

HISTOGENESIS OF PANETH CELLS IN THE HUMAN SMALL INTESTINE.

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ABSTRACT: The epithelial lining of the small intestine consists of various types of cells. Paneth cells, which represent one of the major epithelial cell group, are abundantly present in the depths of the intestinal glands. However, they are not present in the early weeks of foetal development. Limited number of literatures in the past have mentioned about the time of appearance of Paneth cells. The present study aims to find out the time of appearance and the gradual histological changes of the Paneth cells in the small intestine of human foetuses.

KEY WORDS: Paneth cells, immune system, secretory granules, time of appearance.

INTRODUCTION: The epithelium of the small intestine is of endodermal origin. The endodermal cells differentiate into various types of secretory and absorptive cells. Remaining layers are derived from splanchnic mesoderm (1). Paneth cells differentiate at the base of the crypts at about 11 and 12 weeks of gestational age (2). Intestinal cells are constantly renewed with a constant migration of cells from the base of the crypt to the villi. Paneth cells, however, are not observed in regions above the base of the crypt (3). In normal and transgenic mice, cryptdin-positive cells having morphologic appearance of Paneth cells, with apical secretory granules demonstrable by Phloxine-Tartrazine stain were observed on the 7th postnatal day (4).

Paneth cells are numerous in the deeper parts of the intestinal crypts, particularly in the duodenum. They are pyramidal in shape with a round or ovoid nucleus situated near the base. The basal cytoplasm is basophilic and the numerous secretory granules at the cell apex stain with acid dyes. Paneth cells are rich in Zinc and they also secrete Lysozyme, an antibacterial substance. They are long lived and are not observed in mitosis (5). They may occur in the stomach and large bowel in certain pathological conditions: for example they are frequently present in the colonic mucosa in ulcerative colitis (6).

Paneth cells release granules into the lumen of the crypts of Lieberkuhn in the small intestine where their component proteins participate in mucosal immunity (7). The granules contain a number of proteins associated with roles in host defense including lysozymes, secretory phospholipase A2, and alpha-defensins termed cryptdins.

MATERIALS AND METHODS: The present study was carried out on 36 normal human foetuses of various gestational ages ranging from 9 weeks to term. Depending on the gestational ages, the

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foetuses were then divided into groups to determine the period at which specific changes occurred. The number of foetuses in each group was as follows:

1. 9-12 weeks – 6
2. 13-16 weeks – 10
3. 17-20 weeks – 12
4. 21-24 weeks -- 8

The age of the foetuses were calculated from obstetrical history and by measuring the crown- rump length (CRL). The foetuses were collected from the Department of Obstetrics and Gynaecology and PPP Centre, RIMS, with due permission from the Medical superintendent and proper consent from the concerned party. The consent obtained was collectively for all postgraduate students undertaking Thesis works in the Institute during that particular period (2005-2007). The specimens collected were mostly stillbirths and a few were the products obtained after induced abortions (under the provisions of the MTP Act of India, 1971).

Specimens collected were subjected to tissue processing. Primary fixation was done in 10% Formalin for a period of 7-10 days followed by secondary fixation in neutral buffered Formalin. After satisfactory fixation, the tissues were trimmed and washed in 50% alcohol. This was followed by the normal procedure of dehydration, clearing, paraffin embedding, paraffin block making, sectioning, mounting, rehydration and staining. The Rotary Microtome was used for making serial sections of the specimen.

The histological stains used for the study were:

1. Haematoxylin and Eosin stain
2. Phloxine-Tartrazine stains for Paneth cell granules.

DISCUSSION: Paneth cells were described as pyramidal cells with round or oval nuclei. They were seen in the lower half of the intestinal crypts along with less differentiated cells (8). Paneth cells contain numerous secretory granules at the cell apex, which stained with acid dyes (9, 10). The population of Paneth cells in an intestinal crypt at a particular time was about 20 with a turn over time of 3 weeks (11). Paneth cells may occur in the stomach and large bowel in pathological conditions (5). They exhibit intensely eosinophilic granules with H &E stain. These cells were even more clearly demonstrated by Phloxine-Tartrazine method, which stained the granules scarlet red (12).

Paneth cells appeared at 11-12 weeks of foetal development. They were seen at the bottom of the intestinal crypts (2). In developing mice, Paneth cell granules were positive for Phloxine-Tartrazine stain only at 7th postnatal day (4).

In the present study, Paneth cells were detected at 14 weeks. However, proper demonstration of supranuclear cytoplasmic granules with Phloxine-Tartrazine stain was possible only at 24 weeks, and at this age they were comparable to the histological appearance of adult Paneth cells.

CONCLUSION: In the early stages of development i.e. upto about 9 weeks, the epithelial lining of the human small intestine was simple columnar in almost all places. Few areas showed pseudostratified columnar epithelium. Paneth cells appeared at 14 weeks of gestational age. Proper demonstration of apical cytoplasmic granules was possible at 24 weeks.

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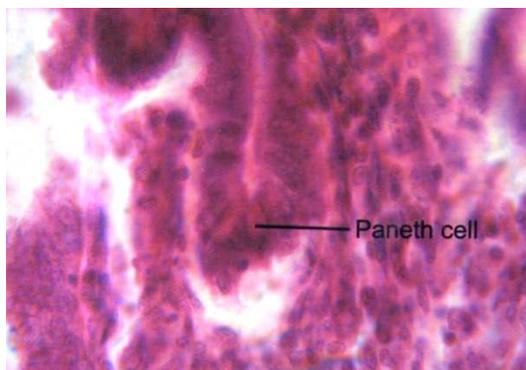
RESULTS AND OBSERVATIONS: The histological staining procedures have given the following results as shown in the various photographs and the adjoining text boxes below.



9-12 weeks: The epithelium in this group was simple columnar in most areas. In some places the epithelium appeared pseudostratified columnar as seen in Fig.1. No cells resembling Paneth cells were observed.

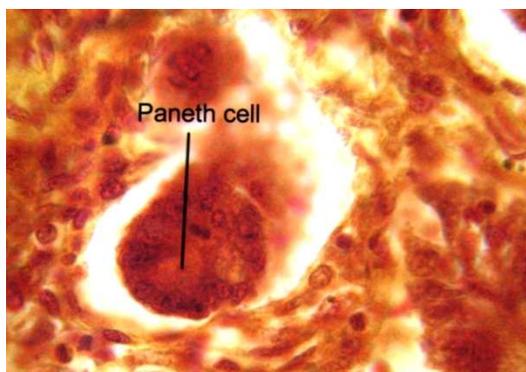
Fig. 1 Human small intestine at 9 weeks (H&E Stain)

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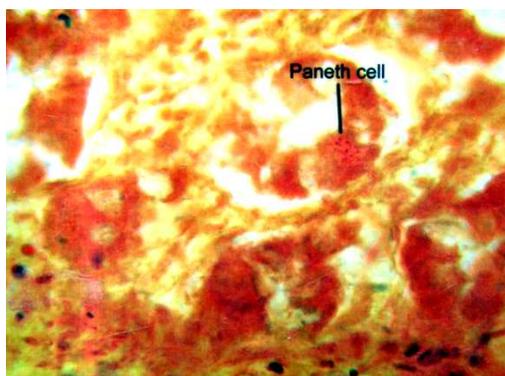
13-16 weeks: Paneth cells were seen at the base of the intestinal glands. They were pyramidal in shape with the nuclei situated basally. However, the apical cytoplasm could not be stained for any secretory granules (Fig. 2).

Fig. 2 Paneth cells at the depth of the crypt of Lieberkuhn (H&E stain).



17-20 weeks: With Phloxine-Tartrazine stain, Paneth cells were observed in the crypts of the intestinal glands. The cells still did not show secretory granules (Fig.3).

Fig. 3 Paneth cells in the intestinal glands at 16 weeks (Phloxine-Tartrazine stain)



21-24 weeks: Paneth cells in this age group were positive for secretory granules which stained bright red with Phloxine-Tartrazine as shown in Fig.4.

Fig. 4 Paneth cells showing secretory granules in a 24 weeks foetus (Phloxine-Tartrazine stain)