PATHOLOGICA 2022;114:22-31;
DOI: 10.32074/1591-951X-338

Review

Histopathology of intestinal villi in neonatal and paediatric age: main features with clinical correlation - Part II

Chiara Rossi[1], Gloria Simoncelli[2], Giovanni Arpa[1], Alessandra Stracuzzi[3], Paola Parente[4], Matteo Fassan[5,6], Alessandro Vanoli[1], Vincenzo Villanacci[2]

[1] Unit of Anatomic Pathology, Department of Molecular Medicine, Fondazione IRCCS Policlinico San Matteo, University of Pavia, Pavia, Italy; [2] Institute of Pathology, Spedali Civili di Brescia, Brescia, Italy; [3] Pathological Anatomy Unit, Department of Diagnostic and Laboratory Medicine, IRCCS Bambino Gesù Children's Hospital, Rome, Italy; [4] Pathology Unit, Fondazione IRCCS Casa Sollievo della Sofferenza, San Giovanni Rotondo, Italy; [5] Department of Medicine (DIMED), Surgical Pathology Unit, University of Padua, Padua, Italy; [6] Veneto Institute of Oncology, IOV-IRCCS, Padua, Italy

Summary

In this paper, we will continue the description of histological findings of infantile and paediatric small bowel alterations with the main clinical pictures and differential diagnosis. We emphasise once again the need to evaluate the biopsies in an adequate clinical contest and with a systematic approach, including epithelial alterations, lamina propria changes, mucosal architecture, and the distribution of inflammation, together with other morphological signs more specific of certain diseases. We describe the histological findings of coeliac and Crohn's disease, gastrointestinal food allergic diseases, Langerhans cell histiocytosis, nutritional deficiencies and infections. Finally, we suggest the principal issues in the drafting the pathological report for appropriate interpretation and usefulness in clinical practice.

Key words: coeliac disease, Crohn's disease, gastrointestinal food allergic diseases, langerhans cell histiocytosis, infections, paediatric enteropathies, small bowel

Received and accepted: July 30, 2021
Pubblished online: December 2, 2021

Correspondence
Paola Parente
Pathology Unit, Fondazione IRCCS Ospedale Casa Sollievo della Sofferenza, viale Cappuccini 1, 71013 San Giovanni Rotondo (FG), Italy
Tel.: +39 0882 410365
Fax: +39 0882 410411
E-mail: paolaparente77@gmail.com

Conflict of interest
The Authors declare no conflict of interest.

How to cite this article: Rossi C, Simoncelli G, Arpa G, et al. Histopathology of intestinal villi in neonatal and paediatric age: main features with clinical correlation - Part II. Pathologica 2022;114:22-31. https://doi.org/10.32074/1591-951X-338

Coeliac disease

Coeliac disease (CD) is an immune-mediated systemic disorder elicited by gluten, a protein found in wheat, barley, rye, spelt, and kamut. It occurs in genetically predisposed individuals, mostly females (male/female ratio 1:2) [1]. The age at presentation vary from early childhood to elderly, according to the age at introduction of gluten in the diet, the quantity consumed and individual sensitivity [1,2]. In Western countries, the prevalence of histologically confirmed CD is around 0.6% [3]. In the last decades, due to sensitive and specific screening tests, there has been a significant increase in the number of new cases of CD [4]. Diagnostic workup is carried through a thorough evaluation of clinical, serological, genetic and histological aspects. In order to achieve a correct diagnosis, it is essential to examine the patients while they are still exposed to gluten. In fact, a gluten-free diet may alter the clinical, serological and histological features of CD, making it unrecognisable [5].

Clinical presentation

According to Caio et al., CD presentation can be subdivided in two main phenotypes: intestinal and extra-intestinal, which may occur individually or in combination [4]. In paediatric patients, the classic presentation

consists in loss of appetite, abdominal distention, diarrhoea and failure to thrive. Growth retardation and delayed puberty may be major manifestations in adolescents [1,4]. Extra-intestinal manifestation are common in both children and adults and are mainly related to malabsorption or chronic inflammation. They include iron deficiency-related microcytic anaemia (most frequent) or macrocytic anaemia due to folic acid and/or vitamin B12 deficiency. Most patients also manifests osteopenia or osteoporosis due to altered absorption of calcium and vitamin D. Neurological manifestation may include headache, paraesthesia, depression, cerebellar ataxia and gluten encephalopathy. Other symptoms include aphthous stomatitis, dental enamel hypoplasia, and hepatitis. CD can be associated with different immune disorders including dermatitis herpetiformis, type 1 diabetes, alopecia, Hashimoto's thyroiditis and selective IgA deficiency [1,4,6].

Histological diagnosis and classification

Histological evaluation still remains of critical importance in CD diagnosis. Should undergo an intestinal biopsy [2]:

- Individuals with positive serology, characterised by the presence of IgA class antitransglutaminase (tTGA) and antiendomysial antibodies (EMA), and children younger than 2 years with isolated IgA antigliadin (AGA) positivity.
- Individuals with deficiency of IgA, positive for tTGA IgG (and even children aged < 2 years with positivity for AGA IgG with or without tTGA IgG).
- Individuals in whom coeliac disease is strongly suspected, with a severe malabsorption syndrome, irrespective of antibody test results.

To obtain a valid diagnosis, at the time of the biopsy, the patient must be on a normal diet containing gluten. Biopsy should be performed in the duodenal bulb and in the distal duodenal portion. It is recommended to collect at least 4 biopsies, 2 for each of the areas mentioned above [3]. Particularly in children, the evaluation of the duodenal bulb can be essential in CD diagnosis. Multiple studies have shown that CD modifications can be restricted to the duodenal bulb in 2.5% to 13% of patients, especially in children [7-9]. In a multicentre study of 102 paediatric patients performed by Bonamico et al, it has been demonstrated that involvement of the duodenal bulb was present in all subjects, in 25% of whom it was the only site of injury [10]. Furthermore, a study published by De Leo and Villanacci shows that bulb duodenal analysis led to a 12% increase in CD diagnosis, emphasising the critical role of bulb duodenal biopsies in CD [11] (Fig. 1). Positioning of biopsies on cellulose acetate filters is recommended. This method ensures proper orientation of the histological sample [2].

The distinctive histological features of CD are:

1 *Increased intraepithelial T lymphocytes*: a value > 25 T lymphocytes/100 enterocytes (lymphocytosis).
2 *Crypt hyperplasia*: extension of the regenerative epithelial crypts associated with the presence of > 1 mitosis per crypt.
3 *Villous atrophy*: decrease in villous height, alteration of normal crypt/villous ratio (3:1) until total

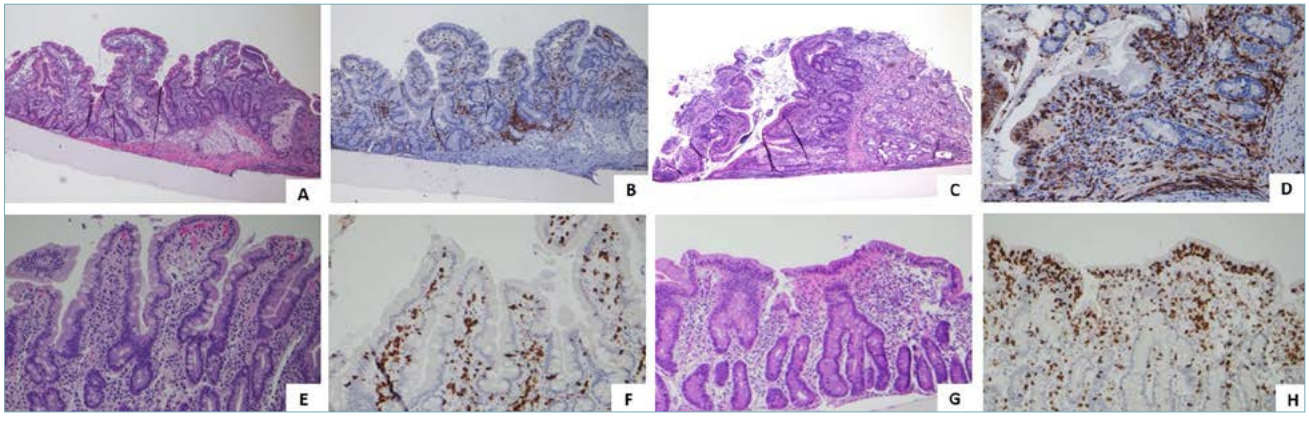

Figure 1. The same case: A-B normal villi in distal duodenum; T lymphocytes < 25/100 epithelial cells. A H&E 4x, B CD3 immunostain 4x; C-D moderate-severe villous atrophy in bulb; T lymphocytes > 25/100 epithelial cells (Type 3B + 3C/Grade A + B2) with pathological increase of T lymphocytes. C H&E 4x, D CD3 immunostain 4x. The same case: E-F normal villi in distal duodenum; T lymphocytes < 25/100 epithelial cells. E H&E 10x, F CD3 immunostain 10x; G-H severe villous atrophy in bulb; T lymphocytes > 25/ 100 epithelial cells (Type 3C/Grade B2) with pathological increase of T lymphocytes. G H&E 10x, H CD3 immunostain 10x.

disappearance of villi. This assessment requires proper orientation of the biopsies.

Up to now, several classifications exists to describe the histopathological alteration of CD, and they are briefly reported in Table I [2,12-15].

Unfortunately none of the above mentioned characteristics is pathognomonic for CD; for an accurate diagnosis the histological pattern has to be integrated with clinical, serological and genetic data[5].

In the histological report, atrophic lesions (Type 3a, 3b or 3c/Grade B) should be indicated as "consistent with CD", while for non-atrophic lesions with intraepithelial lymphocytosis (Type 1 or 2/Grade A) the term "suggestive for CD" is more appropriate. In the conclusive diagnosis the pathologist should only give a description of the lesion, stressing the idea that these injuries are not exclusive of CD and should therefore necessarily be placed in the right clinical setting and supported by serological and genetic confirmation [2].

Immunohistochemistry

The application of immunohistochemistry in CD diagnosis can be helpful to clearly identify the lymphocytosis condition. Albeit the increase of intraepithelial T lymphocyte can be observed also on haematoxylin-eosin stain, we suggest, especially in the easly forms, the use of CD3 monoclonal antibodies for a more accurate evaluation of the epithelial lyphocytosis [2,5]. CD8 monoclonal antibodies can be useful in elderly patients, in order to exclude a refractory form of CD, not responsive to a gluten-free diet. In this condition, regarded by many as pre-lymphomatous, the expression of CD8 may be reduced [2].

Differential diagnosis

CD shares its duodenal histopathologic features with a large variety of intestinal disorders. Numerous conditions are associated with increased intraepithelial lymphocytes, with or without villous blunting [5,16]. Therefore clinical and serological correlation is mandatory. Regarding the paediatric population, the main conditions that must be ruled out are Helicobacter pylori gastritis, parasitic infections and gastrointestinal food-protein enteropathies, common variable immunodeficiency, and autoimmune enteropathy [17]; these conditions are discussed in part I of this review [18], in following sections of this work, and other papers in this same Special Issue [19].

In regards to collagenous sprue, small bowel biopsies may show an increase in chronic inflammatory cells in the lamina propria in association with increased intraepithelial lymphocytes and patchy epithelial degenerative changes; the detection of a subepithelial band-like collagen deposit in the proximal small bowel may lead the pathologist to a correct diagnosis [1,20].

Table I. Coeliac disease classification systems.

Marsh Classification		
Type 1 – Infiltrative lesion Villi within normal morphological limits (normal villous/crypt ratio 3:1) Increased number of intraepithelial lymphocytes (greater than 25/100 epithelial cells)	**Type 2 – Hyperplastic lesion** Villi architecturally within normal morphological limits (like type 1) Increased number of intraepithelial lymphocytes (greater than 25/100 epithelial cells) (like type 1) Hyperplasia of the glandular elements (regenerative aspects highlighted by the reduced mucinous activity and increased number of mitoses).	**Type 3 – Destructive lesion** Varying degrees of villous atrophy associated with hyperplasia of glandular crypts Surface enterocytes with reduced height, irregular brush border and sometimes cytoplasmic vacuoles Increased number of intraepithelial lymphocytes (like type 1 and 2 lesions).
Oberhuber modifications		
3a: mild villous atrophy and pathological increase of intraepithelial lymphocytes. **3b**: moderate villous atrophy and pathological increase of intraepithelial lymphocytes **3c:** total villous atrophy and pathological increase of intraepithelial lymphocytes		
Corazza-Villanacci Classification		
Grade A lesions Normal villi but with a pathological increase in intraepithelial lymphocytes	**Grade B lesions** **B1:** villus/crypt ratio is less than 3:1 and pathological increase of T lymphocytes **B2:** villi are no longer identifiable and pathological increase of intraepithelial lymphocytes	
Congenital defects of small intestine epithelial differentiation		
Grade A – non atrophic type No architectural changes (villous/crypt ratio preserved) and increased IELs count (> 25/100 epithelial cells)	**Grade B – atrophic type** Villous atrophy (mild-moderate-severe degree), crypt hyperplasia (mitoses > 1/crypt) and increased IELs count (> 25/100 epithelial cells).	

Crohn's Disease in childhood

Crohn's disease (CrD) is, together with ulcerative colitis, an inflammatory bowel disease (IBD), a chronic, multifactorial, immune-mediated disorder with a relapsing and remitting course, which may cause, to a variable degree, inflammation of the entire digestive tract [21,22].

Incidence and prevalence of CrD greatly varies by geographic region, having the highest prevalence in Europe and North America. Nevertheless, since 1990, CrD incidence is accelerating in the newly industrialized countries, such as Asia, South America and Africa, while being stable or even decreasing in North America and Europe [23].

Up to 30% of CrD cases develops during childhood or adolescence [24], with an annual incidence ranging from 2 to 5 per 100,000 children [25]. According to patient age at IBD diagnosis, it is possible to distinguish 2 main groups in IBDs: very-early-onset IBDs (VEO-IBDs), diagnosed before 6 years of age, and early-onset IBDs, diagnosed between 6 and 16 years of age [21]. For a more detailed overview of VEO-IBD, we invite the reader to refer to another paper in this Special Issue by Parente et al. [26].

IBD pathophysiology is still not completely known; nonetheless, genetic, environmental, immunological and microbiome-related factors have been proved to contribute, at various rates, to its development [22].

Paediatric CrD is demonstrated to have a more severe behaviour compared to its adult form [27,28], partly because some of VEO-IBDs are represented by a monogenic form (i.e., XIAP deficiency, interleukin 10 signalling defect, IPEX-like), which typically has an aggressive phenotype and less frequently responds to traditional treatments [22].

Common symptoms and signs of paediatric CrD include abdominal pain, diarrhoea, weight loss, growth failure, anorexia, malaise, fatigue, anaemia and fever [22]. Beside clinical evaluation, imaging of the small bowel, esophagogastroduodenoscopy, ileo-colonoscopy and multiple biopsies from the gastrointestinal tract are recommended to achieve a correct diagnosis [25,29].

Upper gastrointestinal involvement appears to be very frequent in paediatric CrD, having an incidence rate of 30-70% [30,31] and resulting to be significantly more frequent in patients with an extensive ileocolonic inflammation [32]. The endoscopic finding usually observed in small bowel include erythema, oedema, erosions, aphthous ulcers, mucosal granularity, pseudopolyps, "cobblestone" appearance of mucosa and stenosis [32,33]. Small intestine histological abnormalities may include patchy or diffuse, chronic or mixed, inflammation in lamina propria, erosions, lymphoid aggregates, cryptitis, partial villous atrophy and non-caseating, epithelioid or giant cells, granulomata [32-34]. Although no single endoscopic finding is pathognomonic, some of them (i.e., aphthous ulcers in any part of the gastrointestinal tract, mucosal skip lesions, ulcerations or strictures of the terminal ileum, significant perianal disease), particularly if coupled with biopsies showing typical IBD-associated lesions (mostly if they include granulomata), may make straightforward the diagnosis of CrD and help in the differential diagnosis with the much rarer form of ulcerative colitis involving the upper gastrointestinal tract [22,25,29,32,34].

Gastrointestinal food allergic diseases

Gastrointestinal food allergic diseases are classified in 3 groups, with regard to their underlying pathogenesis: i) IgE-mediated, which occur very rapidly and frequently have a systemic, severe involvement, ii) mixed IgE/non-IgE-mediated, such as the previously discussed EGIDs (chapter 10), iii) non-IgE-mediated, in which circulating food-specific IgE are typically absent and onset of gastrointestinal symptoms is generally delayed after food allergens exposure [35].

Non-IgE-mediated gastrointestinal food allergic diseases (non-IgE-GI-FA) may be further subclassified into 3 main disorders, according to how extensively the digestive tract is involved: a) food protein-induced proctocolitis, limited to colorectum and characterised by mild symptoms and by a high relative frequency among non-IgE-mediated food allergic disorders, b) food protein-induced enteropathy (FPE), affecting the small intestine and characterised by a moderate severity of symptoms, c) food protein-induced allergic enterocolitis syndrome (FPIES), the least common non-IgE-GI-FA, characterised by severe symptoms and involving whole gastrointestinal tract [35].

FPE is relatively uncommon, accounting for about a fifth of the number of the coeliac patients, and it seems to be decreasing over time [36]. FPIES is a rare disease, with a cumulative incidence ranging from 0.34 to 0.7% in infancy [37,38]. Co-atopy is highly represented in non-IgE-GI-FA, affecting up to 44% [39] and 55% [40] of FPE and FPIES patients, respectively.

Non-IgE-GI-FA pathophysiology is not completely clarified; a combined implication of both cellular immunity [41] and innate immune system [42], in the absence of a notable humoral immunity [43], has been suggested.

Clinics and oral food challenge (OFC) are sufficient for FPIES diagnosis, while histologic confirmation is mandatory for FPE diagnosis [35]. The commoner culprit foods involved in are cow's milk, soy, rice, poultry, fish, wheat, eggs, fruits, corn and vegetables [35].

Common histological findings in jejunal biopsies of FPE and FPIES patients include mild to severe villous atrophy [44], crypt hyperplasia and, occasionally, an increased number of eosinophils and intraepithelial lymphocytes [36]. These alterations are independent to the offending food [44]. Ultrastructurally, epithelial cells display abnormally located nuclei and short and furry microvilli containing large aggregates of lysozymes [36].

Langerhans cell histiocytosis

Langerhans cell histiocytosis (LCH) is, albeit its rarity, one of the most common histiocytic disorder, characterised by a clonal proliferation, and consequent accumulation, of immature Langerhans cells within different organs [45-47]. With regard to its localisation, LCH may be classified into three main groups: single-system single-site, single-system multi-site and multisystem disease, with or without risk organ involvement [47]. Estimated annual incidence for LCH is 2-9 cases/1 million people, with a peak incidence between 1-4 years of age [48,49], and male to female ratio of 1.5:1 [50]. LCH with gastrointestinal involvement is quite rare, usually affects patients under 2 years of age, with a 2-fold male predominance, and is associated with a multisystemic presentation and a poor prognosis [51].
Gastrointestinal involvement of LCH may have various presentations, comprising vomiting, failure to thrive, haematochezia, intractable diarrhoea, malabsorption, constipation, abdominal pain, protein-losing enteropathy, intestinal perforation and strictures. In these patients, esophagogastroduodenoscopy usually shows erosions or ulceration of duodenal mucosa [51].
Molecular studies have showed that up to 75% of LCHs harbour mutually exclusive mutations of BRAF (V600E), MAP2K1 and N/KRAS genes [45,52], all of which determinate the activation of the mitogen-activated protein kinase/extracellular signal-regulated kinase (MAPK/ERK) cascade.
Histology remains the gold standard for LCH diagnosis. Biopsy samples display, in lamina propria, a proliferation of medium-sized mononuclear cells, characterised by ovoid nuclei with a longitudinal nuclear groove (the so-called “coffee-bean” appearance) and a moderate amount of eosinophilic cytoplasm, admixed with a variable number of eosinophils, macrophages, stromal cells, multinucleated giant cells and T cells [53]. In fact, LCH cells have been proved to produce a large variety of proinflammatory cytokines and chemokines, inducing the migration of several inflammatory cells in the involved site [47,53]. Diagnosis requires confirmation of the histiocytic tumour’s nature by immunohistochemistry. Langerhans cells express CD1a, CD207/langerin, S100 and may also result positive for CD68, vimentin and p53 [46]. Electron microscopy typically shows pathognomonic cytoplasmic tennis racquet-shaped organelles, the Birbeck granules, which have been demonstrated to strictly correlate with the immunohistochemical expression of CD207/langerin [54].

Nutritional deficiencies (zinc and iron deficiencies and kwashiorkor)

Nutritional deficiency is defined as a low or insufficient intake of micronutrients (i.e. vitamins and dietary minerals) and macronutrients (i.e. protein, carbohydrates and fat), due to dietary or inherited causes. During childhood, many nutritional deficiencies may lead to retardation in growth and development.
Nowadays, malnutrition affects predominantly children < 5 years of age living in low-income and middle-income countries [55].
Zinc is a micronutrient essential for growth, and the immune,nervous and endocrine systems, particularly during infancy [56]. Since preterm infants are in negative zinc balance at birth due to a lower capacity for gut absorption, they have an increased risk of zinc deficiency [57].
Two rare, inherited disorders, caused by mutations in zinc transporters genes, may determinate zinc deficiency in children: i) *acrodermatitis enteropathica*, an autosomal recessive disease, characterised by cutaneous and gastrointestinal manifestations, due to a reduction in intestinal zinc absorption [56,58], ii) *transient neonatal zinc deficiency*, causative for low concentration of zinc in mother’s breast milk [56].
Microscopically, duodenal biopsies of patients with *acrodermatitis enteropathica* show loss of villous architecture, with flattened villi, increased inflammatory infiltrate in lamina propria and reactive changes in epithelial cells, such as cuboidal cellular shape, enlarged nuclei and open chromatin distribution [58]. Furthermore, ultrastructural examination of intestinal epithelial cells has revealed the presence of numerous intracellular vescicles [59]. After treatment, cellular and villous modifications have been proved to fully recover [58].
Iron deficiency, also causative for anaemia, is another rare cause for the alteration of small intestine villous pattern in paediatrics. It has been estimated that, during infancy, only 1/7,000 children is affected[60]. Reported duodenal-jejunal changes include blunting, shortening or fusion of villi, coupled with dense infiltration of inflammatory cells in lamina propria. After iron therapy, small intestine histology generally improves or returns to normal [61].
Kwashiorkor is a form of severe malnutrition, caused by low protein intake.

Histological studies, mostly performed on malnourished children from underdeveloped and developing countries, have depicted variable lesions in small intestine mucosa, ranging from mild shortening to flattening and thickening of villi, according to the different quality of the examined individuals' diet [62]. Ultrastructural studies on jejunal mucosa have shown an accumulation of fat in the endoplasmic reticulum, in the Golgi apparatus and in the cytoplasm of epithelial cells, as well as in the intercellular space and in lamina propria. These findings probably result from a derangement of fat metabolism [63]. Furthermore, a shortening of microvilli, rounded mitochondria and an increase in polyribosomes have also been observed [64]. All these histological and ultrastructural changes may be reversed after an adequate refeeding [62].

Infections

A complete discussion of all the infections that can affect the small intestine in paediatric age is beyond the scope of this review; it is still of note, however, to mention some relevant infective diseases that can act as mimickers of organic disorders affecting the villous epithelium of the duodenum.

Helicobacter pylori (HP) gastric infection is rather common in the paediatric age, and can cause a peptic duodenitis, with increased IELs and normal or minimally atrophic villous architecture [65]. Reactive hyperplasia of Brunner glands and gastric metaplasia may be present, and the latter has been shown to harbour HP in a small percentage of cases. Nevertheless, the identification of HP on gastric epithelium is required for a diagnosis of HP-related peptic duodenitis.

Other infectious disease that can cause villous atrophy with increased intraepithelial lymphocytes and must therefore be considered in the differential diagnosis in a paediatric patient are viral and parasitic infections; in the latter category, *Giardia lamblia* infection should be ruled out before diagnosing an organic disorder of the villi [66]. Cryptosporidiosis, albeit less frequent, is another known parasitic infection mimicking villous atrophy in the duodenum [67,68]. As for *G. lamblia*, identification of the offending parasite on the brush border of the affected villi in a child should prompt careful examination of the laboratory exams and history so as to avoid overdiagnosis of an organic condition.

A final note of caution must be issued when examining biopsies of the small intestine in which a prominent macrophage infiltrate is recognized; careful examination and use of special stains can help differentiate storage disorders from the more common infections by intracellular microorganisms: *Tropheryma whipplei* (PAS-D-positive, Acid-fast-negative) and *Mycobacterium avium* complex (Acid-fast-positive, PAS-D-negative) are the most common conditions that must be excluded [69].

Pathological report

The wide variety of conditions and indications that may lead to the performance of a small bowel biopsy in the paediatric age mandates that the pathologist maintains a certain degree of flexibility in writing the report. Table II and Table III report respectively a sum-

Table II. Main histologic features of paediatric small bowel mucosal disorders.

Disorder	Villous atrophy	Intraepithelial lymphocytosis	Lamina propria inflammation	Characteristic histologic features
Congenital enzymatic and transport deficiencies				
Congenital disaccharidase deficiencies	absent	absent	absent	normal small bowel histology
Congenital lipid trafficking deficiencies	absent	absent	absent	fat-filled, multivacuolated enterocytes; lipid droplets in the intercellular/extracellular spaces (only in Anderson disease)
Ion and nutrient transport deficiencies	rare and usually mild	absent	possible	dense inspissated mucus in cystic fibrosis
Congenital defects of small intestine epithelial differentiation				
Microvillous inclusion disease	present, usually severe	absent	present	absence of brush border; PAS+ (or CD10+) inclusions on the apex of enterocytes
Congenital tufting enteropathy	present, usually severe	absent	mild	"tufts" of teardrop-shaped surface enterocytes; crypt dilatation and hyperplasia; reduced EpCAM expression

continues

Table II. Main histologic features of paediatric small bowel mucosal disorders (*follows*).

Enteroendocrine cell dysgenesis	possible, usually mild	absent	absent	absence of chromogranin-positive neuroendocrine cells
Trico-hepato-enteric syndrome	present	absent	absent	none
Autoimmune disorders				
Coeliac disease	present	present	present	crypt hyperplasia, regenerative changes
Autoimmune enteropathy	present, usually severe	absent	present	absence or severe reduction in goblet and Paneth cells
Immunodeficiencies				
Common variable immune deficiency	present	present	present	absence of plasma cells, nodular lymphoid hyperplasia
Selective IgA deficiency	present	present	present	absence of plasma cells, nodular lymphoid hyperplasia
Chronic granulomatous disease	absent	absent	absent	granulomas with pigment-laden macrophages in the crypt, with possible extension to the villus if particularly florid
Graft-versus-host disease	present, usually mild	absent	mild (if present)	epithelial apoptotic bodies, gland destruction and loss of Paneth cells
Infections and bacterial overgrowth syndrome	rare and patchy	rare	possible	none
Gastrointestinal food allergic diseases	present, mild to moderate	possible	absent	histological alterations in biopsy from for protein-induced enteropathy (FPE) and for protein-induced allergic enterocolitis syndrome (FPIES)
Eosinophilic gastroenteritis	absent	absent	present	dense lamina propria infiltration by eosinophils; crypt abscesses or cryptitis may be present
Crohn's disease	possible, usually mild	absent	present	granulomata, erosions, lymphoid aggregates, cryptitis
Ulcerative colitis-associated duodenitis/backwash ileitis	absent	absent	present	chronic active inflammation and crypt distortion
Lymphangectasia	possible	absent	absent	dilated lymphatic vessels in the superficial or deep mucosa
Storage diseases				
Congenital disorders of glycosylation	present	absent	present	small bowel manifestations are usually present only with deficit of phosphomannose isomerase (MPI-CGD, fomer CGD type Ib) and mutations in the gene encoding alpha-1,3-glucosyltransferase (ALG6-CDG, formerly CDG type Ic)
Mucopolysaccharidosis	absent	absent	absent	scarce; reported enlarged lymphatic vessels in the lamina propria in Sanfilippo syndrome
Lysosomal acid lipase deficiency (LAL-D)	possible	absent	present	PAS+ foamy macrophages in the lamina propria in Wolman disease and CESD
Tangier disease	possible	absent	present	PAS-negative foamy macrophages in the lamina propria
Glycogen storage diseases	possible, usually mild	absent	present	mild alterations possibly found in GSD type Ib as a Crohn-like enteritis
Glycolipid storage disorders	absent	absent	absent	enlarged ganglion cells with foamy cytoplasm in Fabry disease
Nutritional deficiencies (zinc, iron, kwashiorkor)	possible, usually mild	usually absent	possible	cellular and villous alterations regress after supplementation of the lacking nutrients
Langherans Cell Histiocytosis	absent	absent	present	medium-sized mononuclear cells, characterised by ovoid nuclei with a longitudinal nuclear groove and a moderate amount of eosinophilic cytoplasm (CD1a+, langerin +, S100+)
Necrotising Enterocolitis	absent	absent	absent	ischaemic colitis with patchy or diffuse haemorrhagic necrosis of the mucosa, coagulative necrosis of the muscular layers

Table III. Most common histologic mimics of small bowel pathologic conditions and their differential diagnosis.

Diagnosis	Histology	Differential diagnosis
Melanosis and pseudomelanosis	Dark, pigmented macrophages in the lamina propria	Lysosomal storage diseases, chronic granulomatous disease
Barium granuloma	Granulomatous reaction with fine, refractile material	Lysosomal storage diseases, IBD, chronic granulomatous disease
Pseudolipomatosis	Clear spaces in the submucosa, without epithelial or endothelial lining	Lymphangectasias
Foreign body injury	Nonspecific necrosis, inflammation; may progress to ulcer or perforation	Ischaemic enteritis
Drug-related injury	Nonspecific enteritis	IBD

mary of the most common histological modifications of the pathologic conditions reported in this review and some common non-pathological mimics of disease [70]. An extensive and thorough description of the findings should be provided in the pathological report, including: i) a note on the orientation of the sample, especially if it is suboptimal, fragmented or there are crushing artefacts that could impair the correct evaluation of the structure of the villous, ii) degree of villous atrophy, iii) presence, distribution and composition of the inflammatory infiltrate. iv) any additional relevant histologic sign, v) changes in the submucosal or muscular layers, if present in the biopsy sample, vi) presence of microorganisms. When the diagnosis is straightforward and guidelines exist to grade and report a specific condition, the latter should be suitably applied and reported in the final pathological report; however, in many cases, the final diagnosis can only be reached by integrating the histological picture with the clinical and laboratory information.

Conclusions

Diagnosis of small bowel disorders and their specific aetiology requires an integrated clinical and pathological approach. In-depth knowledge of histopathological lesions and the main differential diagnosis are imperative in daily pathological practice for appropriate management of these patients.

References

1 Russo P, Ruchelli ED. Pathology of pediatric gastrointestinal and liver disease. New York: Springer-Verlag, Inc. 2004 https://doi.org/10.1007/978-1-4419-9066-2

2 Villanacci V, Ceppa P, Tavani E, et al. Coeliac disease: the histology report. Dig Liver Dis Off J Ital Soc Gastroenterol Ital Assoc Study Liver 2011;43 Suppl 4:S385-95. https://doi.org/10.1016/S1590-8658(11)60594-X

3 Al-toma A, Volta U, Auricchio R, et al. European Society for the Study of Coeliac Disease (ESsCD) guideline for coeliac disease and other gluten-related disorders. Published online 2019. https://doi.org/10.1177/2050640619844125

4 Caio G, Volta U, Sapone A, et al. Celiac disease: a comprehensive current review. BMC Med 2019;17:142. https://doi.org/10.1186/s12916-019-1380-z

5 Villanacci V, Vanoli A, Leoncini G, et al. Celiac disease: histology-differential diagnosis-complications. A practical approach. Pathologica 2020;112:186-196. https://doi.org/10.32074/1591-951X-157

6 Casella G, Bordo BM, Schalling R, et al. Neurological disorders and celiac disease. Minerva Gastroenterol Dietol 2016;62:197-206.

7 Boschee E, Lacson A, Turner J, et al. Duodenal bulb histology in paediatric celiac disease: a case-control study. J Can Assoc Gastroenterol 2020;3:210-215. https://doi.org/10.1093/jcag/gwz014

8 Gonzalez S, Gupta A, Cheng J, et al. Prospective study of the role of duodenal bulb biopsies in the diagnosis of celiac disease. Gastrointest Endosc 2010;72:758-765. https://doi.org/10.1016/j.gie.2010.06.026

9 Evans KE, Aziz I, Cross SS, et al. A prospective study of duodenal bulb biopsy in newly diagnosed and established adult celiac disease. Am J Gastroenterol 2011;106:1742-1837. https://doi.org/10.1038/ajg.2011.171

10 Bonamico M, Thanasi E, Mariani P, et al. Duodenal bulb biopsies in celiac disease: a multicenter study. J Pediatr Gastroenterol Nutr 2008;47:618-622. https://doi.org/10.1097/mpg.0b013e3181677d6e

11 De Leo L, Villanacci V, Ziberna F, et al. Immunohistologic analysis of the duodenal bulb: a new method for celiac disease diagnosis in children. Gastrointest Endosc 2018;88:521-526. https://doi.org/10.1016/j.gie.2018.05.014

12 Oberhuber G, Granditsch G, Vogelsang H. The histopathology of coeliac disease: time for a standardized report scheme for pathologists. Eur J Gastroenterol Hepatol 1999;11:1185. https://doi.org/10.1097/00042737-199910000-00019

13 Corazza GR. Coeliac disease. J Clin Pathol 2005;58:573-574. https://doi.org/10.1136/jcp.2004.023978

14 Corazza GR, Villanacci V, Zambelli C, et al. Comparison of the interobserver reproducibility with different histologic criteria used in celiac disease. Clin Gastroenterol Hepatol 2007;5:838-843. https://doi.org/10.1016/j.cgh.2007.03.019

[15] Villanacci V. The histological classification of biopsy in celiac disease: time for a change? Dig Liver Dis 2015;47:2-3. https://doi.org/10.1016/j.dld.2014.09.022

[16] Robert ME, Crowe SE, Burgart L, et al. Statement on Best Practices in the Use of Pathology as a Diagnostic Tool for Celiac Disease A Guide for Clinicians and Pathologists. Am J Surg Pathol 2018;42e44-e58. https://doi.org/ 10.1097/PAS.0000000000001107

[17] Francalanci P, Cafferata B, Alaggio R, et al Pediatric autoimmune disorders with gastrointestinal expression: from bench to bedside. Pathologica 2021 Epub December 2. https://doi.org/10.32074/1591-951X-339

[18] Rossi C, Simoncelli G, Arpa G, et al. Histopathology of intestinal villi in neonatal and paediatric age: main features with clinical correlation - Part I. Pathologica 2021 Epub December 2. https://doi.org/10.32074/1591-951X-337

[19] Campora M, Mastracci L, Carlin L, et al. Pathologist's approach to paediatric and neonatal eosinophilic gastrointestinal disorders. Pathologica 2021 In Press.

[20] Freeman HJ. Collagenous sprue. Can J Gastroenterol 2011;25:189-192. https://doi.org/10.1155/2011/821976

[21] Bequet E, Sarter H, Fumery M, et al. Incidence and phenotype at diagnosis of very-early-onset compared with later-onset paediatric inflammatory bowel disease: a population-based study [1988-2011]. J Crohns Colitis Published online October 31, 2016:jjw194. https://doi.org/10.1093/ecco-jcc/jjw194

[22] Oliveira SB, Monteiro IM. Diagnosis and management of inflammatory bowel disease in children. BMJ Published online May 31, 2017:j2083. https://doi.org/10.1136/bmj.j2083

[23] Ng SC, Shi HY, Hamidi N, et al. Worldwide incidence and prevalence of inflammatory bowel disease in the 21st century: a systematic review of population-based studies. Lancet 2017;390(10114):2769-2778. https://doi.org/10.1016/S0140-6736(17)32448-0

[24] Abraham BP, Mehta S, El-Serag HB. Natural history of pediatric-onset inflammatory bowel disease: a systematic review. J Clin Gastroenterol 2012;46:581-589. https://doi.org/10.1097/MCG.0b013e318247c32f

[25] IBD Working Group of the European Society for Paediatric Gastroenterology, Hepatology and Nutrition. Inflammatory bowel disease in children and adolescents: recommendations for diagnosis-the porto criteria. J Pediatr Gastroenterol Nutr 2005;41:1-7. https://doi.org/10.1097/01.MPG.0000163736.30261.82

[26] Parente P, Pastore M, Grillo F. Very Early Onset-IBD: evidence for the need of a multidisciplinary approach. Pathologica 2021 Epub December 2. https://doi.org/10.32074/1591-951X-336

[27] Puntis J, McNeish AS, Allan RN. Long term prognosis of Crohn's disease with onset in childhood and adolescence. Gut 1984;25:329-336. https://doi.org/10.1136/gut.25.4.329

[28] Pigneur B, Seksik P, Viola S, et al. Natural history of Crohn's disease: Comparison between childhood- and adult-onset disease. Inflamm Bowel Dis 2010;16:953-961. https://doi.org/10.1002/ibd.21152

[29] PA. NAS for PG hepatology, and nutrition. Differentiating ulcerative colitis from Crohn disease in children and young adults: report of a working group of the North American Society for Pediatric Gastroenterology, Hepatology, and Nutrition and the Crohn's and Colitis Foundation of America. J Pediatr Gastroenterol Nutr 2007;44:653-674. https://doi.org/10.1097/MPG.0b013e31805563f3

[30] Lenaerts C, Roy CC, Vaillancourt M, et al. High incidence of upper gastrointestinal tract involvement in children with Crohn disease. Pediatrics 1989;83:777-781.

[31] Cameron DJS. Upper and lower gastrointestinal endoscopy in children and adolescents with Crohn's disease: a prospective study. J Gastroenterol Hepatol 1991;6:355-358. https://doi.org/10.1111/j.1440-1746.1991.tb00870.x

[32] Castellaneta SP, Afzal NA, Greenberg M, et al. Diagnostic role of upper gastrointestinal endoscopy in pediatric inflammatory bowel disease: J Pediatr Gastroenterol Nutr 2004;39:257-261. https://doi.org/10.1097/00005176-200409000-00006

[33] Schmidt-Sommerfeld E, Kirschner BS, Stephens JK. Endoscopic and histologic findings in the upper gastrointestinal tract of children with Crohn's Disease: J Pediatr Gastroenterol Nutr 1990;11:448-454. https://doi.org/10.1097/00005176-199011000-00004

[34] Abdullah BA, Gupta SK, Croffie JM, et al. The role of esophagogastroduodenoscopy in the initial evaluation of childhood inflammatory bowel disease: a 7-year study: J Pediatr Gastroenterol Nutr 2002;35:636-640. https://doi.org/10.1097/00005176-200211000-00009

[35] Labrosse R, Graham F, Caubet J-C. Non-IgE-mediated gastrointestinal food allergies in children: an update. Nutrients 2020;12:2086. https://doi.org/10.3390/nu12072086

[36] Savilahti E. Food-induced malabsorption syndromes. J Pediatr Gastroenterol Nutr 2000;30(Supplement):S61-S66. https://doi.org/10.1097/00005176-200001001-00010

[37] Katz Y, Goldberg MR, Rajuan N, et al. The prevalence and natural course of food protein-induced enterocolitis syndrome to cow's milk: a large-scale, prospective population-based study. J Allergy Clin Immunol 2011;127:647-653.e3. https://doi.org/10.1016/j.jaci.2010.12.1105

[38] Alonso SB, Ezquiaga JG, Berzal PT, et al. Food protein-induced enterocolitis syndrome: Increased prevalence of this great unknown - results of the PREVALE study. J Allergy Clin Immunol 2019;143:430-433. https://doi.org/10.1016/j.jaci.2018.08.045

[39] Kokkonen J, Haapalahti M, Tikkanen S, et al. Gastrointestinal complaints and diagnosis in children: a population-based study. Acta Paediatr Oslo Nor 1992 2004;93:880-886.

[40] Maciag MC, Bartnikas LM, Sicherer SH, et al. A slice of food protein-induced enterocolitis syndrome (FPIES): insights from 441 children with FPIES as provided by caregivers in the international FPIES association. J Allergy Clin Immunol Pract 2020;8:1702-1709. https://doi.org/10.1016/j.jaip.2020.01.030

[41] Morita H, Nomura I, Orihara K, et al. Antigen-specific T-cell responses in patients with non-IgE-mediated gastrointestinal food allergy are predominantly skewed to TH2. J Allergy Clin Immunol 2013;131:590-592.e6. https://doi.org/10.1016/j.jaci.2012.09.005

[42] Mehr S, Lee E, Hsu P, et al. Innate immune activation occurs in acute food protein-induced enterocolitis syndrome reactions. J Allergy Clin Immunol 2019;144:600-602.e2. https://doi.org/10.1016/j.jaci.2019.04.021

[43] Caubet JC, Bencharitiwong R, Ross A, et al. Humoral and cellular responses to casein in patients with food protein-induced enterocolitis to cow's milk. J Allergy Clin Immunol 2017;139:572-583. https://doi.org/10.1016/j.jaci.2016.02.047

[44] Vitoria JC, Camarero C, Sojo A, et al. Enteropathy related to fish, rice, and chicken. Arch Dis Child 1982;57:44-48.

[45] Emile J-F, Abla O, Fraitag S, et al. Revised classification of histiocytoses and neoplasms of the macrophage-dendritic cell lineages. Blood 2016;127:2672-2681. https://doi.org/10.1182/blood-2016-01-690636

[46] Weltgesundheitsorganisation. WHO Classification of Tumours of Haematopoietic and Lymphoid Tissues. Revised 4th edition. (Swerdlow SH, Campo E, Harris NL, et al., eds.). International Agency for Research on Cancer, 2017.

[47] Jezierska M, Stefanowicz J, Romanowicz G, et al. Langerhans cell histiocytosis in children - a disease with many faces. Recent advances in pathogenesis, diagnostic examinations and treatment. Adv Dermatol Allergol 2018;35:6-17. https://doi.org/10.5114/pdia.2017.67095

[48] Alston RD, Tatevossian RG, McNally RJQ, et al. Incidence and survival of childhood Langerhans cell histiocytosis in Northwest England from 1954 to 1998. Pediatr Blood Cancer 2007;48:555-560. https://doi.org/10.1002/pbc.20884

[49] Guyot-Goubin A, Donadieu J, Barkaoui M, et al. Descriptive epidemiology of childhood Langerhans cell histiocytosis in France, 2000-2004. Pediatr Blood Cancer 2008;51:71-75. https://doi.org/10.1002/pbc.21498

[50] Salotti JA, Nanduri V, Pearce MS, et al. Incidence and clinical features of Langerhans cell histiocytosis in the UK and Ireland. Arch Dis Child 2009;94:376-380. https://doi.org/10.1136/adc.2008.144527

[51] Singhi AD, Montgomery EA. Gastrointestinal tract langerhans cell histiocytosis: a clinicopathologic study of 12 patients. Am J Surg Pathol 2011;35:305-310. https://doi.org/10.1097/PAS.0b013e31820654e4

[52] Grana N. Langerhans cell histiocytosis. Cancer Control 2014;21:328-334. https://doi.org/10.1177/107327481402100409

[53] Senechal B, Elain G, Jeziorski E, et al. Expansion of regulatory T cells in patients with Langerhans cell histiocytosis. PLoS Med 2007;4:e253. https://doi.org/10.1371/journal.pmed.0040253

[54] Lau SK, Chu PG, Weiss LM. Immunohistochemical expression of langerin in langerhans cell histiocytosis and non-langerhans cell histiocytic disorders. Am J Surg Pathol 2008;32:615-619. https://doi.org/10.1097/PAS.0b013e31815b212b

[55] Black RE, Victora CG, Walker SP, et al. Maternal and child undernutrition and overweight in low-income and middle-income countries. Lancet 2013;382(9890):427-451. https://doi.org/10.1016/S0140-6736(13)60937-X

[56] Kambe T, Fukue K, Ishida R, et al. Overview of Inherited zinc deficiency in infants and children. J Nutr Sci Vitaminol (Tokyo) 2015;61(Supplement):S44-S46. https://doi.org/10.3177/jnsv.61.S44

[57] Ackland ML, Michalczyk A. Zinc deficiency and its inherited disorders -a review. Genes Nutr 2006;1:41-49. https://doi.org/10.1007/BF02829935

[58] Kelly R, Davidson GP, Townley RR, et al. Reversible intestinal mucosal abnormality in acrodermatitis enteropathica. Arch Dis Child 1976;51:219-222. https://doi.org/10.1136/adc.51.3.219

[59] Hosoyamada T. Clinical studies of pediatric malabsorption syndromes. Fukuoka Igaku Zasshi Hukuoka Acta Medica 2006;97:322-350.

[60] Lundström U, Perkkiö M, Savilahti E, et al. Iron deficiency anaemia with hypoproteinaemia. Arch Dis Child 1983;58:438-441. https://doi.org/10.1136/adc.58.6.438

[61] Guha DK, Walia BN, Tandon BN, et al. Small bowel changes in iron-deficiency anaemia of childhood. Arch Dis Child 1968;43:239-244. https://doi.org/10.1136/adc.43.228.239

[62] Stanfield JP, Hutt MS, Tunnicliffe R. Intestinal biopsy in kwashiorkor. Lancet Lond Engl. 1965;2(7411):519-523. https://doi.org/10.1016/s0140-6736(65)91474-1

[63] Theron JJ, Wittmann W, Prinsloo JG. The fine structure of the jejunum in kwashiorkor. Exp Mol Pathol 1971;14:184-199. https://doi.org/10.1016/0014-4800(71)90064-5

[64] Shiner M, Redmond AOB, Hansen JDL. The jejunal mucosa in protein-energy malnutrition. A clinical, histological, and ultrastructural study. Exp Mol Pathol 1973;19:61-78. https://doi.org/10.1016/0014-4800(73)90041-5

[65] Ensari A. Gluten-sensitive enteropathy (celiac disease): controversies in diagnosis and classification. Arch Pathol Lab Med 2010;134:826-836. https://doi.org/10.1043/1543-2165-134.6.826

[66] Koot BG, ten Kate FJ, Juffrie M, et al. Does giardia lamblia cause villous atrophy in children?: a retrospective cohort study of the histological abnormalities in giardiasis. J Pediatr Gastroenterol Nutr 2009;49:304-308. https://doi.org/10.1097/MPG.0b013e31818de3c4

[67] Brown I, Bettington M, Rosty C. The role of histopathology in the diagnosis and management of coeliac disease and other malabsorptive conditions. Histopathology 2021;7888-105. https://doi.org/10.1111/his.14262

[68] Panarelli NC, Lamps LW. Infectious disorders of the upper gastrointestinal tract (excluding Helicobacter pylori). Diagn Histopathol 2020;26:556-565. https://doi.org/10.1016/j.mpdhp.2020.10.003

[69] Panarelli NC. Infectious diseases of the upper gastrointestinal tract. Histopathology 2021;78:70-87. https://doi.org/10.1111/his.14243

[70] Odze RD, Goldblum JR. Odze and goldblum surgical pathology of the GI tract, liver, biliary tract, and pancreas.; 2015. Accessed June 16, 2021. https://www.clinicalkey.com/dura/browse/bookChapter/3-s2.0-C20110697145