world at much less cost with more happiness than tears around.

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SCIENTIFIC CORRESPONDENCE

Apoptosis as a novel mode of secretion in endocrine corpuscles of Stannius of a teleost (*Clarius batrachus*)

The corpuscles of Stannius (CS) are tiny endocrine glands located in or close to kidneys of holostean and teleostean fishes[1–3]. These glands are a source of stanniocalcin, a fast-acting homodimeric glycoproteinous hormone, which reduces the uptake of calcium from ambient water via gill and gut[4,5]. Recently stanniocalcin has been localized in tissues of other vertebrates including humans, where it functions as regulator of female reproductive system[6,7]. In spite of increased research on fish CS, some areas like cell types and mode of release of stanniocalcin are still debatable. We have, therefore, investigated the CS of an obligatory air-breathing freshwater catfish *Clarius batrachus* (Ham.) to collect more information.

Ultra-thin sections were prepared to study the cellular composition of the corpuscles of Stannius of *Clarius batrachus*. The CS from the kidney were excised and prefixed in cacodylate gluteraldehyde (buffered – 0.15 M, pH 7.2) solution at room temperature for 10 min. The tissue was then fixed in a similarly buffered solution of 1% osmium tetraoxide for 2 h at 0°C and then embedded in Epon. Ultrathin sections were stained with lead citrate and examined under electron microscope (Phillips EM 300).

The corpuscles of Stannius in *C. batrachus*, are small irregular shaped white bodies which lie on the peritoneal side in the posterior region of the kidney. At ultrastructural level (Figures 1 and 2) three structurally different cell types are clearly evident. The predominant cells (Type 1) have electron dense cytoplasm, large secretory granules and abundant endoplasmic reticulum. In contrast, the second type of cells (Type 2) are less numerous and are characterized by electron translucent cytoplasm, scarce membrane bound small granules. These cells possess unique cytoplasmic extensions amidst type 1 cells. We have also encountered a third type of cells (Figure 2) which can be identified by their dark colour, small size, shrunken nuclei separated by nuclear membrane and condensed cytoplasm. Furthermore, the most conspicuous feature of these cells is the presence of a large number of vacuoles of irregular shape which gives them an exhausted appearance. The large secretory granules are also separated from the surrounding membrane by wide space around them. The Golgi areas are scarce. The mitochondria are small and electron dense. The nuclei of these third type of

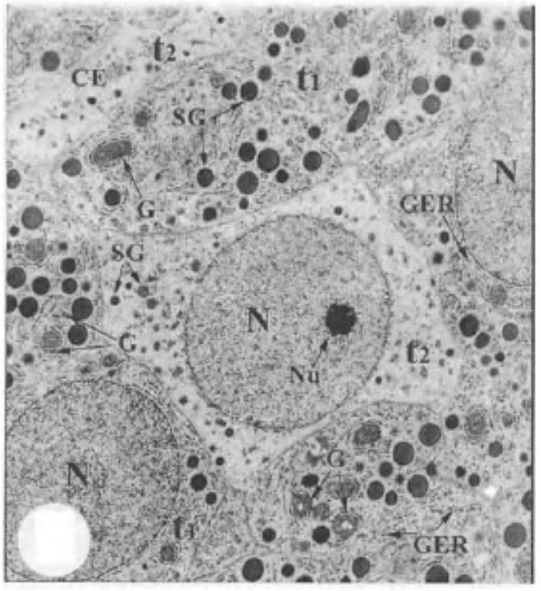

Figure 1. Section of CS of *C. batrachus*, showing two cell types. The type 1 (t_1) cells are characterized by large secretory granules (SG), abundant Golgi bodies (G) and granular endoplasmic reticulam (GER) in electron dense cytoplasm, whereas type 2 (t_2) cells possess electron translucent cytoplasm, small size secretory granules (SG), large nucleus (N) with dense nucleolus (Nu). (× 26400).

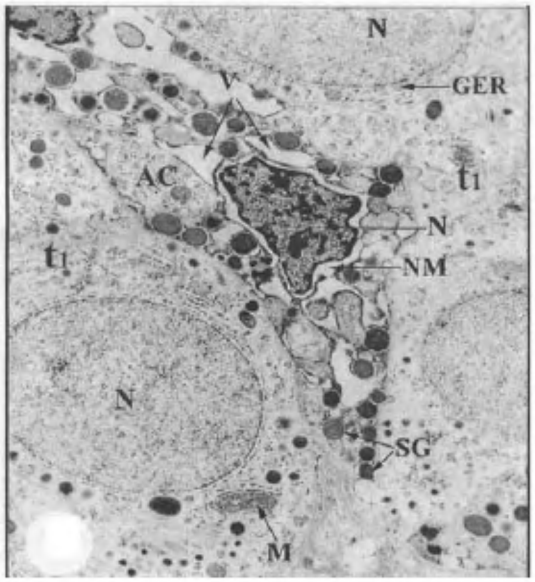

Figure 2. Section of CS of *C. batrachus*, showing apoptotic (AC) form of type 1 (t_1) cells. Note densely vacuolated cytoplasm (V), secretory granules (SG) with peripheral lumen, densified shrinking nucleus (N) separated from the nuclear membrane (NM). Note abundant granular endoplasmic reticulum (GER), (× 26400).

cells are multilobate and with amorphous chromatin patches.

Ultrastructural studies have revealed the presence of various cell types in the CS of fishes[1,2,4,8]. The general consensus is that the freshwater species possess two types of cells (Types 1 and 2) in their corpuscles while marine forms have just one. However, exceptions have been reported[9]. Dark cells similar to the present type III cells have also been reported[10,11] and the investigators have attributed these cells to be artifacts resulting from anoxia during fixation. One of the histological features directly related to the secretory activities of the CS as observed by many workers[5], is that it varies in different zones of the gland indicative of asynchronous secretion by gland cells. This feature has been attributed to a strategy by the gland for placing the burden of secretion on one region, while allowing the remainder to concentrate on renewed hormone synthesis. Another feature is the presence of degenerating or detached cell debris in the lobular lumen of the CS in light microscopical observations[11–13], suggesting holocrine secretion, an evidence that has never been confirmed at ultrastructural level. We suggest that these dark cells in the CS of *C. batrachus*, represent apoptotic degeneration[14], of type 1 cells rather than a specific cell type. Apoptosis represents a physiological mode of cell death. The sequential phases of apoptosis-like progressive cytoplasmic and nuclear densification, widening of the nuclear envelope, secretory granular membrane and also of GER membrane, are clearly evident in these dark cells. The apoptotic mode of release of the secretory product has not been reported by any endocrine cells and in the present case it is suggestive of high turnover by these cells.

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