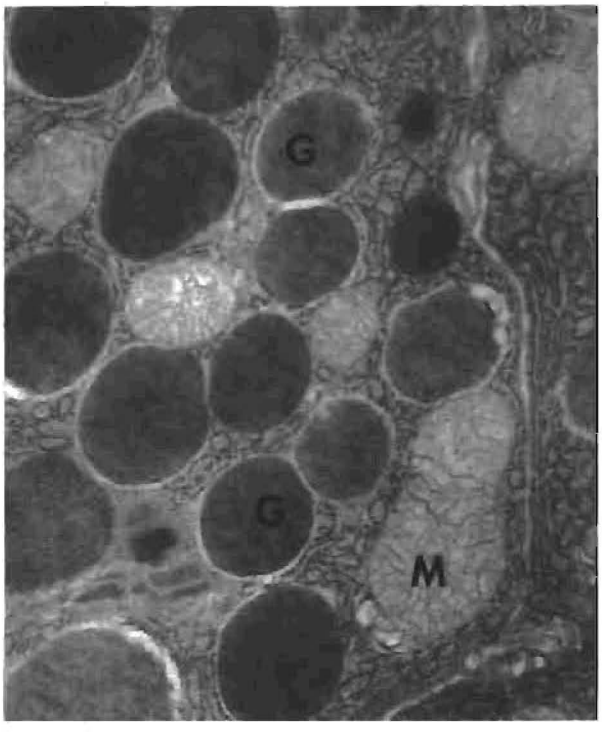

Figure 2 Mature cell of the oesophageal glands showing abundant secretory granules (G) and mitochondrion (M) with branching *cristae*. Note the various RER profiles amongst the secretory granules. × 9500.

Mitochondria

The oval-shaped mitochondria were sparse and their branching *cristae* traversed the entire width (Figure 2). The mitochondrial matrix was less electron-dense than the rest of the organelles.

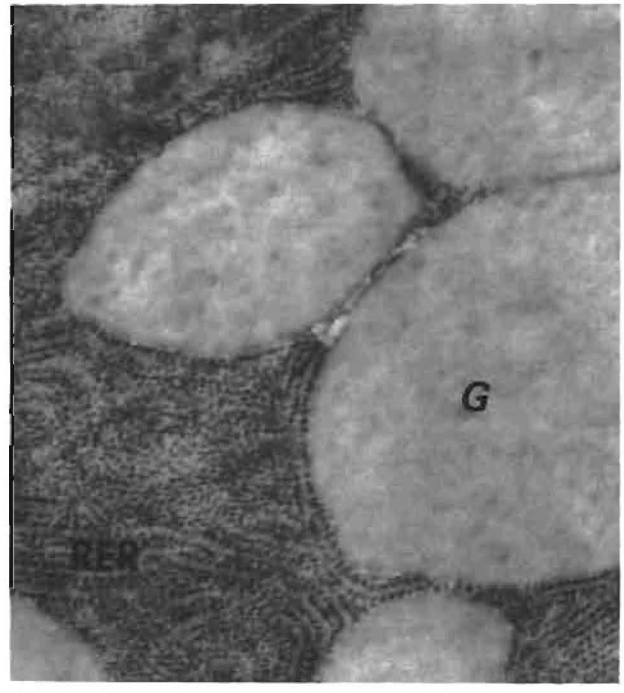

Figure 3 A cell of the oesophageal glands showing maturing secretory granules (G) and RER. Note that the secretory granules are closely associated with RER. × 28,000.

Secretory granules

Secretory granules of various sizes and electron-density were present in the cytoplasm. In the actively synthesizing cells (maturing cells) the granules were closely associated with the RER and together they occupied most of the cytoplasm (Figures 2 & 3). In the mature cells, the granules replaced most cell organelles. The granule contents passed into the lumen after fusion of their membranes with the outer limiting membrane of the cell and then rupturing at the point of fusion (Figure 4).

Nucleus

The roundish nuclei were situated in the basal region of the cell. Clumps of the electron-dense chromatin material in it were randomly dispersed and often clung to the inner surface of the membrane (Figure 1). The matrix was uniformly granular and of low electron-density. Nucleoli were not always visible in the nuclei.

Gastric glands

Chief cells were pear-shaped and their narrow parts enclosed the lumen of the gastric gland.

Cell boundaries

The long microvilli partially obliterated the lumen of the gastric gland. Laterally, the apposing cell membranes had junctional complexes. The apposing membranes interdigitated extensively as they approached the basement membrane.

Mitochondria

Mitochondria of various sizes and shapes were abundant in the supranuclear cytoplasm (Figure 5). Their *cristae* traversed the whole width of the mitochondria whose ground matrix was electron-dense.

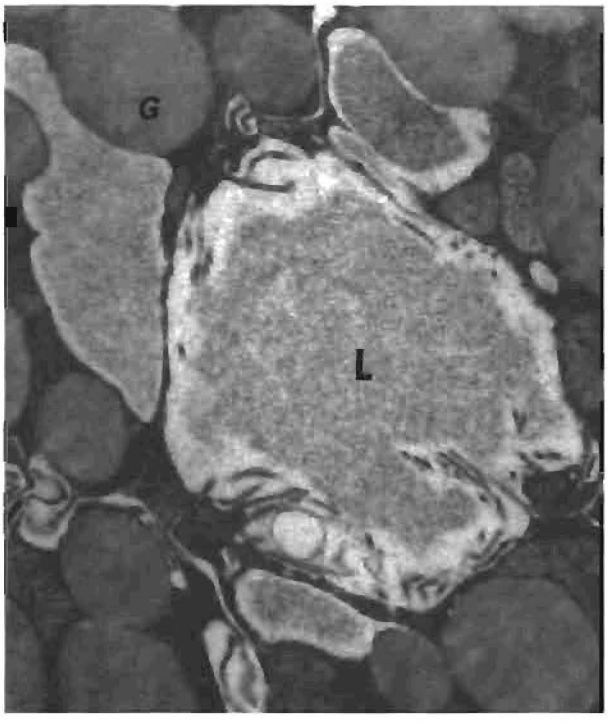

Figure 4 Secretory granules (G) being secreted into the lumen (L) of an acinus which already contains a copious amount of the secretion. × 9500.

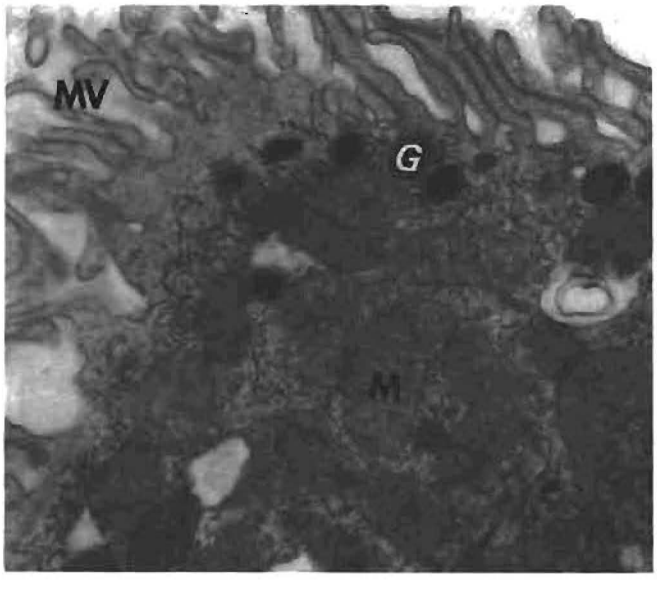

Figure 5 The supranuclear cytoplasm of the gastric cell. Note the well formed microvilli (MV) and abundant mitochondria (M). The few secretory granules (G) are electron-dense and small in size. Compare with Figure 2. x 9500.

Endoplasmic reticulum

The endoplasmic reticulum was predominantly of the rough type (RER) and was present as single randomly distributed flattened cisternae. Free ribosomes were present in the apical cytoplasm which also contained a system of randomly orientated smooth-surfaced vesiculotubules. These tubules were not present in the rest of the cytoplasm of the cell.

Secretory granules

Very few electron-dense secretory granules were present in the cytoplasm (Figure 5). The mode of their secretion into the lumen of the gastric gland was similar to that of the secretory granules of the oesophageal glands.

Nucleus

The oval nucleus lodged in the extreme basal region of the cell rested with its flattened surface on the basement membrane. Clumps of electron-dense chromatin were attached to the inner surface of the membrane and also randomly distributed in the finely granulated nuclear matrix. A centrally situated nucleolus was present.

Discussion

It is generally accepted that RER is best developed in cells elaborating a protein-rich secretory product. The maturing cells of the oesophageal glands of *K. maculata* contained a large amount of RER whilst in the matured cells, the RER together with the secretory granules were the predominant organelles. This is in accordance with the usual accumulation of RER in protein synthesizing cells such as the gastric chief cells and the pancreatic exocrine cells (Rubin, Jeffries & Sleisenger 1968; Murray 1970; Geuze 1971; Noaillac-Depeyre & Gas 1978). The present results confirm that the fine structural organization of the cells of the oesophageal glands and the gastric chief cells of *K. maculata* is for protein biosynthesis.

In *K. maculata*, the fine structure of the secretory granules from the cells of the oesophageal glands resembles, with the exception of size and electron-density, the fine structure of those from the gastric chief cells. Both granules also showed similar staining characteristics (Hirji & Nikundiwe 1982). Thus the secretory granules from the cells of the oesophageal glands appear to be similar to the pepsinogen granules of the gastric chief cells. Further studies are needed in order to establish the biochemical composition of the secretory granules of amphibians and provide a comparison with the pepsinogen from the gastric chief cells of other vertebrates (Samloff 1971).

Jordan (1927) observed that in some amphibians more pepsinogen was produced by the oesophageal glands than in the gastric glands. The relative quantity of secretory granules present in the cells of the oesophageal glands and the gastric chief cells shows that in *K. maculata*, more pepsinogen is produced in the oesophagus than in the stomach. Probably this observation is true for most Ranidae (Hirji & Nikundiwe 1982).

The gastric glands of *K. maculata* are composed entirely of chief cells. The presence of smooth-surfaced vesiculo-tubules and abundant mitochondria in the supranuclear cytoplasm is currently considered to be the major fine structural requisite for the production of hydrocloric acid (HCl) in the chief cells of the lower vertebrates (Rebolledo & Vial 1979). On morphological evidence, the gastric chief cells of *K. maculata* probably secrete HCl in addition to pepsinogen. Currently, the term oxynticopeptic cell is preferred for gastric cells responsible for the dual secretion of HCl and pepsinogen (Rebolledo & Vial 1979). The cells of the oesophageal glands on the other hand appear to be involved in the biosynthesis of pepsinogen which is activated by HCl in

the stomach to pepsin. The combined pepsinogen output of the oesophageal and gastric glands is therefore responsible for peptic digestion in the stomach of *K. maculata* and other amphibians which possess oesophageal glands.

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