

An Ultrastructural Study on Endocrine Cells in the Pyloric Region of the Korean Hedgehog(*Erinaceus koreanus*)

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한국산 고슴도치 유문부에 있어서 내분비세포의 전자현미경적 연구

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초 록

한국산 고슴도치 유문부에 있어서 내분비세포를 전자현미경적으로 관찰하여 EC, ECL, D1 및 G형 등의 4종류의 세포를 분류하였다.

EC세포는 고전자밀도의 다형태성 과립을 가지며, 과립내에는 더 높은 전자밀도의 dense body가 보였다. 과립의 크기는 160~530 nm였다.

ECL세포는 원형 또는 난원형의 과립을 가지며, 이들 과립내용물의 전자밀도는 높고, 편재하거나 공포상으로 출현하였다. 과립의 크기는 200~560 nm였다.

D1세포는 원형의 과립을 가지며, 과립의 전자밀도는 낮고, 때로 한계막과 과립내용물 사이에 좁은 halo를 나타내었다. 과립의 크기는 130~400 nm였다.

G세포는 원형 또는 난원형의 과립을 가지며, 비교적 낮은 전자밀도의 과립내용물과 한계막 사이에 좁은 halo를 보였다. 과립의 크기는 140~370 nm였다.

Introduction

Gastrointestinal endocrine cells of several mammalian species have undergone extensive ultrastructural study. As many as 18 different endocrine cell types have been identified in the mammalian gastroentero-pancreatic(GEP) endocrine system at the ultrastructural level (Solcia *et al.*, 1981a). However, only one study has described their ultrastructure in the hedgehogs (Matsuzaki *et al.*, 1985).

The present study describes the ultrastructural features of several endocrine cell types in the pyloric region of the adult Korean hedgehogs.

Materials and Methods

Adult Korean hedgehogs of both sexes were killed by ether anesthesia. For electron microscopy, 10 small pieces of the mucosa (about 2mm²) were taken from pyloric region. The samples were immediately immersed in 2.5% glutaraldehyde buffered with 0.1M cacodylate

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(pH 7.4) for 24hrs, washed in 0.1 M cacodylate buffer (pH 7.4), and then post-fixed in 2% OsO₄ in the same buffer for 2hrs at room temperature, dehydrated and embedded in Epon-812. Ultrathin sections were contrasted with uranyl acetate and lead citrate for electron microscopy.

Results

In the present study, 4 types of the endocrine

cells (gastric-type EC, ECL, D₁ and G cell) in the pyloric region of the hedgehog were classified on the basis of the ultrastructures of the cytoplasmic granules and designated according to the Santa Monica 1980 classification of gastroenteropancreatic endocrine-paracrine cells (Solcia *et al.*, 1981b). The ultrastructures of each endocrine cell types are summarized in Table 1.

Table 1. Ultrastructures of Secretory Granules in Endocrine Cells of the Pyloric Region of the Hedgehog (*Erinaceus koreanus*)

Cell types	Granular Size(nm)	Shape of Granules	Contents
EC (Gastric-type)	160~530	pleomorphic, dense bodies in dense matrix	high density, many granules
ECL	200~560	round or oval	high dense core and halo
D ₁	130~400	round	low density, sometimes dense core and clear halo
G	140~370	round or oval	low density, a few granules dense core and clear halo

The endocrine cells observed were primarily pyramidal or spherical in shape. The secretory granules generally were observed in the basal cytoplasm but on occasion were found in the supra- and para-nuclear cytoplasm as well. Depending on the cell type, the secretory granules were variable in size, electron density and internal structure.

The gastric-type EC cells were characterized by the presence of mostly pleomorphic granules showing a wide variation in size (160~530 nm) and in high electron density (Fig. 1). Some granules showed highly dense bodies in a dense matrix, and some ones also showed a clear narrow space between the core and membrane (Fig. 1a).

Granules of ECL cells were round or oval in shape with ranging from 200 to 560nm in a diameter (Figs. 2, 3). The granule contents with

a high electron density were located eccentrically in a limiting membrane with a wide space (Fig. 2, inset, Fig. 3). Some granules showed a small amount of content or empty.

D₁ cells were round or oval in shape. The granules of these cells were round in shape with ranging 130 to 400 nm in a diameter (Fig. 4). D₁ cells showed small and round granules with low electron density. Some granules showed occasionally a narrow halo between the limiting membrane and the electron dense materials (Fig. 4, inset). The microfilaments and a few lipid droplets were rarely seen in the cytoplasm.

G cells were characterized by the presence of round or oval granules with low electron density (140~370 nm in diameter, Figs. 5, 6). Some granules showed a narrow halo between the limiting membrane and the core (Fig. 6). A few

mitochondria, well-developed Golgi complex, and some rER were seen throughout the cytoplasm.

Discussion

Matsuzaki *et al.* (1985) reported 5 types of endocrine cells in the proximal duodenum of the echidna ultrastructurally. However, 7 types of endocrine cells, 5-hydroxytryptamine-, motilin-, gastrin-, somatostatin-, bovine pancreatic polypeptide-, cholecystokinin-, and substance P-immunoreactive cells, were identified immunocytochemically at the light microscopic level in the proximal duodenum of the echidna (Yamada *et al.*, 1985).

In the present ultrastructural study of the hedgehog pyloric region, 4 types of endocrine cells were classified by the ultrastructural characteristics of their secretory granules.

The ultrastructures of the hedgehog gastric-type EC cells were found to be identical with those of other mammalian gastric EC cells which contained many pleomorphic granules with high electron density (Capella *et al.*, 1969; Sasagawa *et al.*, 1970; Capella and Solcia, 1972; Sato *et al.*, 1978; Yoshino *et al.*, 1978; Rubin, 1972; Helančer, 1981; Oomori, 1983; Matsuzaki *et al.*, 1985; Lee and Lee, 1986). Some granules of EC cells showed a clear space between the core and membrane in the same cell which is in accordance with Oomori (1983) and Solcia *et al.* (1975). Furthermore, the granules of the hedgehog gastric-type EC cells often display the internal dense bodies in a dense matrix, which is in agreement with Capella *et al.* (1969), Vassallo *et al.* (1971), Capella and Solcia (1972), Solcia *et al.* (1975) and Oomori (1983).

In the present investigation, the ultrastructures of ECL cells were found to be identical with other mammalian ECL cells which contained relatively large, round or oval, and irre-

gular granules with high electron density or coarsely granular matrix eccentrically situated in the wide space (Forssmann *et al.*, 1969; Vassallo *et al.*, 1969; Capella *et al.*, 1969, 1971; Kobayashi *et al.*, 1971; Rubin, 1972; Lee and Lee, 1986). ECL cells were mainly recognized in the fundus and pylorus (Caella *et al.*, 1969, 1971; Vassallo *et al.*, 1971). It was reported that human ECL cells contained prominent filaments and glycogen bodies (Rubin, 1972), but such features were not observed in the hedgehog ECL cells.

In the present study, the ultrastructure of D₁ cells were shown to be identical with other mammalian D₁ cells which contained small, round secretory granules and low density (Sasagawa *et al.*, 1970; Kobayashi *et al.*, 1971; Capella and Solcia, 1972; Oomori, 1983). It has been observed that the abundant microfilaments in the cytoplasm (Sasagawa *et al.*, 1970; Kobayashi *et al.*, 1971; Oomori, 1983) and the secretory granules with a clear halo (Forssmann *et al.*, 1969). Also in the present observation, the features of the microfilaments were found in the hedgehog D₁ cells.

In the present investigation, the hedgehog G cells at ultrastructural level were generally in accordance with other mammalian G cells. It has been observed that the more or less microfilaments in the cytoplasm (Sasagawa *et al.*, 1970; Oomori, 1983) and the cytoplasmic process with microvilli reached the pyloric lumen (Vassallo *et al.*, 1969; Sasagawa *et al.*, 1970; Oomori, 1983), but such features were not observed in the hedgehog G cells.

These investigation described ultrastructurally 4 types of endocrine cells that are present in the hedgehog pyloric region. Previous immunohistochemical studies have identified the polypeptide hormones of some endocrine cell types

in the gastrointestinal mucosa of various mammals; EC cells producing serotonin, ECL cells producing histamine, D₁ cells producing somatostatin, G cells producing gastrin (Sundler *et al.*, 1976; Alumets *et al.*, 1977; Forssmann *et al.*, 1977). Also the differences between animal species and the functional roles of endocrine cells require further investigations.

Summary

Endocrine cells in the pyloric region of the hedgehog were studied ultrastructurally. 4 types of endocrine cells classified as gastric-type EC, ELC, D₁ and G cells were observed in this region. The gastric-type EC cells contained pleomorphic granules with high electron density and highly dense bodies in a dense matrix. ECL cells were characterized by the presence of round or oval granules with high electron density. Some granules of ECL cells showed a small amount of content or empty. D₁ cells contained round and small granules with low electron density. They occasionally showed a narrow halo between the limiting membrane and the dense materials. G cells were characterized by the presence of round or oval granules with low electron density. Some granules of these cells showed a narrow halo between the limiting membrane and the core.

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Figure Legends

- Fig. 1.** A gastric-type EC cell. $\times 12,000$.
Note the pleomorphic granules with high electron density.
- Fig. 1a.** Higher magnification of a basal portion of the EC cell. $\times 30,000$. Note the internal highly dense bodies in a dense matrix. A clear narrow space between the core and membrane is also seen (arrow).
- Fig. 2.** An ECL cell. $\times 12,000$.
Inset ($\times 30,000$) shows higher magnification of a part of the cell. Note the round or oval granules with high electron dense and a widely spaced membrane.
- Fig. 3.** An ECL cell. $\times 12,000$.
Note the granules showing a small amount of content or empty.
- Fig. 4.** A D_1 cell. $\times 17,000$.
Note the small and round granules with low electron density. Inset ($\times 30,000$) shows higher magnification of a basal part of the D_1 cell. The microfilaments are seen in the cytoplasm (arrow).
- Fig. 5.** A G cell. $\times 12,000$.
Note the round or oval granules with low electron density.
- Fig. 6.** Higher magnification of a part of the G cell. $\times 30,000$. Note the granules with a narrow halo between the limiting membrane and the core. Well-developed Golgi complex is also seen.





