ENDOCRINE CELLS IN THE ALIMENTARY TRACT OF THE SHEEP

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The isolation and purification of peptide hormones from the alimentary tract of several species has enabled the production of antisera specific to parts of these peptides which can be used for immunocytochemical localization of the secretory cells producing the different peptides. This report concerns the presence and localization of endocrine cells in the alimentary tract of the sheep which have been identified by indirect immunofluorescence using an antiserum specific to the C-terminal part of gastrin and cholecystokinin (CCK) and another antiserum to gastric inhibitory polypeptide (GIP).

Material and Methods

TISSUE. — Sheep were killed by intravenous injection of sodium pentobarbitone. For gastrin/CCK histochemistry, whole body perfusion with 5-8 l normal saline followed by 5 l phosphate buffered formalin (10%) saline was usually carried out before removing pieces of tissue for secondary fixation in Bouin's solution. For GIP investigations, fresh tissue was fixed immediately in Bouin's fluid for 12-24 h. Fixed tissues were processed to paraffin wax (56 °C melting point) and 6μM sections were cut with a Spencer rotary microtome.

ANTISERA. — C-terminal porcine gastrin antiserum (rabbit) was generously provided by Professor R.A. Gregory and Dr G.J. Dockray of the University of Liverpool, who also supplied pure porcine gastrin for specificity controls with the antiserum. GIP antiserum (rabbit) was generously provided by Professor V. Marks of the University of Surrey.

Fluorescein isothiocyanate conjugated goat anti-rabbit IgG (FITC-IgG) was purchased from Miles Laboratories (UK) Ltd., Slough, Bucks.

Bovine serum albumin was purchased from Sigma Chemical Co., London.

IMMUNOFLUORESCENCE. — Dewaxed sections were washed with phosphate buffered saline (PBS) at pH 7.4 and stained by an indirect method of immunofluorescence (Coons, 1956).

Gastrin and CCK: Sections were incubated with gastrin antiserum diluted to 1:200 with bovine serum albumin (2%) in PBS for 30 min at 27 °C. Sections were washed with PBS and then incubated with FITC-IgG (1:10 dilution in PBS) for 15 min at 27 °C. Finally, sections were washed with PBS and mounted in phosphate buffered glycerol for examination under the fluorescence microscope.

GIP: Sections were incubated in a moistened chamber with GIP antiserum diluted to 1:1400 with bovine serum albumin (2%) in PBS for 48 h at 4 °C. Final washing and mounting were as for gastrin-CCK sections.

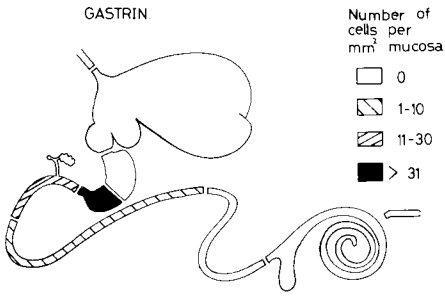

Fig. 1. — Diagram of the localization of gastrin-like immunoreactivity in the alimentary tract of the sheep.

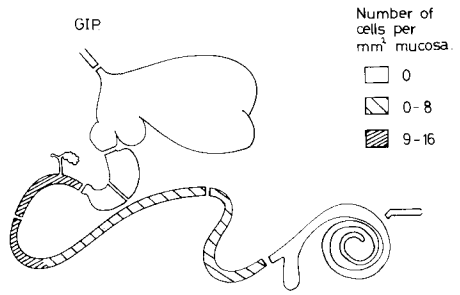

Fig. 2. — Diagram of the localization of GIP-like immuno-reactivity in the alimentary tract of the sheep.

Results and Discussion

GASTRIN-CCK. — Sheep gastrin has the same five C-terminal amino-acids as porcine gastrin and, because CCK also has the same C-terminal amino-acids, the porcine gastrin antiserum would be expected to bind to sheep cells secreting gastrin or CCK. The distribution of cells having immunoreactivity to this gastrin antiserum is shown in figure 1. The cells occurred throughout the mucosa of the abomasal antrum and pylorus and, on the basis of their distribution, it is assumed they are gastrin secretory cells. They had a characteristic pyramidal shape with an apical pole projecting into the lumen of the glandular crypts, as illustrated in plate 1. The distribution of these abomasal cells is similar to that first described by McGuigan (1968) in human and porcine stomach material.

Specifically staining cells were also found scattered through the crypts, villi and Brunner's glands of the upper small intestine. It is probable that these cells represent a mixture of gastrin and CCK secretory cells.

GIP. — To our knowledge, sheep GIP has not yet been isolated and purified so that the observations with porcine GIP antiserum have to be qualified accordingly. Nevertheless, specifically immunoreactive cells were found in the upper small intestine of the sheep as illustrated in figure 2. The cells were similar in appearance to those first described by Polak *et al.* (1973) in the intestine of man and dog. A typical example is shown in plate 2. GIP cells had a characteristic tear-drop shape with an apical pole projecting into the gland lumen. They were predominantly localized in the deeper parts of the crypts of Lieberkuhn.

N.W.B. is an A.R.C. postgraduate scholar.

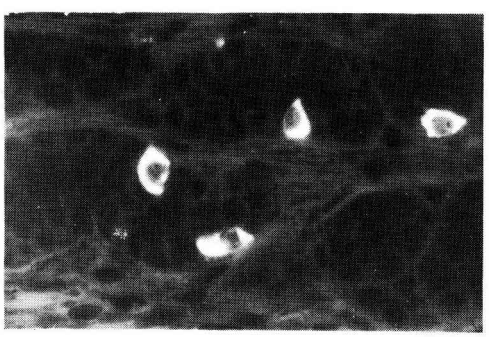

Plate 1. — Gastrin cells in the mucosa of the abomasal antrum in the sheep. x 400 (Ilford film FP40, 30 s exposure).

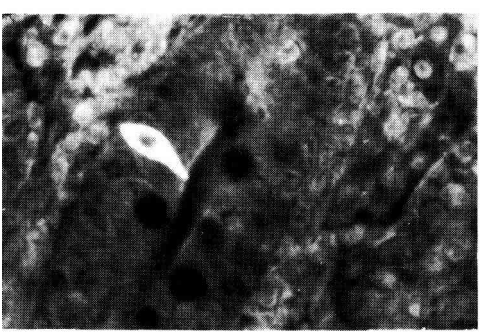

Plate 2. — GIP cell in a crypt of Lieberkuhn of the duodenum in the sheep. x 400 (Ilford film FP40, 30 s exposure).

References

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