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Case report

Conspicuous enterocytic binding pattern for peanut lectin and malignant histiocytosis of the intestine

Biopsy examination and autopsy findings of a 33-year-old man with glutensensitive enteropathy

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Summary. We present a case of a 33 year old male with a history of early childhood diarrhoea and more recently diagnosed gluten sensitive enteropathy, who died with active disease, ulcerative proctosigmoiditis and desquamative erythrodermia associated with toxin induced shock. Autopsy revealed a tumour restricted to lymph nodes of the mesentery and the retroperitoneum. This is considered to be malignant histiocytosis of the intestine (MIH). Immunohistological examination of the diagnostic jejunal biopsy showed a pathological binding pattern for peanut lectin (PNL) within the enterocytes. This may be an expression of disturbed production or secretion of a product rich in non-reducing terminal D-galactosyl residues.

Key words: Coeliac disease – peanut lectin – malignant histiocytosis of the intestine

The aetiology and pathogenesis of gluten sensitive enteropathy is still a matter of discussion. Findings which suggest a primary immunopathic process are confronted by increasing amounts of data suggesting that the toxic gliadin fractions might only on the basis of an enterocytic defect interact with the gut associated lymphoreticular system. In this case of coeliac disease with numerous related complications and a lethal outcome there was a conspicious enterocytic binding pattern for PNL, a lectin specific for non-reducing terminal D-galactosyl residues (Uhlenbruck, et al. 1969; Lotan et al. 1975). This may be an indication of disturbed enterocyte function in this disorder.

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Clinical history

In may 1980, 20 months before death, a 32 year-old farmer K.H.Z. complained of an increased frequency of diarrhoea and abdominal spasms which had bothered him for years. His clinical history included growth retardation dating from the 7th month of life with regular diarrhoea for the first 2 years and repeated pneumonia during childhood.

Symptomatic therapy was ineffective, x-ray-examinations 6 months later gave no positive results. In the course of hospitalization a jejunal biopsy was taken and the diagnosis "probably gluten-sensitive enteropathy" was made. The patient followed a glutenfree diet for 3 months without significant improvement. Some time later a perioral skin eruption was interpreted as dermatitis herpetiformis (Duhring). In the course of his holiday in Greece the patient reported having felt "better than ever", however, coming back home, abdominal pain and frequent diarrhoea reappeared.

Meanwhile a progressive weight loss became evident (II/80: 74 kg; II/71: 66 kg; IX/81: 60 kg). 4 months before his death, he was hospitalized for frequent watery diarrhoea, sore thorat, vomiting and crural cramps. Under intravenous nutrition the clinical picture improved rapidly. Diagnostic gluten exposition was followed by a relapse with diarrhoea and acidosis (e.g.: pH: 7,2; BE: −11) so severe that the ion balance could only be reinstated with great difficulty. After a short time he was discharged with an extremly strict diet. Re-admission because of an alleged dietary mistake was necessary only 6 weeks later. At the time of admission his body weight was 56 kg, pH 7.1 and BE −16.6. Body-temperatures (more than 39° C) in the course of parenteral nutrition responded well to antibiotics. Fecal culture revealed Klebsiellae. The last 10 days were nevertheless characterized by severe hyperpyrexia whereas the white blood counts never exeded 8,000/µl. Gastroscopy revealed a marked change in the duodenal mucosa with loss of Kerckring's folds, duodenitis and a duodenogastrical reflux. Duodenal biopsy led to the histological diagnosis of chronic duodenitis with villous atrophy and goblet cell depletion. Coloscopy showed numerous coin-shaped ulcera of the sigmoid an the rectum, which spread in the following days. Histology proved an acute and non-specific procto-colitis. Enterococci could be demonstrated in smear-culture. During the last few days a desquamative erythroderma appeared. A state of shock with an increasing respiratory distress lead to death by hypoxemic heart failure at Jan. 1st, 1982.

Autopsy

The autopsy of a malnourished young man with severe desquamative erythroderma showed a conspicious small intestine with coarse mucosal folds, mosaic-like aspect of the thickened mucosa (Fig. 1) and prominent Peyer's patches. The thickened root of mesentery contained enlarged tumourous and amorphous lymph nodes (Fig. 2). The para-aortic, peripancreatic and portal lymph nodes were likewise affected. Numerous coin-shaped mucosal ulcerations were found in the sigmoid colon and the rectum. The liver showed a moderate fatty infiltration, the lungs had a marked chronic oedema, the myocardium was pale and flabby and showed dilatation. Signs of congestion were evident. The kidneys showed cloudy swelling.

Material and methods

Apart from histological routine staining 2–4 µ thick paraffin sections from biopsy and autopsy material were immunostained by means of commercially available antibodies (Dako, Denmark; Nordic Immunology, Netherlands; E.Y. Lab. Inc. California) using the peroxidase-antiperoxidase technique for detecting immunoglobulin fractions (γ-, α-, μ-, ε-, κ-, λ-, and J-chains), lysozyme, and binding sites for PNL. Incubation of slides with the lectin was done according to Klein et al. (1981), the fixed PNL was localized by means of an unlabelled anti-PNL as first step antibody of the conventional PaP-method (Möller 1982a). PNL reactivity to the tissue was blocked by adding D-Galactose to the lectin solution. Controls done by omitting the lectin or the anti-lectin gave negative results.

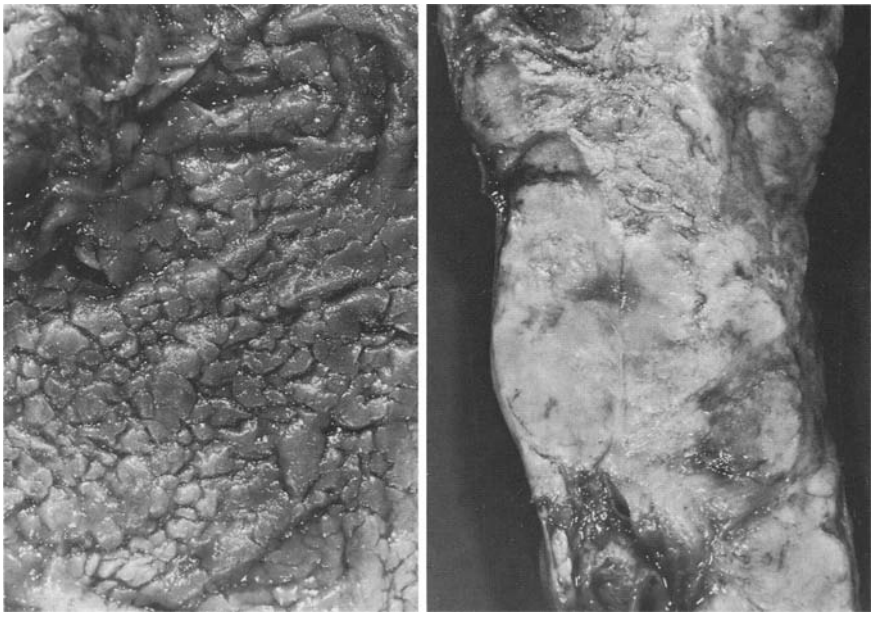

Fig. 1. Proximal jejunum: loss of Kerckring's folds, tyical mosaic-like aspect (approx. ×2.2)

Fig. 2. Cross-section of the root of the mesentary tumourous infiltration of the lymph nodes and the adipose tissue (approx. ×2.2)

Histological results

Histological examination of the diagnostic jejunal biopsy taken 8 months prior to death reveals changes corresponding to the postmortem findings: villous atrophy, crypt hyperplasia, goblet cell reduction, a raised mitotic rate among the enterocytes and a dense and polymorphic inflammatory infiltration of the tunica propria (Fig. 3). The plasma cells display a polytypic staining pattern, the number of immuno-stained cells per area decrease in the following order: $J > \alpha > \lambda > \kappa$ (Fig. 3b) $\gg \mu \gg \gamma = \varepsilon$. Lysozyme containing mononuclear cells are numerous (Fig. 3c), but not atypical in appearance. Intraepithelial immunoglobulin-negative lymphocytes are increased in number (Figs. 4b, c). The strongly lysozyme positive Paneth-cells are scarce (Fig. 3c). There is a conspicious epithelial binding pattern for PNL differing obviously from the normal state (Fig. 4a): an exclusive supranuclear coarse and vesicular binding site is detectable throughout the mucosa apparent in only a minority of enterocytes (Fig. 4b, c). This pattern corresonds approximately to the region of the normal crypt-orifice (Fig. 4a, two arrows).

All findings are in agreement with the diagnosis of coeliac disease. There is no evidence of blastic infiltration or of crypt lesions caused by atypical histiocytic aggregates in the biopsy or autopsy material, the latter was rather poorly preserved.

The structure of the neoplastic lymph nodes is incompletely effaced but

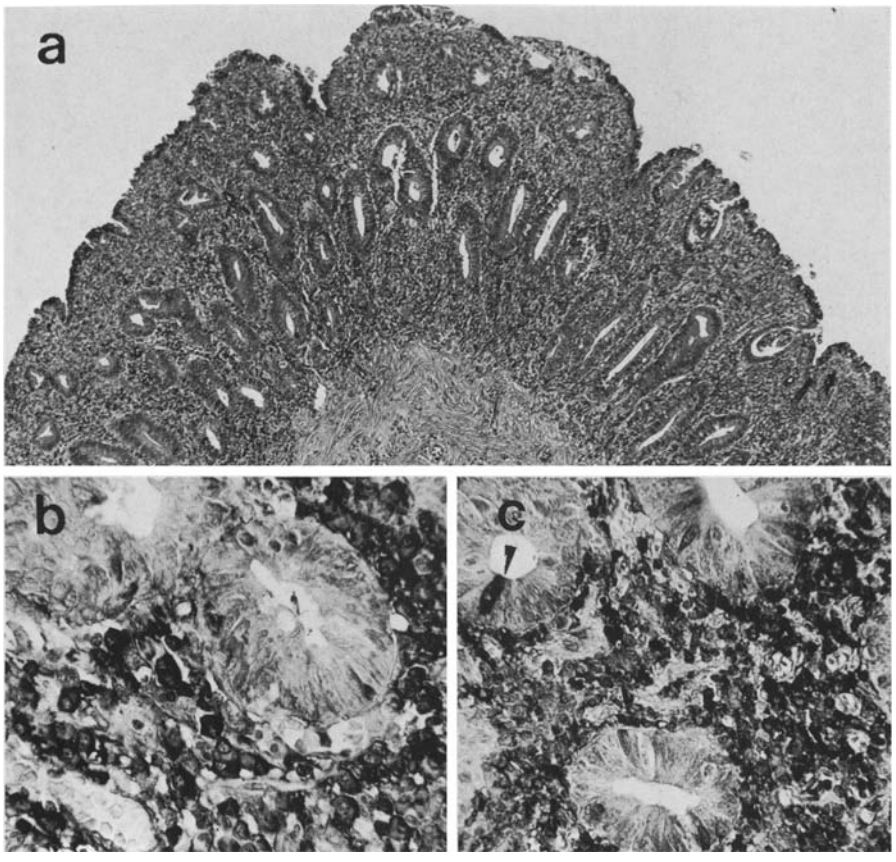

Fig. 3a–c. Jejunal biopsy, taken 8 months prior to death: **a** Villous atrophy, crypt hyperplasia, loss of goblet cells, closely packed inflammatory infiltration of the lamina propria (H-E, × 52). **b** Subepithelial κ-chain producing plasma cells (PaP: anti-κ, × 320). **c** Dense monocytic/macrophagocytic population; a lysozyme-positive Paneth cell (→) (PaP: anti-lysozyme, × 200)

the normal cellular constituents are completely replaced by atypic and pleomorphic cells (Fig. 5). The proliferating cells form bipolar mitoses, have a rounded but non-lymphoid structure, display phagocytic activity and resemble sinus histiocytes (Fig. 5b). Grotesque giant cells are numerous (Fig. 5c). These changes correspond to the malignant histiocytosis of the intestine (MHI) described by Isaacson and Wright (1978a and b). Immunostaining did not give a definitive result because of the post mortem degradation of the tissue.

The bowel ulcers are non-specific. The cutis contains a scanty perivascular lymphocytic infiltration. The spleen displays excessive erythrophagocytosis of sinus histiocytes and marked atrophy of the white pulp. The liver shows centrolobular globular fatty infiltration and an increase in phagocytic activity of sinusoidal cells and Kupffer cells. There is also increased red cell phagocytosis in the bone marrow. All tissues are additionally affected by congestion and disseminated intravascular coagulation.

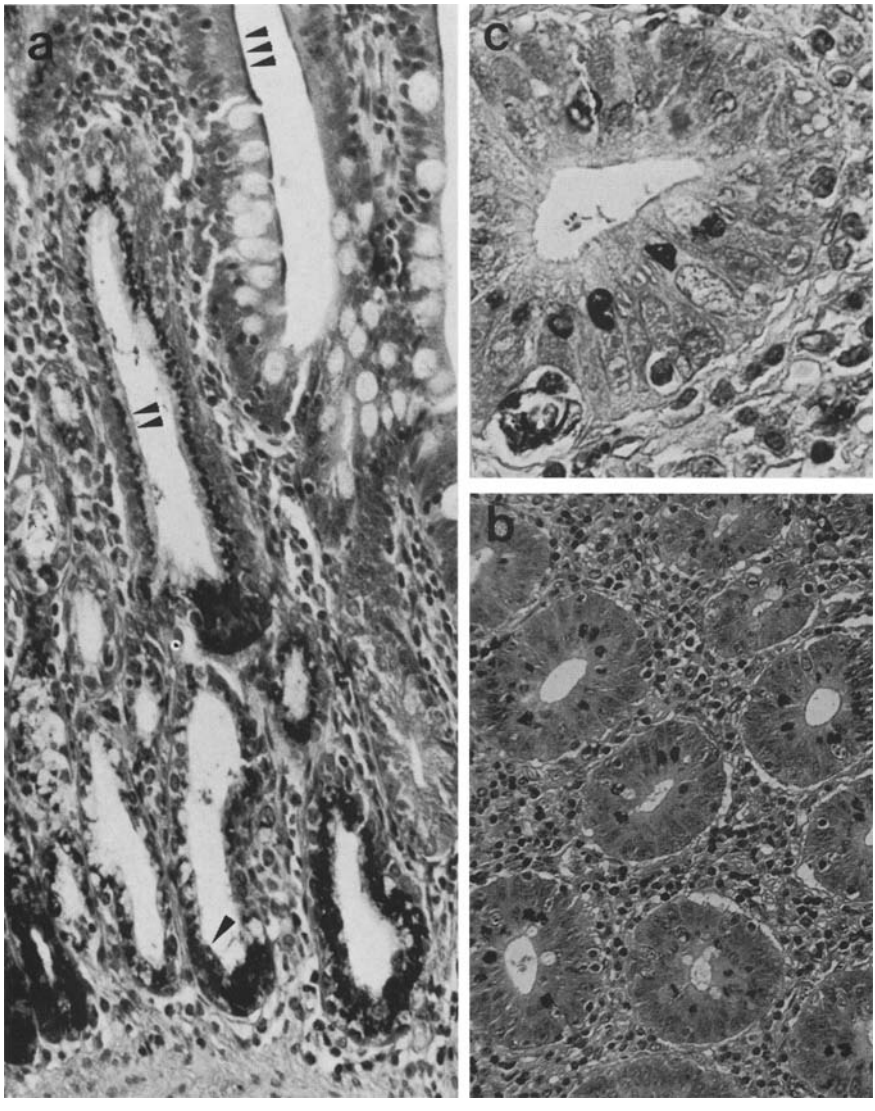

Fig. 4a–c. Peanut lectin in enterocytes. **a** PNL-binding pattern in normal jejunal mucosa (Meckel's diverticulum from a 16 years-old boy): coarse, vesicular, peri- and supranuclear at the crypt base level (↑); granular, distinctly supranuclear at the crypt orifice (↑↑); associated to the luminal surface of villi (↑↑↑) (PaP: PNL, anti-PNL, ×300). **b** Coeliac disease: PNL binding pattern of single enterocytes, approximately corresponding to that of the apical crypt level (PaP: PNL, anti-PNL, ×300). **c** Like **b**, Mitosis and two intraepithelial lymphocytes (PaP: PNL, anti-PNL, ×700)

Discussion

The case presents a chronically progressive enteropathy dating back to infancy, with several rare complications terminally. The gluten sensitivity was diagnosed only months prior to death, due to an aggravation of the

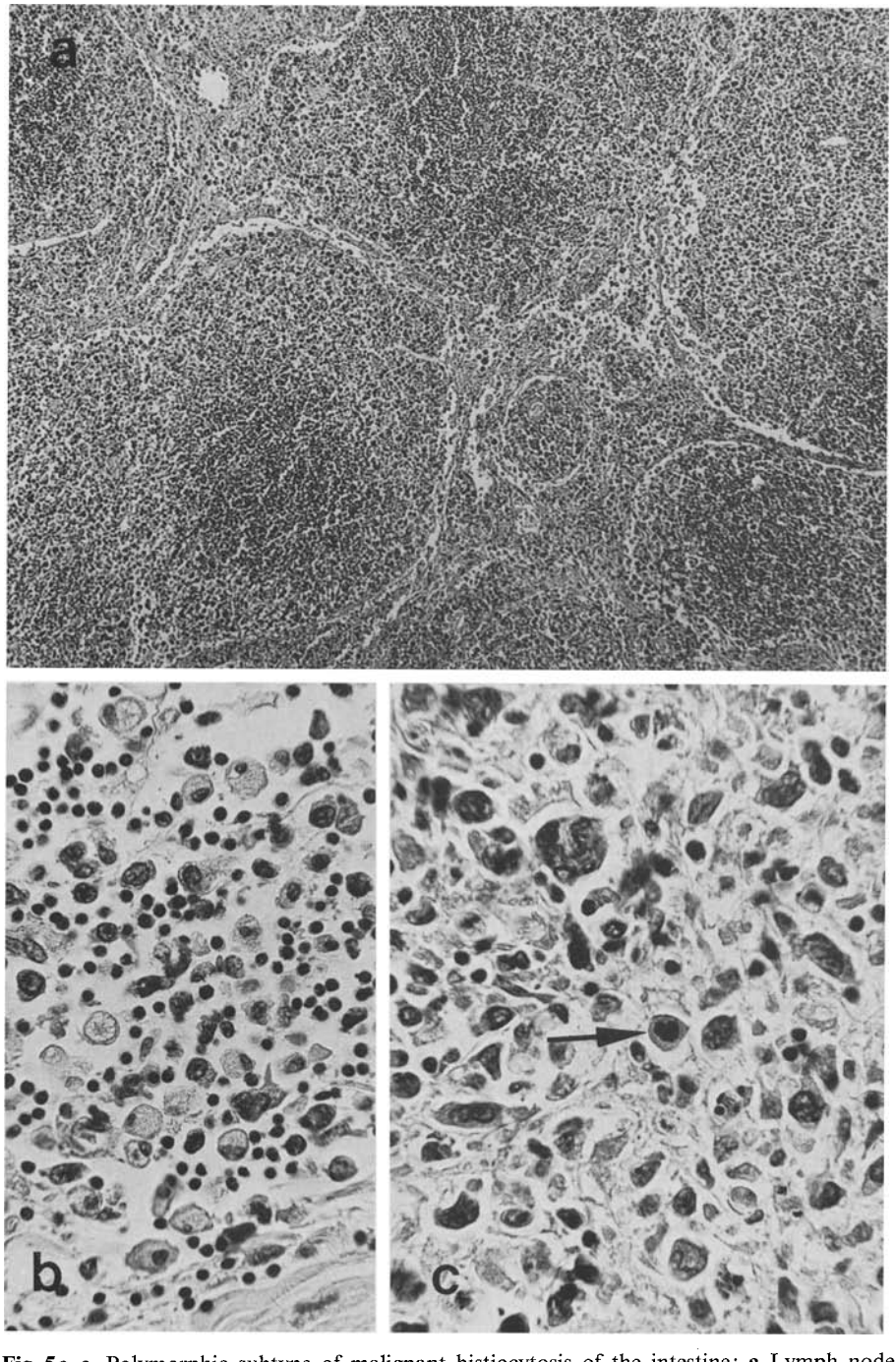

Fig. 5a–c. Polymorphic subtype of malignant histiocytosis of the intestine: **a** Lymph node, survey: partially effaced structure, loss of follicles, sinusoidal and perisinusoidal infiltration (hematoxilin, ×52). **b** Lymph node, detail: tyical and atypical sinus histiocytes with marked phagocytic activity (hematoxilin, ×400). **c** Lymph node, detail of perisinusoidal area: atypical histiocytes, giant cells, mitosis (→) (hematoxilin, ×400)

disease which lead to repeated hospitalizations. Whether the state deteriorated because of the basic disease or because of the development of a malignant tumour of the intestinal lymph nodes, must stay open to question. The terminal decompensation may be attributed to toxinemia due to ulcerative proctosigmoiditis and erythroderma, as repeated blood cultures failed to verify septicemia.

The association of coeliac disease and ulcerative colitis has been reported (Kitis et al. 1980); in spite of its rarety the assumptions of a pathogenic correlation seemed justifiable. Cutaneous complications of gluten-sensitive enteropathy are well known (Otto et al. 1981; Perkkiö et al. 1981; Marsh 1982a). The incidence of primary intestinal malignant lymphomas is raised in coeliac disease (Otto et al. 1981; Isaacson and Wright 1978; Brandt et al. 1978). Thus, all parallel disorders in this young man can be more or less directly related to his gluten-sensitivity.

An entity within the intestinal lymphomas, arising more frequently in coeliac disease and malabsorption syndrome, was recently separated by Isaacson and Wright from the lymphocytogenic tumours. It is a special form of malignant histiocytosis, not identical with the classical histiocytic medullary reticulosis Robb-Smith (Scott and Robb-Smith 1939) and was termed "malignant histiocytosis of the intestine (MHI)" (Isaacson and Wright 1978a and b; Isaacson 1981; Isaacson et al. 1979 and 1982).

The histiocytic character of the tumour can be demonstrated by its phagocytic capacity and by means of immunohistology by its lysozyme and α_1-antitrypsin content. This case differs in two aspects from previously published data: a) Our patient was by far the youngest. The youngest previously reported patient was 43 at diagnosis. The early onset may be due to the life long history of enteropathy. b) The tumour was restricted to the draining lymph nodes of the intestine. Otto et al. (1981) characterize the primary intestinal lymphomas as hyperplasiogenic tumours, arising from a chronic antigenic overload. The origin of a histiocytic neoplasm in the lymph drainage of a gut with coeliac disease suggest an increased influx of toxic substances through a defective enterocyte barrier.

The histological and immunohistological data obtained from the diagnostic jejunal biopsy have their equivalents in recent literature (Falchuk et al. 1974; Cooper et al. 1981; Hodges et al. 1979; Perkkiö et al. 1981; Selby et al. 1981; Simpson et al. 1981; Ward et al. 1979; Marsh 1982b).

Binding sites for peanut lectin, a protein with a high affinity to the disaccharide beta-D-Gal (1>3) Gal NAC (Lothan et al. 1975), are numerous in biological systems (Watanabe et al. 1981; Rose et al. 1981; Reisner et al. 1977; Klein et al. 1981; Möller 1982a, b) but not ubiquitious, at least not in an immunohistologically detectable amount. However a definied technique with a stable sensitivity is able to furnish organotypical and cytotypical binding patterns. This is the first report on PNL-receptors in human jejunal mucosa. As Fig. 4a shows, the enterocytes of a healthy jejunal mucosa (from a Meckel's diverticulum) display a locality-dependent binding pattern. At the crypt base level, there is a course vesicular shaped and peri- or supranuclear located PNL-site (↑). At the crypt orifice it is small, granular and distinctly supranuclear (↑↑). On the villi PNL-binding is confined to

the enterocytic brush order (↑↑↑) while goblet cells and Paneth cells lack the receptor. This pattern corresponds well to that recently found by Sato and Spicer (1982) who furnished ultrastructural data on the PNL-affinity of various alimentary epithelial cells of the mouse: columnar cells of the jejunum disclosed a specific binding in Golgi cisternae and on the tips and sides of the microvilli. (There was, however, a positive staining of goblet cells.). Considering the migration of enterocytes in the course of their maturation (Booth 1970) this phenomenon may be interpreted as evidence of secretion. The PNL-reactive disaccharide might be part of a (protective?!) secretion product. This would then be synthesized within the perinclear endoplasmic reticulum of the immature crypt cell ('enteroblast', Booth 1970), collected in the Golgi field and packed up in vesicles. At the crypt base level those would be exreted and the product would become part of the glycocalyx of the mature cell. In a recently published investigation Pestalozzi et al. (1982) demonstrated a distribution pattern of galactosyltransferase in human enterocytes which seems to be identical with the PNL pattern. As PNL recognizes galactosyl residues, this correspondence may be a hint to the nature of the structure decorated by this lectin.

In the reported case of coeliac disease the secretion process seems to be incomplete and only realized in a minority of enterocytes (Fig. 4b, c).

Evaluation of this single observation in a major collective study is presently underway. If reproducible, the conspicious PNL binding pattern described could possibly serve as a new argument for a primary enterocyte defect in coeliac disease.

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