Microvillus Inclusion Disease and Tufting Enteropathy

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Introduction

The Larger Group of "Intractable Diarrheas of Infancy"

Before focusing on microvillus inclusion disease and tufting enteropathy, we briefly review similarly presenting entities. In 1968 Avery, Villavicencio and Lilly were the first to describe a severe chronic diarrhea in 20 infants and they named it "infantile intractable diarrhea"; according to their description "(it) was prolonged and intractable despite extensive hospital therapy" [1].

This syndrome was defined on the basis of some clinic characteristics, namely: (1) diarrhea of more than 2 weeks duration, (2) age, less than 3 months, (3) three or more stool cultures negative for bacterial pathogens, (4) necessity of intravenous rehydration, and (5) prolonged and intractable diarrhea despite hospital therapy.

The death rate was very high: 9 out of the 20 babies (45 %) in Avery et al.'s record had died; it was even higher in Hyman et al.'s (70 %) record [2].

Heterogeneity and lack of specificity are evident in Avery's original report: different pathologies were grouped in it, some of which with a diagnosis were well defined even at that time. Only autoptic material was available for the first cases, and only after the introduction of total parenteral nutrition (TPN) at the beginning of the 1970s [3] it was possible to study the matter more in depth, thanks to proximal small-intestinal biopsy [4] and later on to the development of endoscopic techniques, which were safe and adequate for the infant as well. It became consequently possible to discriminate different causes for the so-called intractable diarrhea of infancy [5] but its definition superimposes on the definition of "protracted diarrhea of infancy": The latter has duration in common with it, but a failure to gain weight is enough to define the picture [6].

Many cases of "protracted diarrhea of infancy" are diet associated, as a consequence of cow milk or lactose intolerance or malnutrition. Malnutrition causes intestinal atrophy and consequently a malabsorption syndrome and diarrhea, which apparently gets better with fasting. These features have almost disappeared in the developed countries.

The main causes of "intractable diarrhea of infancy," including more severe and longer forms, can thus be summed up (see also Table 1.1):

Autoimmune Enteropathy

This rare disorder mostly occurring in young infants and children (6–18 months old), is characterized by severe diarrhea and small-intestinal mucosal atrophy resulting from immune-mediated injury. It remains a challenging diagnosis because of its clinicopathologic variability. This entity is dealt with in Chap. 2.

Small-Intestinal Enteropathy of Unknown Origin

This entity could be a variation of autoimmune enteropathy, as the increase in inflammatory cells in the lamina propria shows. It appears in less than 12-month-old infants, with a lower death rate compared to those with autoimmune enteropathy, but it can be very severe. Infants can be dependent from TPN [5].

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Table 1.1 Main causes of protracted diarrhea in infancy

Small-intestinal enteropathy of unknown origin
Intractable ulcerating enterocolitis of infancy
Congenital enterocyte heparan sulfate deficiency
Congenital intestinal integrin deficiency
Congenital secretory diarrheas
Congenital chloridorrhea
Congenital Na-losing diarrhea
Autoimmune enteropathy
Diseases of the intestinal epithelium
Microvillus inclusion disease
Tufting enteropathy

Intractable Ulcerating Enterocolitis of Infancy

A rare disease initially described in 1991 in five children presenting in the first year of life with intractable diarrhea, ulcerating stomatitis, and large ulcers with overhanging edges throughout the colon within the first year of life [7]. The affected infants can show such a severe colitis that a subtotal colectomy is necessary, even if long-term prognosis is good. It has been suggested that the affected children have a genetically determined primary immune dysregulation [8].

Congenital Enterocyte Heparan Sulfate Deficiency

Described in 1995 in three infants who, within the first weeks of life, presented secretory diarrhea and massive enteric protein loss [9]. The small-intestinal mucosa is normal on light microscopy, but histochemical examinations show a complete absence of enterocyte heparan sulfate. The sulfated glycosaminoglycans of the basocellular membrane are mostly deficient, particularly heparan sulfate, while distribution of vascular and lamina propria glycosaminoglycans is normal [9]. Diarrhea is so severe to make TPN necessary, associated to repeated albumin infusions because of severe protein-losing enteropathy. Studies in men and mice show that heparan sulfate is essential in maintaining intestinal epithelial barrier function [10], and that the specific loss of heparan sulfate proteoglycans from the basolateral surface of intestinal epithelial cells is common to many forms of protein-losing enteropathy [11].

Congenital Intestinal Integrin Deficiency

In 1999, Lachaux et al. described an intractable diarrhea starting from 9 days after birth, associated to pyloric atresia and total epithelial detachment of gastric and intestinal mucosa. Immunofluorescence analysis showed α6β4 integrin deficiency at the intestinal epithelium—lamina propria junction [12].

Mutations in α6 or β4 integrins cause junctional epidermolysis bullosa with pyloric atresia. In 2008, two Kuwaitian brothers with pyloric atresia were described, respectively affected by intractable diarrhea and episodes of protein-losing enteropathy, with a novel mutation in β4 integrin not associated to its reduced expression in tissues [13].

Congenital Secretory Diarrheas

Includes congenital chloridorrhea and congenital sodium diarrhea, dealt with in Chap. 36.

Diseases of the Intestinal Epithelium

Microvillus inclusion disease and tuft enteropathy are the best-known diseases of the intestinal epithelium causing intractable diarrhea of infancy.

In 1994, Girault et al. described eight infants with early-onset severe watery diarrhea associated to facial deformities and unusual tufts of woolly hair with trichorrhexis nodosa. Duodenal biopsies showed moderate to severe villous atrophy, with normal or hypoplastic crypts; colon biopsies were grossly normal. As a consequence, severe malabsorption was present. All patients had no antibody response to immunization antigens; the immunological response to vaccinations was poor. Five children died despite TPN [14]. Two children from the series of Girault et al. had hepatic cirrhosis; six additional patients had signs and symptoms compatible with this new "syndromic diarrhea", associated to hepatic involvement (Tricho-Hepato-Enteric Syndrome, THES) characterized by fibrotic livers with marked hemosiderosis [15–17].

Nine different mutations in *TTC37* gene (5q14.3–5q21.2) were found in 12 children from 11 families with classical features of THES. *TTC37* codes for a protein that has been named thespin (THES ProteIN) [18].

Enlarged platelets with abnormal α-granule secretion can be observed in some patients. The estimated incidence of the syndrome is 1 in 400,000 to 1 in 500,000 live births.

Microvillus Inclusion Disease

In 1978, Davidson et al. described five infants presenting an intractable diarrhea of infancy characterized by secretive diarrhea and malabsorption, starting in the first hours after birth with hypoplastic villous atrophy in the small-intestinal biopsy. Four of these infants had a deceased brother who had presented similar features.

In one of these infants, the electron microscopy identified the presence of a peculiar abnormality of the microvilli of the enterocytes [19] (Fig. 1.1).

Three new cases with the same clinic and histological characteristics of the latter were described in France in 1982,

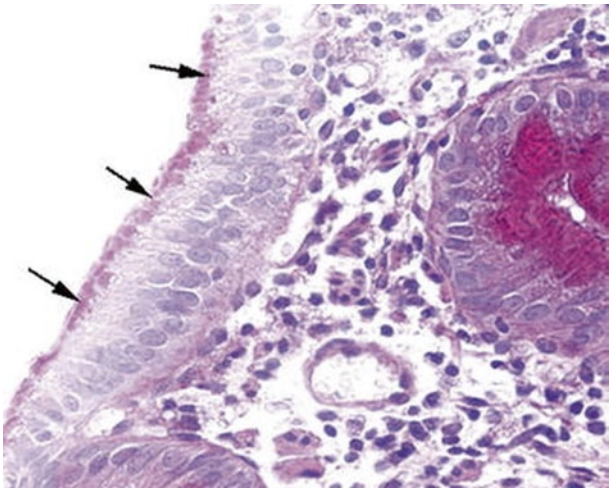

Fig. 1.1 Microvillous in the original label inclusion disease. PAS staining highlights abundant PAS-positive material *(arrows)* in the apical part of the enterocyte cytoplasm. PAS × 260. *PAS* peroidic acid-Schiff (Reprinted from Ref. [20], Fig. 1, with kind permission from Springer Science and Business Media)

and the four of them were grouped in a new disease called congenital microvillus atrophy [21, 22]. Two new cases were described in Great Britain in 1985 [23] and one in Italy in 1986; a subsequently born brother of the latter resulted affected too [24]. A survey completed in 1987 among centers known for their involvement in pediatric gastroenterology identified more than 30 cases worldwide. Additional cases were later published.

In 1989, Cutz et al. proposed the use of the term "microvillus inclusion disease" to highlight the characteristic ultrastructural lesions of the disease [25].

Clinical presentation: case report

First child of parents with no blood relation, A.G. was born after 37 weeks of gestation, the pregnancy having been complicated by a risk of miscarriage in the 5th month. His weight was 3500 g.

The infant was hospitalized when he was 40 days old because of an abundant diarrhea (15–20 evacuations a day of liquid stools), starting on the 6th day of its life and resistant to numerous dietary and pharmacological therapies.

Entering the hospital, the patient weighed 2800 g, it was in severe general conditions with dystrophia and dehydration; a TPN was therefore immediately started. The acid–basic balance showed hyponatremic acidosis (pH 7.2; EB −8,3; Na 128 mEq/l). The secretive nature of diarrhea was confirmed by its entity (about 100 ml/kg/die) with a total absence of oral nutrition and with the persistence of TPN in progress.

Moreover, the typical absence of ionic gap in the stools was present: osmolality 226 mOsm/l, Na 86 mEq/l, K 23.5 mEq/l (gap 7 mOsm/l).

Loperamide and chlorpromazine increased intestinal absorption, but did not change the clinical picture.

Microbiological examinations included an electronic microscope examination of the feces for the identification of viruses and the search for enterotoxigenic bacteria and parasites with specific methods were repeatedly negative.

The abdominal ultrasound showed adrenal hyperplasia associated to hyperaldosteronism (1160 ng/ml, v.n. < 125 ng/ml).

Jejunal biopsy showed a picture of villus atrophy with no hyperplastic crypts and periodic acid-Schiff (PAS)-positive material stored in the apical cytoplasm of enterocytes. Electron microscopy was diagnostic for microvillus inclusion disease.

Microvillus Inclusion Disease is a Congenital Secretory Diarrhea Starting in Neonatal Age

Severe diarrhea typically appears in the first days of life, usually within the first 72 h, and it is immediately life threatening. The stools are watery, and the stool output is 100–500 ml/kg/day when the infant is fed, a volume comparable to or higher than that observed in cholera. The diarrhea is of secretory type; therefore, it persists at a stable rate of 50–300 ml/kg/day despite fasting, and the electrolyte content of the stools is increased, without an osmotic gap. However, the mucosal atrophy causes osmotic diarrhea. For this reason, feeding increases the fecal output and oral feeding in nutritionally significant amounts is impossible. Due to the high output, patients can lose up to 30% of their body weight within 24 h, resulting in profound metabolic acidosis and severe dehydration, unless vigorous intravenous rehydration is started.

In a small percentage of cases (which was in the past considered to be around 20%, and presently around 5% of the cases [26]), diarrhea starts later in life, between 1 and 3 months, usually at 6–8 weeks of age. This less severe form has been denominated late-onset microvillus inclusion disease, while the classical form beginning at neonatal age has been denominated early-onset microvillus inclusion disease[27].

The hallmark of the disease is the electron microscopic finding of disrupted enterocytic microvilli (i.e., digitations of the apical membrane of the intestinal epithelial cell protruding into the lumen) and the appearance of characteristic inclusion vacuoles, whose inner surfaces are lined by typical microvilli. Both lesions are seen only with the electronic microscopy.

A few cases have been termed atypical microvillus inclusion disease, in which the onset can be early or late, but the histologic picture is different, particularly for the absence of detectable microvillus inclusions [28].

Therefore, three variants of the disease have been identified: early-onset microvillus inclusion disease, late-onset microvillus inclusion disease, and atypical microvillus inclusion disease. However, because of the sparse distribution of microvillus inclusions, it is not certain that their absence could be limited to the sample.

Microvillus inclusion disease is usually characterized by growth retardation and some developmental delay later in infancy. No other specific findings can be detected. However, the disease can be associated with other abnormalities, indicated in Table 1.2.

Histologic Findings

Findings from duodenal biopsy must not be considered diagnostic. Histologic results of duodenal biopsy samples can range from essentially normal to mildly abnormal, showing the following:

- Thin mucosa caused by hypoplastic villus atrophy
- Diffuse villus atrophy (loss of villus height)
- Crypt hypoplasia

PAS staining of the intestinal biopsy sample does not show the usual linear staining along the brush border, but reveals PAS-positive material in the apical cytoplasm. The PAS staining material corresponds to the increased number of electron dense secretory granules in the epithelium. The abnormal pattern of staining appears in the upper crypt region and continues over the villus [29] (Fig. 1.2).

PAS accumulates in low crypts in atypical microvillus atrophy, in upper crypts in congenital microvillus atrophy, and in low villi in late-onset microvillus atrophy.

Table 1.2 Anomalies described in association to microvillus inclusion disease

Meckel diverticula	Abdominal adhesions
Inguinal hernias	Renal dysplasia
Absent corpus callosum	Hydronephrosis
Mesenteric duct remnants	Craniosynostosis
Abnormal vertebrae	Down syndrome
Aganglionic megacolon	Hematuria
Pneumocystis jiroveci pneumonia	Dihydropyrimidinase deficiency
Autosomal dominant hypochondroplasia	Microcephaly
Renal Fanconi syndrome	Other renal problems
Hypophosphatemic rickets	Diabetes
Cardiac problems	Pulmonary problems
Liver dysfunction	Multiple hepatic adenomas

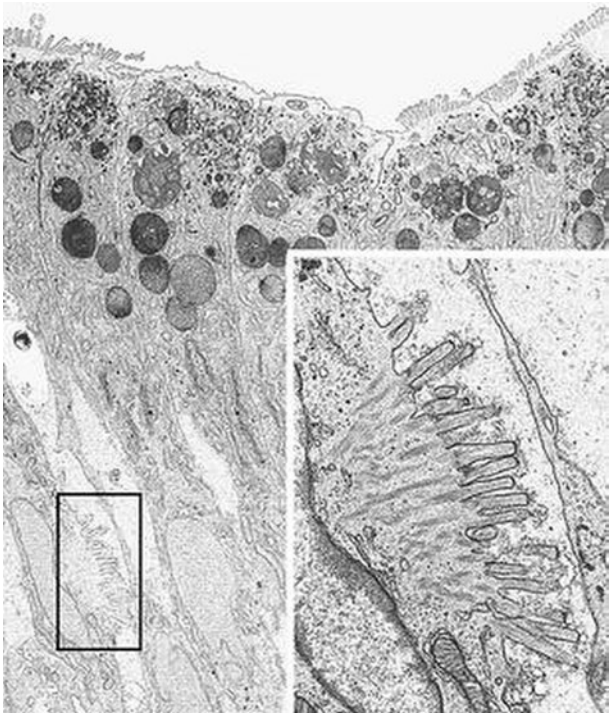

Fig. 1.2 Microvillous in the original label inclusion disease. Villous enterocytes: the *boxed area* shows microvilli on the lateral membrane. *Inset*: Enlargement of the *boxed* area × 6200, *inset* × 22,500. (Reprinted from [20, Fig. 5], with kind permission from Springer Science and Business Media)

Similar results were obtained with anti-CD10 immunohistochemistry: In affected children, the normal linear staining in surface enterocytes is absent, while prominent cytoplasmic reactivity is seen [30]. CD10 is a neutral membrane-associated peptidase; thus, abnormal stain findings with PAS or anti-CD10 immunohistochemistry are expressions of the abnormalities in microvillar structure.

Rectal biopsy findings demonstrate microvillus involutions and an increased number of secretory granules. This test has been proposed as a relatively easy method for making an early diagnosis. Anti-CD10 immunohistochemistry can aid in the diagnosis, because abnormal cytoplasmic CD10 staining of absorptive colonocytes has been observed in microvillus inclusion disease [31].

The diagnosis rests on findings demonstrated by electron microscopy (see Figs. 1.3 and 1.4.). Electron microscopy shows well-preserved crypt epithelium with abundant microvilli. Villus enterocytes are severely abnormal, particularly toward the apices of the short villi. The microvilli are depleted in number, short, and irregularly arranged. Some of the enterocytes contain the typical microvillus involutions, which are intracellular vacuoles where microvilli are observed lining the inner surface. A striking feature is a number of small, membrane-bound vesicles containing electron-dense material (see Figs. 1.3 and 1.4). A few cases have been

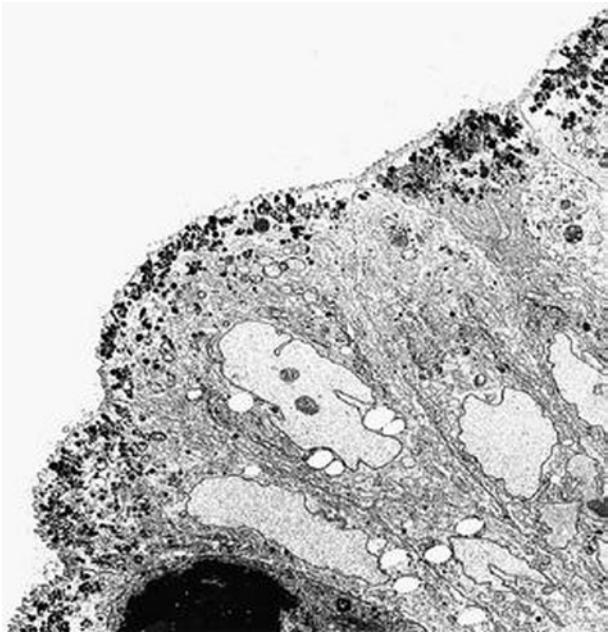

Fig. 1.3 Microvillous in the original label inclusion disease. The apical cytoplasm of villous epithelium shows an increased number of secretory granules associated with microvillus alterations × 2400. (Reprinted from [20, Fig. 4], with kind permission from Springer Science and Business Media)

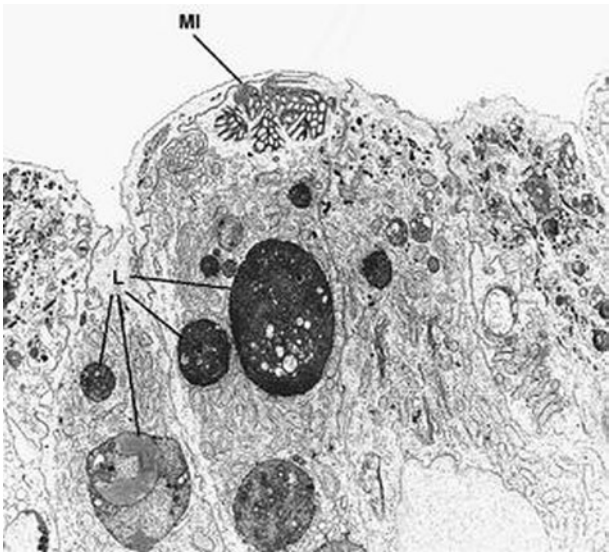

Fig. 1.4 Microvillous in the original label in the original label inclusion disease. The villous enterocytes lack brush-border microvilli, whereas their apical cytoplasm contains a microvillus inclusion *(MI)* and numerous lysosomes *(L)* × 5.500. (Reprinted from [20, Fig. 2], with kind permission from Springer Science and Business Media)

described in which the classic microvillus inclusions are shadowed by other features, such as large aggregates of electron lucent, vermiform membranous vesicles in enterocyte cytoplasm, corresponding to the PAS-positive material [32].

Epidemiology

The cases published or gathered in an online registry were 137 in 2014 [26].

A female preponderance had been observed among the published cases, with a female-to-male ratio of 2:1, but in the total 137 cases there is a 1.54 male to female ratio. A blood relation is present in 41 % of the assessable cases with a genre preference for males. A cluster of cases from the Navajo reservation in northern Arizona suggests an incidence as high as 1 case per 12,000 live births.

Pathophysiology

Due to their alterations, mature enterocytes inefficiently absorb ions and nutrients, causing a malabsorption syndrome; however, the diarrhea is caused mainly by active secretion of water and electrolytes in the intestinal lumen (secretory diarrhea). The pathogenesis of the secretory diarrhea is unknown; it is assumed to result from an imbalance between decreased absorption and unaltered secretion.

Measurement of stool electrolytes and osmolality enables rapid and accurate assessment of the pathogenesis of this chronic diarrhea (osmolar versus secretory) and greatly narrows the differential diagnosis.

Fecal electrolytes demonstrate a typical pattern of secretory diarrhea. Fecal sodium levels are high (approximately 60–120 mEq/l), and no osmotic gap is found. In patients with secretory diarrhea, the following formula applies: 2(Na concentration + K concentration) = stool osmolarity ± 50. In osmotic diarrhea, stool osmolarity exceeds 2(Na concentration + K concentration) by 100 or more.

Secretory diarrhea occurs in the fasting state and is associated with large output losses that cause dehydration and metabolic acidosis.

In osmotic diarrhea, findings on stool microscopy are negative for white blood cells (WBCs), blood (exudative diarrhea), and fat (steatorrhea).

Even if there are data about the anomalies in water and electrolytes transportation in the small intestine, it is not known whether and how the colon mucosa participates to the absorption alterations in the disease.

In one of the Italian cases, we used the technique of rectal perfusion that showed a decrease in sodium absorption, only partially corrected by chlorpromazine administration [33].

Pathogenesis

Severe perturbation of the microvillar cytoskeleton may disrupt the transport of brush border components that have

to be assembled at the apical membrane. The postulated abnormality in the cytoskeleton causes a block in exocytosis, mainly of PAS-positive material (e.g., polysaccharides, glycoproteins, glycolipids, neutral mucopolysaccharides). As a consequence, small secretory granules that contain a PAS-positive material accumulate in the apical cytoplasm of epithelial cells.

In 2008, the presence of mutations in the *MYO5B* gene was described in seven patients (out of ten tested), predominantly of Turkish origin [34]. Homozygous mutations in the same gene were subsequently found in seven cases of Navajo origin; five parents were heterozygote [35]. A total of 41 unique *MYO5B* mutations in 40 patients have been identified so far: in more detail 16 different homozygous mutations in 25 patients, and 25 heterozygous mutations in 15 patients [26].

The *MYO5B* gene codifies myosine Vb, an actin-based motor protein which carries the recycling endosomes to the apical plasma membrane along the actin filaments of the microtubules. The functional deficiency of this protein alters the intracellular trafficking of resident apical plasma membrane proteins to the cell surface, and this could be the cause for the impaired apical brush border membrane development [34]. Actually, in microvillus inclusion disease the *MYO5B* mutations associate to a defective myosin Vb expression in enterocytes.

Myosine Vb carries on its action after having bound to a specific small guanosine-5′-triphosphatase (GTPase) rab proteins, such as Rab 11, located on the surface of recycling endosomes [36]. Thanks to this link the recycling endosomes move along the actine filaments (see Fig. 1.5) [37].

When myosine Vb has an altered function, the recycling endosomes are not carried in a normal way: in the enterocytes of the subjects with microvillus inclusion disease, no regular accumulation of myosine Vb and of the recycling endosome-associated proteins (one of these is Rab 11) can be observed close to the apical membrane, and no specific staining pattern is present [38]. Therefore, the Rab 11 distribution in the enterocytes can be a helpful diagnostic tool [39].

Other biochemical mechanisms depending on myosine Vb which can produce alterations in the structure of the microvilli are presently being studied [40].

Myosine Vb is expressed in all the epithelial tissues and, actually, microvillus inclusions in the stomach and colon, in addition to less well-defined inclusions in gallbladder epithelium and in renal tubular epithelial cells, have been reported in some patients with microvillous inclusion disease (MVID). Nevertheless, no extraintestinal symptoms are generally reported. Two children with renal Fanconi syndrome who carried mutation *MYO5B* did not show alterations in the apical brush border morphology and the PAS staining pattern in renal tubular epithelial cells, which makes it unlikely for it to be the cause of proximal tubular renal dysfunction [41].

Recently, Dutch investigators have found [42] that the mild variant of MVID appears to be caused by loss of function of syntaxin 3 (STX3), an apical receptor involved in membrane fusion of apical vesicles in enterocytes. In fact, whole-exome sequencing of DNA from patients with variant MVID revealed homozygous truncating mutations in STX3; and in addition, patient-derived organoid cultures and overexpression of truncated STX3 in CaCo2 cells recapitulated most characteristics of variant MVID.

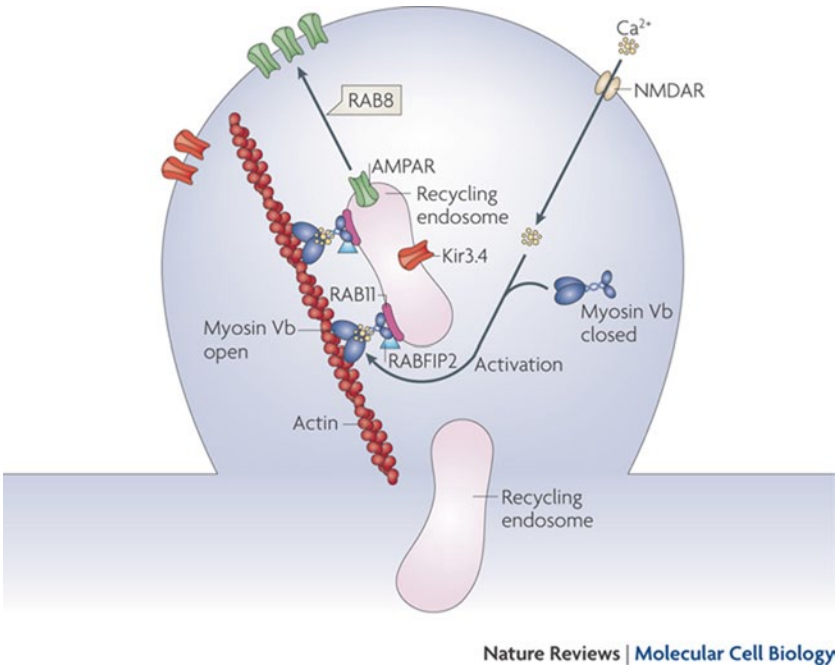

Fig. 1.5 Endocytic recycling. Myosin Vb is a conformation-dependent binding partner of Rab11-FIP2. Activation of myosin Vb induces translocation of recycling endosomes and their cargo. Final transport from the recycling endosome to the cell surface is mediated by Rab8. (Reprinted by permission from Macmillan Publishers Ltd and Nature Publishing Group [37])

Prenatal Diagnosis

Pregnancy and birth are usually normal in individuals with microvillus atrophy, and polyhydramnios is usually absent, in contrast to the clinical picture of patients with other causes of congenital secretory diarrhea [43]. Nevertheless, in some cases, polyhydramnios and bowel dilation in the third trimester have been described [44]. In one case, a high fetal alpha-fetoprotein in the second trimester was observed [45]. Authors have speculated that the fetal alpha-fetoprotein elevation might possibly be caused by in utero body fluid leakage into the amniotic fluid through fetal enteropathy.

Identification of the gene responsible for the disease allows its prenatal diagnosis [46].

Treatment

The prognosis of early-onset microvillus inclusion disease is poor. If patients are untreated, the disease is rapidly fatal because of dehydration and malnutrition.

In late-onset microvillus inclusion disease diarrhea tends to be less severe, and some alimentation is possible.

Medical Care

Agents tentatively given to induce a better growth of the intestinal mucosa (e.g., epithelial growth factor, colostrum) are ineffective. Several drugs (e.g., somatostatin, octreotide, loperamide, chlorpromazine) have been tried to counteract the massive secretory diarrhea in patients with microvillus atrophy; however, none has proven effective.

At present, the only available therapy is TPN. Children with late-onset microvillus inclusion disease usually have less severe diarrhea; with age they can reduce the requirements of TPN to 1–2 per week.

If patients are treated with TPN, their prognosis entirely depends on the complications of this approach. These complications include cholestasis with subsequent liver damage leading to cirrhosis, catheter-related sepsis due to infection with bacterial or fungal agents, and progressive lack of vascular access.

In the observed cases, cholestasis appears worsened by transplant.

The study of eight patients who developed cholestatic liver disease suggests that cholestasis is enhanced by the impairment of the *MYO5B*/*RAB11A* apical recycling endosome pathway in hepatocytes [47].

Surgical Care

Successful outcomes of small-intestinal transplantation have been reported, and evidence suggests that an early transplant might be beneficial. The limited experience accumulated in a few centers worldwide reflects an overall survival rate of approximately 50 % at 5 years after small-bowel transplantation; this is a much better outcome than is seen with other indications for intestinal transplantation [48]. Patients who did not receive colonic transplant weaned later from parenteral nutrition.

The analysis of 16 patients who underwent a small-bowel transplantation shows a lower death rate compared to those who did not (23 versus 37 %) after an average observation period of 3.5 years (but variable between 3 months and 14 years). In all of the cases, apart from the first two, colon had been transplanted too [49].

Although only small series have been reported, evidence suggests that early small-bowel transplantation should be performed, at least in children with early-onset microvillus inclusion disease. Patients with late-onset microvillus atrophy appear to have an improved prognosis.

Transplantation appears to be the only option for patients who do not fare well with long-term TPN (e.g., because of sepsis, liver damage, lack of vascular access). For patients in whom transplantation is successful, a gradual return to a normal diet is considered possible.

In the observed cases, TPN-related cholestasis appears worsened by transplant. Therefore, in children with cholestasis, the worsening of this picture after the transplant points to a combined liver-intestinal transplantation.

Tufting Enteropathy (or Intestinal Epithelial Dysplasia)

In 1994, Reifen et al. described two infants less than a month old with protracted diarrhea. The diarrhea was so profuse to make TPN necessary but it improved when enteral nutrition was interrupted. The jejunal biopsies showed a peculiar picture characterized by the presence of focal aggregations of packed enterocytes whose shape resembled of a teardrop, as a consequence of an apical rounding of the plasma membrane. These focal areas resembled a tuft and that is why the term “tufting enteropathy” was coined [50]. Curiously, a case with the same characteristics was identified among those presented by Davidson et al. in the same paper where the first case of microvillus inclusion disease had been described [19].

Clinical Expression

The incidence of the disease has been estimated to be 1:100,000 live births in Western Europe [51], but it seems higher in people of Arabic origin [52].

The picture is a severe secretory diarrhea starting in the first weeks of life. During pregnancy, there is no polyhydramnios, as in the microvillus inclusion disease and

differently from congenital sodium diarrhea and congenital chloridorrhea.

The alterations in the enterocytes, in any case, cause an accentuation of the diarrhea with nutrition, including total enteral nutrition, as it had already been observed since the first described cases.

There are two different clinical forms: one is isolated and the other is syndromic, associated to different anomalies, particularly to facial dysmorphism with choanal atresia and superficial punctuated keratitis [53, 54].

Pathophysiology

In 2008, a mutation of the gene for *Epithelial Cell Adhesion Molecule* (EpCAM) was identified in two ill children in the same family, and in three children from unrelated kindreds [55]. EpCAM is a transmembrane protein involved in cell proliferation, differentiation, and adhesion.

In 2010, a mutation in *SPINT2* gene was found in a case affected by a syndromic form of tufted enteropathy. *SPINT2* is a transmembrane protein that seems to be involved in epithelial regeneration [56].

It is interesting to note how mutations in *SPINT2* gene are also present in the syndromic congenital sodium diarrhea, where choanal atresia, hypertelorism, and corneal erosions are particularly frequent and anal atresia can be found in certain cases [57].

Analyzing 57 patients, mutations in the gene for EpCAM were found in 73% of the cases, all of them presenting an isolated intestinal disease.

But in 21% of the cases, all showing a syndromic form of the disease, there are mutations the *SPINT2* gene.

According to this study, tufting enteropathy could be separated into at least three genetic classes, each with specific phenotypes [58].

However, it seems impossible at present to discriminate "tufting enteropathy" isolated from the syndromic one, even from a genetic point of view.

Histologic Features

Jejunal biopsy shows a picture of partial villous atrophy associated to crypt hyperplasia. The most characteristic feature, the one which gave the name to the disease, is the presence of "tufts," small focal aggregates of teardrop-shaped enterocytes with an apical rounding (See Fig. 1.6a, b).

The "tufts" are not a characteristic exclusive to intestinal epithelial dysplasia, because they have been observed in other mucosal enteropathies and in normal jejunum. In the latter cases, anyway they were present in <10% of the epithelial surface, while in "tufting enteropathy" they are present in more than 80% of the jejunal surface. But the picture is not always so evident in the earliest period of the disease. Attempts at immunohistochemical analysis (including beta-catenin, E-cadherin, desmoglein, laminins) have not been easily applicable [60]. On the contrary, the staining with EpCAM/MOC31 antibody, an EpCAM antibody clone, showed a sensitivity and specificity of 100% for loss of staining in 15 studied patients [61].

Electronic microscopy shows relatively normal microvilli, and it is not particularly useful for diagnosis, if only to exclude a microvillus inclusion disease.

A mild inflammation of the lamina propria is also present. An infiltration of T lymphocytes within the lamina propria had been observed since the original description, even if inferior to celiac disease, but it sometimes arises suspicion of autoimmune enteropathy [50].

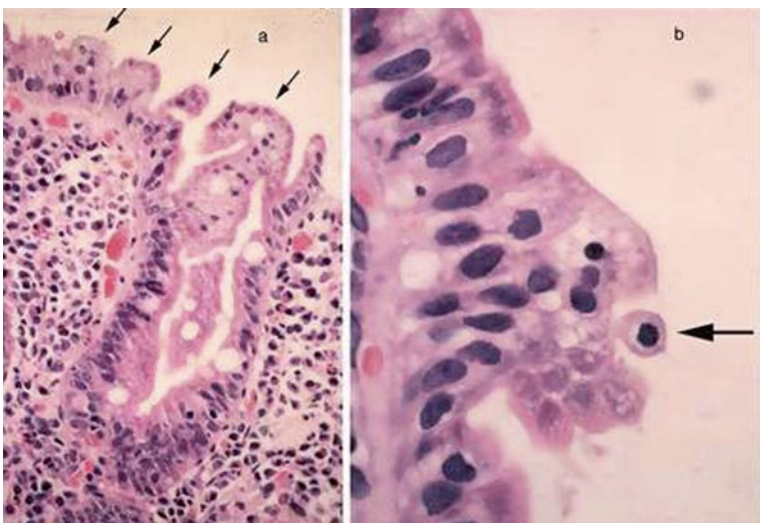

Fig. 1.6 **a** Numerous tufts of enterocytes on the mucosal surface of the duodenum. **b** A characteristic tear-drop-shaped structure *(arrow)* in an epithelial tuft (H&E stain; original magnification: a–x 80; b–x 400). (Reprinted from Ref. [59, Fig. 1], with kind permission from Springer Science and Business Media)

Treatment

Tuft enteropathy is associated to a severe secretory diarrhea, which worsens with nutrition. That is why affected children have to be treated with TPN.

Some cases seem to have a less severe course and they can be given a partial parenteral nutrition [62].

Cases totally dependent on TPN are candidates for intestinal transplantation.

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