The Diagnostic Approach to Monogenic Very Early Onset Inflammatory Bowel Disease

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Patients with a diverse spectrum of rare genetic disorders can present with inflammatory bowel disease (monogenic IBD). Patients with these disorders often develop symptoms during infancy or early childhood, along with endoscopic or histological features of Crohn’s disease, ulcerative colitis, or IBD unclassified. Defects in interleukin-10 signaling have a Mendelian inheritance pattern with complete penetrance of intestinal inflammation. Several genetic defects that disturb intestinal epithelial barrier function or affect innate and adaptive immune function have incomplete penetrance of the IBD-like phenotype. Several of these monogenic conditions do not respond to conventional therapy and are associated with high morbidity and mortality. Due to the broad spectrum of these extremely rare diseases, a correct diagnosis is frequently a challenge and often delayed. In many cases, these diseases cannot be categorized based on standard histological and immunologic features of IBD. Genetic analysis is required to identify the cause of the disorder and offer the patient appropriate treatment options, which include medical therapy, surgery, or allogeneic hematopoietic stem cell transplantation. In addition, diagnosis based on genetic analysis can lead to genetic counseling for family members of patients. We describe key intestinal, extra-intestinal, and laboratory features of 50 genetic variants associated with IBD-like intestinal inflammation. In addition, we provide approaches for identifying patients likely to have these disorders. We also discuss classic approaches to identify these variants in patients, starting with phenotypic and functional assessments that lead to analysis of candidate genes. As a complementary approach, we discuss parallel genetic screening using next-generation sequencing followed by functional confirmation of genetic defects.

Inflammatory bowel diseases (IBDs) are a diverse group of complex and multifactorial disorders. The most common subtypes are Crohn’s disease (CD) and ulcerative colitis (UC).1,2 There is increasing evidence that IBD arises in genetically susceptible people, who develop a chronic and relapsing inflammatory intestinal immune response toward the intestinal microbiota. Disease development and progression are clearly influenced by environmental factors, which have contributed to the rapid global increase in the incidence of IBD in recent decades.3

Developmental, Genetic, and Biological Differences Among Age Groups

IBD location, progression, and response to therapy have age-dependent characteristics.4–10 The onset of intestinal inflammation in children can affect their development and growth. Age of onset can also provide information about the

Abbreviations used in this paper: CD, Crohn’s disease; CGD, chronic granulomatous disease; CVID, combined variable immunodeficiency; EOIBD, early-onset inflammatory bowel disease; HSCT, hematopoietic stem cell transplantation; IBD, inflammatory bowel disease; Ig, immunoglobulin; IL, interleukin; IPEX, immunodysregulation polyendocrinopathy enteropathy X-linked syndrome; NADPH, reduced nicotinamide adenine dinucleotide phosphate; NEMO, nuclear factor ₫ B essential modulator protein; NK, natural killer; PID, primary immunodeficiency; SCID, severe combined immunodeficiency; UC, ulcerative colitis; VEOIBD, very early onset inflammatory bowel disease; WAS, Wiskott–Aldrich syndrome; WES, whole-exome sequencing.

Keywords: Inflammatory Bowel Disease; Crohn’s Disease; Ulcerative Colitis; Unclassified Colitis; Indeterminate Colitis; Immunodeficiency; Pediatrics; IBD Unclassified; Genetics; Next-Generation Sequencing; Whole Exome Sequencing.
type of IBD and its associated genetic features. For example, patients with defects in interleukin (IL)-10 signaling have a particularly early onset of IBD, within the first few months of life. Our increasing understanding of age-specific characteristics has led to changes in the classification of pediatric IBD. Based on disease characteristics, several age subgroups have been proposed that correspond largely to the generally accepted age stages defined by National Institute of Child Health and Human Development pediatric terminology.11

Five major subgroups of pediatric IBD can be summarized according to age (Table 1). The Montreal classification12 originally defined patients with age of onset younger than 17 years as a distinct group of patients with pediatric-onset IBD (A1). The Pediatric Paris modification13 of the Montreal classification12 later defined the pediatric-onset group of IBD as A1 but subdivided those with a diagnosis before 10 years of age as subgroup A1a and those with a diagnosis between 10 and ≤17 years of age as subgroup A1b.13 This reclassification was based on several findings indicating that children with a diagnosis of IBD before 10 years of age develop a somewhat different disease phenotype compared with adolescents or adults. Particular differences that supported the modification were paucity of ileal inflammation and predominance of pancolonic inflammation as well as a low rate of anti-Saccharomyces cerevisiae antibodies in A1a patients with CD, with an increased risk of surgery (colectomy) and biological therapy in A1a patients with UC.13

In this review, we refer to the A1a group as having early-onset IBD (EOIBD). Very early onset IBD (VEOIBD), the subject of this review, represents children with a diagnosis before 6 years of age.14 This age classification includes neonatal, infantile, toddler, and early childhood groups. Proposing an age group between infantile IBD and A1a EOIBD makes sense when taking account that the age of onset is often older than 2 years in multiple relevant subgroups of patients with monogenic IBD (such as those with XIAP deficiency, chronic granulomatous disease [CGD], or other neutrophil defects). On the other hand, from the age of 7 years, there is a substantial rise in the frequency of patients with a diagnosis of conventional polygenic IBD, particularly CD.5,15 This leads to a relative enrichment of monogenic IBD in those with age of onset younger than 6 years. Approximately one-fifth of children with IBD younger than 6 years of age and one-third of children with IBD younger than 3 years of age are categorized as having IBD unclassified (or indeterminate colitis),16 reflecting the lack of a refined phenotyping tool to categorize relevant

<table>
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<th>Group</th>
<th>Classification</th>
<th>Age range (y)</th>
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<tr>
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<td>EOIBD</td>
<td>Paris A1a</td>
<td>Younger than 10</td>
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<td>VEOIBD</td>
<td></td>
<td>Younger than 6</td>
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<td>Infantile (and toddler)</td>
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<td>Younger than 2</td>
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<td>onset IBD</td>
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<td>Neonatal IBD</td>
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<td>First 28 days of age</td>
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The enrichment of monogenic defects in EOIBD and VEOIBD becomes apparent when relating the approximately 1% of patients with IBD younger than 6 years of age and <0.2% younger than 1 year of age to reports that the majority of monogenic disorders can present at younger than 6 years of age and even younger than 1 year of age (Figure 1).

Although it is generally accepted that many patients with VEOIBD have low response rates to conventional anti-inflammatory and immunomodulatory therapy, there is a paucity of well-designed studies to support this hypothesis. Infantile (and toddler) onset of IBD was highlighted in the Pediatric Paris classification because of higher rates of affected first-degree family relatives, indicating an increased genetic component, severe disease course, and high rate of resistance to immunosuppressive treatment.13 Features of autoimmunity with dominant lymphoid cell infiltration are frequently found in infants and toddlers.17 Such patients are likely to have pancolitis; subgroups of patients develop severely ulcerating perianal disease, and there is a high rate of resistance to conventional therapy, a high rate of first-degree relatives with IBD, and increased lethality.4–8 Recent guidelines and consensus approaches on the diagnosis and management of IBD16,19 highlight that children with infantile onset of IBD have a particular high risk of an underlying primary immunodeficiency. An extreme early subgroup, neonatal IBD, has been described with manifestations during the first 27 days of life.4,5,8

Guidelines on the diagnosis and classification of IBD in pediatric patients13,18–22 have addressed the need to recognize monogenic disorders and immunodeficiencies in particular, because these require a different treatment strategy than conventional IBD. Current guidelines do not, however, cover the spectrum of these rare subgroups of monogenic IBD. The identification of an underlying genetic defect is indeed challenging, owing to the orphan nature of these diseases, the wide phenotypic spectrum of disorders, and the limited information available on most genetic defects. This review and practice guide provides a comprehensive summary of the monogenic causes of IBD-like intestinal inflammation and a conceptual framework for the diagnostic evaluation of patients with suspected monogenic IBD. We categorize known genetic defects into functional subgroups and discuss key intestinal and extraintestinal findings. Based on the enrichment of known causative mutations as well as extreme phenotypes in very young children, we have focused on a practical approach to detect monogenic disorders in patients with VEOIBD and infantile IBD in particular. Because there is only modest biological evidence to support age-specific categorization of IBD above infantile IBD and within the EOIBD subgroup, we also discuss disease- and gene-specific ages of onset of intestinal inflammation (Figure 1).

**Epidemiology of Pediatric IBD**

Approximately 20% to 25% of patients with IBD develop intestinal inflammation during childhood and adolescence.
IBD in children younger than 1 year of age has been reported in approximately 1% and VEOIBD in approximately 15% of pediatric patients with IBD. VEOIBD has an estimated incidence of 4.37 per 100,000 children and a prevalence of 14 per 100,000 children. The incidence of pediatric IBD is increasing. Some studies have reported that the incidence of IBD is increasing particularly rapidly in young children, although not all studies have confirmed this observation.

Polygenic and Monogenic Forms of IBD

Twin studies have provided the best evidence for a genetic predisposition to IBD, which is stronger for CD than UC. Conventional IBD is a group of polygenic disorders in which hundred(s) of susceptibility loci contribute to the overall risk of disease. Meta-analyses of (genome-wide) association studies of adolescent- and adult-referral-based IBD cohort, n = 1605 patients comprising CD, UC, and IBD unclassified (IBDU). Symbols represent individual patients. Bars represent the age range of case series if individual data were not available. The age ranges of infantile IBD, VEOIBD, EOIBD, and Montreal/Paris classification A1a, A1b, A2, and A3 are shown for reference. Age of onset data refer to references provided in Table 2. Additional references for disease subgroups are provided in Supplementary Information for Figure 1.
important to consider that these 163 loci individually contribute only a small percentage of the expected heritability in IBD.26 This suggests that IBD, including CD and UC, can be regarded as a classic polygenic disorder. Findings from initial genome-wide pediatric association studies focused on adolescents and confirm a polygenic model.27,28 There are no well-powered genome-wide association studies of patients with EOIBD or VEOIBD.

Although most cases of IBD are caused by a polygenic contribution toward genetic susceptibility, there is a diverse spectrum of rare genetic disorders that produce IBD-like intestinal inflammation.29 The genetic variants that cause these disorders have a large effect on gene function. However, these variants are so rare in allele frequency (many private mutations) that those genetic signals are not detected in genome-wide association studies of patients with IBD. With recent advances in genetic mapping and sequencing techniques and increasing awareness of the importance of those “orphan” disorders, approximately 50 genetic disorders have been identified and associated with IBD-like immunopathology (for a partial summary, see Uhlig29). For simplicity, we refer to these disorders in the following text as monogenic IBD, even if there is a spectrum of penetrance of the IBD phenotype. We will compare those monogenic forms of IBD with polygenic conventional IBD.

All data suggest that the fraction of monogenic disorders with IBD-like presentation among all patients with IBD correlates inversely with the age of onset. Despite a growing genotype spectrum, monogenic disorders still account for only a fraction of VEOIBD cases. The true fraction is unknown. In a study of 66 patients who developed IBD at ages younger than 5 years, 5 patients were found to carry mutations in IL10RA, 8 in IL10RB, and 3 in IL10.30 All patients developed symptoms within the first 3 months of life.30 A recent study detected 4 patients with presumed pathogenic XIAP mutations in a group of 275 patients with pediatric IBD (A1a/A1b Paris classification) and 1047 patients with adult-onset CD (A2 and A3 Montreal classification).31 Because all patients with XIAP variants were infantile to adolescent male patients with CD, this could suggest an approximate prevalence of 4% among young male patients with IBD. However, studies like these focus on specific genes and may have strong selection bias toward an expected clinical subphenotype. They might therefore overestimate the frequency of specific variants. Analysis of large, multicenter, population-based cohorts is needed to determine the proportion of cases of VEOIBD caused by single gene defects and to estimate penetrance.

Monogenic defects have been found to alter intestinal immune homeostasis via several mechanisms (Table 2). These include disruption of the epithelial barrier and the epithelial response as well as reduced clearance of bacteria by neutrophil granulocytes and other phagocytes. Other single-gene defects induce hyperinflammation or auto-inflammation or disrupt T- and B-cell selection and activation. Hyperactivation of the immune response can result from defects in immune inhibitory mechanisms, such as defects in IL-10 signaling or dysfunctional regulatory T-cell activity.

### Epithelial Barrier and Response Defects

Genetic disorders that affect intestinal epithelial barrier function include dystrophic epidermolysis bullosa,32 Kindler syndrome,33 familial diarrhea caused by dominant activating mutations in guanylate cyclase C,34 X-linked ectodermal dysplasia and immunodeficiency,35 and ADAM17 deficiency.36

X-linked ectodermal dysplasia and immunodeficiency, caused by hypomorphic mutations in IKBKG (encodes nuclear factor κB essential modulator protein [NEMO])34 and ADAM17 deficiency35 cause epithelial and immune dysfunction. Recently, TTC7A deficiency was described in patients with multiple intestinal atresia, with and without severe combined immunodeficiency (SCID) immunodeficiency.36,37 Hypomorphic mutations in TTC7A without intestinal strictureing or severe immunodeficiency, most likely due to a defect in epithelial signaling.38

### Dysfunction of Neutrophil Granulocytes

Variants in genes that affect neutrophil granulocytes (and other phagocytes) predispose people to IBD-like intestinal inflammation. Chronic granulomatous disease is characterized by genetic defects in components of the phagocyte reduced nicotinamide adenine dinucleotide phosphate (NADPH) oxidase (phox) complex. Genetic mutations in all 5 components of the phagocyte NADPH oxidase (phox)—gp91-phox (CYBB), p22-phox (CYBA), p47-phox (NCF1), p67-phox (NCF2), and p40-phox (NCF4)—are associated with immunodeficiency and can cause IBD-like intestinal inflammation.

As high as 40% of patients with CGD develop CD-like intestinal inflammation.39–41 Multiple granulomas and the presence of pigmented macrophages can indicate the group of defects histologically. Missense variants in NCF2 that affect RAC2 binding sites have recently been reported in patients with VEOIBD.42 Recently, several heterozygous functional hypomorphic variants in multiple components of the NOX2 NADPH oxidase complex were detected in patients with VEOIBD that do not cause CGD-like immunodeficiency but have a moderate effect on reactive oxygen species production and confer susceptibility to VEOIBD.43

Tumor necrosis factor α inhibitors can resolve intestinal inflammation in patients with CGD but could increase the risk of severe infections in patients with CGD.44 Allogenic hematopoietic stem cell transplantation (HSCT) can cure CGD and resolve intestinal inflammation.45–47 Monocytes produce high levels of IL-1 in patients with CGD, and an IL-1 receptor antagonist (anakinra) has been used to treat noninfectious colitis in those patients.48

In addition to CGD, a number of another neutrophil defects are associated with intestinal inflammation. Defects in glucose-6-phosphate translocase (SLC37A4)49,50 and glucose-6-phosphatase catalytic subunit 3 (G6PC3)50 are associated with congenital neutropenia (and other distinct features) but also predispose people to IBD. Leukocyte adhesion deficiency type 1 is caused by mutations in the gene encoding CD18 (ITGB2) and is associated with...
Table 2. Genetic Defects and Phenotype of Monogenic IBD

<table>
<thead>
<tr>
<th>Group</th>
<th>Syndrome/disorder</th>
<th>Gene</th>
<th>Inheritance</th>
<th>CD-like</th>
<th>Granuloma</th>
<th>UC-like</th>
<th>Epithelial defect (apoptosis)</th>
<th>Disease location (1–5)</th>
<th>Perianal fistula/abscess</th>
<th>Penetrating fistulas</th>
<th>Strictures</th>
<th>Skin lesions</th>
<th>Autoimmunity, inflammation</th>
<th>HLH/MASS</th>
<th>Neoplasia</th>
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<td>Epithelial barrier</td>
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<td>Familial hemophagocytic lymphohistiocytosis type 5</td>
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<th>Syndrome/disorder</th>
<th>Gene</th>
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<th>UC-like</th>
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<th>Disease location (1–5)</th>
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NOTE. Genetic defects are grouped according to functional subgroups. Gene names refer to HUGO gene nomenclature. CD-like and UC-like were marked only when patient characteristics in the original reports were described as typical CD or UC pathologies. Unclassified or indeterminate colitis is the not specified default option. Disease location is classified as follows: 1, mouth; 2, enteropathy; 3, enterocolitis; 4, isolated ileitis; 5, colitis; 6, perianal disease. Epithelial defects refer in particular to finding of epithelial lining nonadherent at the basal membrane or increased epithelial apoptosis and epithelial tufting. Key laboratory findings are provided in Supplementary Table 1, and examples of additional defects of possible or unclear relevance are listed in the Supplementary Information for Table 1.

HLH, hemophagocytic lymphohistiocytosis; AR, autosomal recessive; eb, epidermolysis bullosa; X, X-linked; A, arthritis; vasc, vasculitis; n, nail; h, hair; AD, autosomal dominant; e, eczema; f, folliculitis/pyoderma; SJ, Sjögren syndrome; p, psoriasis; AIHA, autoimmune hemolytic anemia; AN, autoimmune neutropenia; PSC, primary sclerosing cholangitis; HT, Hashimoto thyroiditis; AIH, autoimmune hepatitis; TID, type 1 diabetes mellitus; MAS, macrophage activation syndrome; NSIP, non-specific interstitial pneumonitis; S, serositis.

aPersonal information and communication.
defective transendothelial migration of neutrophil granulocytes. Patients typically present with high peripheral granulocyte counts and bacterial infections, and some present with IBD-like features.51,52

CD-like disease is a typical manifestation of glycogen storage disease type Ib, characterized by neutropenia and neutrophil granulocyte dysfunction.40,49,53 Granulocyte colony-stimulating factor has been used to treat neutropenia and neutrophilia in patients with glycogen storage disease type Ib.53

In addition to neutrophil defects, defects in several other genes, including WAS, LRBA, BTK, CD40LG, and FOXP3, can lead to autoantibody-induced or hemophagocytosis-induced neutropenia. These multidimensional mechanisms of secondary immune dysregulation indicate the functional complexity of some seemingly unrelated genetic immune defects and the broad effects they might have on the innate immune system.

**Hyperinflammatory and Autoinflammatory Disorders**

VEOIBD has been described in a number of hyperinflammatory and autoinflammatory disorders such as mevalonate kinase deficiency,54,55 phospholipase C-γ2 defects,66 familial Mediterranean fever,57-59 Hermansky–Pudlak syndrome (type 1, 4, and 6),60-64 X-linked lymphoproliferative syndrome type 165 and type 2,66-68 or familial hemophagocytic lymphohistiocytosis type 5.69

Among these, mevalonate kinase deficiency is a prototypic autoinflammatory disorder, characterized by increased activation of caspase-1 and subsequent activation of IL-1β.70 Inhibiting IL-1β signaling with antibodies that block IL-1β or IL-1 receptor antagonists can induce complete or partial remission in patients, including those with VEOIBD.54,55,71

X-linked lymphoproliferative syndrome 2 is caused by defects in the XIAP gene. At least 20% of patients with XIAP defects develop a CD-like immunopathology with severe fistulizing perianal phenotype.66-68,72,73 In these patients, Epstein–Barr virus infections can lead to life-threatening hemophagocytic lymphohistiocytosis. Originally associated with a poor outcome after HSCT,74 less toxic induction regimens could improve the prognosis and cure this form of IBD.67,73

**Complex Defects in T- and B-Cell Function**

IBD-like immunopathology is a common finding in patients with defects in the adaptive immune system. Multiple genetic defects that disturb T- and/or B-cell selection and activation can cause complex immune dysfunction, including immunodeficiency and autoimmunity as well as intestinal inflammation. Disorders associated with IBD-like immunopathology include B-cell defects such as common variable immunodeficiency (CVID), hyper-immunoglobulin (Ig) M syndrome, and agammaglobulinemia.75-79 Several other primary immune deficiencies, such as Wiskott-Aldrich syndrome80 (WAS) and atypical SCID or Omenn syndrome81,82 can also cause IBD-like intestinal inflammation.

**CVID, Agammaglobulinemia, and Hyper IgM Syndrome**

Patients with CVID have clinical features of different types of IBD, spanning CD, UC, and ulcerative proctitis–like findings.83,84 Although CVID is largely polygenic, a small proportion of cases of CVID have been associated with specific genetic defects. CVID type 1 is caused by variants in the gene encoding the inducible T-cell costimulator (ICOS),85-86 whereas CVID type 8 is caused by variants in LRBA.87-89 Patients with these mutations can present with IBD-like pathology. Recently, IBD and CVID-like disease was described in a family with IL-21 deficiency.90

Patients with agammaglobulinemia, caused by defects in BTK or PIK3RI, as well as patients with subtypes of hyper IgM syndrome caused by defects in CD40LG, AICDA, or IKKβ can develop IBD-like immunopathology.75-79 It is worth considering that several other immunodeficiencies, not regarded as primary B-cell defects, are similarly associated with low numbers of B cells and/or Igss (such as those caused by variants in SKIV2L and TTC37; see Table 2 and Supplementary Table 1).

**WAS**

WAS is a primary immunodeficiency. Many patients with WAS present with UC-like noninfectious colitis during early infancy.80 The syndrome is caused by the absence or abnormal expression of the cytoskeletal regulator WASP and is associated with defects in most immune subsets (effector and regulatory T cells, natural killer [NK] T cells, B cells, dendritic cells, macrophages, NK cells, and neutrophils).91 In addition to features of UC, patients develop many other autoimmune complications. Allogeneic bone marrow transplantation is the standard of care for those patients.80 Patients who are not candidates for bone marrow transplantation have been successfully treated with experimental gene therapy approaches.92,93

**Atypical SCID Defects**

Patients with atypical SCID defects have residual B- and T-cell development and oligoclonal T-cell expansion.94 VEOIBD is commonly observed in patients with atypical SCID due to hypomorphic defects in multiple genes such as DCLRE1C, ZAP70, RAG2, IL2RG, LG4, ADA, and CD3G.91,92,95 This list of genes is likely not complete, and it seems reasonable to assume that most genetic defects that cause T-cell atypical SCID also cause IBD.

A subset of patients with SCID present with severe eczematous rash (Omenn syndrome).91 It is not clear whether residual lymphocye function in patients with hypomorphic TTC7A mutations is a precondition for IBD or contributes to VEOIBD.93 Intestinal and skin lesions also develop in patients with SCID due to graft-versus-host disease in response to maternal cells.92
Hoyeraal–Hreidarsson Syndrome

Hoyeraal–Hreidarsson syndrome is a severe form of dyskeratosis congenita characterized by dysplastic nails, lacy reticular skin pigmentation, and oral leukoplakia. It is a multiorgan disorder. Patients with mutations in RTEL1\textsuperscript{97,98} or DKC1\textsuperscript{99–101} can develop SCID and intestinal inflammation.

Regulatory T Cells and IL-10 Signaling

Loss-of-function defects in IL-10 and its receptor (encoded by IL10RA and IL10RB)\textsuperscript{102–106} cause VEOIBD with perianal disease and folliculitis within the first months of life. All patients with loss-of-function mutations that prevent IL-10 signaling develop IBD-like immunopathology, indicating that these defects are a monogenic form of IBD with 100% penetrance.\textsuperscript{106,107} The anti-inflammatory cytokine IL-10 is secreted by natural and induced regulatory T cells (in particular, intestinal CD4\textsuperscript{+} FOXP3\textsuperscript{+} and Tr1 cells), macrophages, and B cells. Many intestinal and extraintestinal cell types express the IL-10 receptor and respond to IL-10. Defects in IL-10 receptor signaling affect the differentiation of macrophage M1/M2, shifting them toward an inflammatory phenotype.\textsuperscript{108} Defects in IL-10 signaling are associated with extraintestinal inflammation such as folliculitis or arthritis and predispose to B-cell lymphoma.\textsuperscript{102,103,109} Conventional therapy options are largely not effective in patients with IL-10 signaling defects, but allogeneic matched or mismatched HSCT can induce sustained remission of intestinal inflammation.\textsuperscript{30,102,103,107,110}

X-linked immune dysregulation, polyendocrinopathy, enteropathy syndrome (IPEX) is caused by mutations in the transcription factor FOXP3. Those mutations affect natural and induced regulatory T cells, causing autoimmunity and immunodeficiency but also enteropathy in a large percentage of patients with colitis.\textsuperscript{111,112} The intestinal lesions that develop in patients with IPEX can be classified as graft-versus-host disease–like changes with small bowel involvement and colitis, celiac disease–like lesions, or enteropathy with goblet cell depletion.\textsuperscript{113}

Antibodies against enterocytes and/or antibodies against goblet cells can be detected in the serum of patients with IPEX.\textsuperscript{113} IPEX-like immune dysregulation with enteropathy can also be caused by defects in IL-2 signaling in patients with defects in the IL-2 receptor \( \alpha \) chain (IL2RA, encoding CD25)\textsuperscript{114,115} or a dominant gain of function in STAT1 signaling.\textsuperscript{116}

Other Disorders and Genes

IBD or IBD-like disorders have been described in patients with several other disorders. In some disorders, there is no well-defined plausible functional mechanism. For example, patients with trichohepatoenteric syndrome have presumed defects in epithelial cells that lead to intractable diarrhea.\textsuperscript{117,118} However, an adaptive immune defect might also cause this disorder, because the patients have Ig deficiencies that require Ig substitution.

Several genes, described in the Supplementary Information for Table 1, are associated with a single or less well-defined case report of patients who developed IBD-like features. Some of these patients might happen to have intestinal inflammation by coincidence, and even several case reports cannot exclude a publication bias.

Heterozygous defects in the PTEN phosphatase are associated not only with multiple tumors but also immune dysregulation and autoimmunity.\textsuperscript{119} Inflammatory polyps are common among patients with PTEN hamartoma tumor syndrome and indeterminate colitis, and ileitis is a rare complication.\textsuperscript{119} The functional mechanism involved in intestinal inflammatory polyps and intestinal inflammation is not clear because heterozygous mutations in PTEN are not associated with conventional immunodeficiency and affect multiple cell types.

Very early onset enteropathies and intestinal infections are described in several monogenic immunodeficiency and/or autoinflammation disorders, including defects in the itchy E3 ubiquitin ligase activity encoded by the ITCH gene, defects in E3 ubiquitin ligase HOIL-1 encoded by HOIL1, and gain of function defects in IKBA (see Supplementary Information for Table 1). It is not clear what activates the inflammatory events in those patients; it could be pathogenic microbes in the intestine, food, or IBD-like intestinal inflammation induced by the commensal microbiota.

Additional disorders are associated with intestinal inflammation without immunodeficiency or without known epithelial mechanisms. For example, some patients with Hirschsprung disease, an intestinal innervation and dysmotility disorder, develop enterocolitis associated with dominant germline mutations in RET.\textsuperscript{120,121} One possible pathomechanism could be increased bacterial translocation due to bacterial stasis leading to subsequent inflammation.

Despite multiple reports of complement system deficiencies and IBD, this group of disorders is not clearly defined. MASP2 deficiency has been reported in a patient with pediatric-onset IBD. However, reports of intestinal inflammation in several other complement defects are much harder to interpret because those patients present with inconsistent disease phenotypes; some are less well documented and could be simple chance findings (see Supplementary Information for Table 1).

Why Should We Care About Monogenic Defects?

It is a challenge to diagnose the rare patients with monogenic IBD, but differences in the prognosis and medical management argue that a genetic diagnosis should not be missed. As a group, these diseases have high morbidity and subgroups have high mortality if untreated. Based on their causes, some require different treatment strategies than most cases of IBD.

Allogeneic HSCT has been used to treat several monogenic disorders. It is the standard treatment for patients with disorders that do not respond to conventional treatment, those with high mortality, or those that increase susceptibility to hematopoietic cancers (eg IL-10 signaling defects, IPEX, WAS, or increasingly XIAP deficiency). Introduction of HSCT as a potentially curative treatment option
for intestinal and extraintestinal manifestations of these disorders has changed clinical practice.30,73,74,107,111

However, there is evidence from mouse models and clinical studies that patients with epithelial barrier defects are less amenable to HSCT, because this does not correct the defect that causes the disease (eg, NEMO deficiency or possibly TTC7A deficiency). For example, severe recurrence of multiple intestinal atresia after HSCT in patients with TTC7A deficiency16,37 indicates a contribution of the enterocyte defect to pathogenesis. Due to the significant risk associated with HSCT, including graft-versus-host disease and severe infections, it is important to determine the genetic basis of each patient’s VEOIBD before selecting HSCT as a treatment approach.

Understanding the pathophysiology of a disorder caused by a genetic defect can identify unconventional biological treatment options that interfere with specific pathogenic pathways. Patients with mevalonate kinase deficiency or CGD produce excess amounts of IL-1β, so treatment with IL-1β receptor antagonists has been successful.54,55 This treatment is not part of the standard therapeutic repertoire for patients with conventional IBD. Access to individualized genotype-specific therapies is particularly important, because it might avoid both surgery (including colectomy) and the adverse effects of medical therapy in patients who are unlikely to benefit from conventional IBD therapies in the long term.

A further incentive to establish a specific genetic diagnosis is the ability to anticipate complications. Some patients should be screened for infections (such as for Epstein–Barr virus infection status in XIAP defects) or cancer (including B-cell lymphomas in patients with IL-10 receptor deficiency109 or skin and hematopoietic malignancies in Hoyeraal–Hreidarsson syndrome). Genetic information can also identify patients who should be screened for extraintestinal manifestations such as idiopathic thrombocytopenic purpura, autoimmune hemolytic anemia, autoimmune neutropenia, or autoimmune hepatitis (Table 2).

Knowledge of the genetic predisposition can reduce the time to detect associated complications.

Families who are aware of the genetic basis of their disease can receive genetic counseling.

### When Should We Suspect Monogenic IBD?

The timely diagnosis of monogenic IBD requires assessments of intestinal and extraintestinal disease phenotypes in conjunction with the histopathology and appropriate laboratory tests to exclude allergies or infections.18,19 Classification of clinical, endoscopic, histological, and imaging findings into CD-like and UC-like phenotypes can be helpful but is not sufficient to differentiate patients with a monogenic disorder from conventional idiopathic CD (such as discontinuous, transmural inflammation affecting the entire gastrointestinal tract, fistulizing disease, or granuloma formation) or UC (a continuous, colonic disorder with crypt abscess formation and increases in chronic inflammatory cells, typically restricted to the lamina propria). Histopathologists use nonspecific terms such as IBD unclassified in a relevant proportion of patients with VEOIBD, including monogenic forms of IBD. In the absence of highly specific and sensitive intestinal histological markers of monogenic forms of IBD, extraintestinal findings and laboratory test results are important factors to focus the search for monogenic forms of IBD (Table 3 and Figure 2). A phenotypic aide-mémoire summarizing the key findings to ensure that a careful clinical history for VEOIBD and examination to narrow the search for an underlying monogenetic defect is YOUNG AGE MATTERS MOST (YOUNG AGE onset, Multiple family members and consanguinity, Autoimmunity, Thriving failure, Treatment with conventional medication fails, Endocrine concerns, Recurrent infections or unexplained fever, Severe perianal disease, Macrophage activation syndrome and hemophagocytic lymphohistiocytosis, Obstruction and atresia of intestine,

### Table 3. Pivotal Prompts for Suspecting Monogenic IBD

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<thead>
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<th>Key points</th>
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<td>Very early age of onset of IBD-like immunopathology</td>
<td>Likelihood increases with very early onset, particularly in those younger than 2 years of age at diagnosis</td>
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<tr>
<td>Family history</td>
<td>In particular consanguinity, predominance of affected males in families, or multiple family members affected</td>
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<tr>
<td>Atypical endoscopic or histological findings</td>
<td>For example, extreme epithelial apoptosis or loss of germinal centers</td>
</tr>
<tr>
<td>Resistance to conventional therapies</td>
<td>Such as exclusive enteral nutrition, corticosteroids, and/or biological therapy</td>
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<td>Skin lesions, nail dystrophy, or hair abnormalities</td>
<td>For example, epidermolysis bullosa, eczema, folliculitis, pyoderma or abscesses, woolen hair, or trichorrhexis nodosa</td>
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<tr>
<td>Severe or very early onset perianal disease</td>
<td>Fistulas and abscesses</td>
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<td>Lymphoid organ abnormalities</td>
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<td>Recurrent or atypical infections</td>
<td>Intestinal and nonintestinal</td>
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<td>Hemophagocytic lymphohistiocytosis</td>
<td>Induced by viral infections such as Epstein–Barr virus or cytomegalovirus or macrophage activation syndrome</td>
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<td>For example, arthritis, serositis, sclerosing cholangitis, anemia, and endocrine dysfunction such as thyroiditis, type 1 diabetes mellitus</td>
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<tr>
<td>Early development of tumors</td>
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</tbody>
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Skin lesions and dental and hair abnormalities, and Tumors). An important component of management is to solicit advice from a specialist in VEOIBD.

Very early age of onset of intestinal symptoms and IBD-like endoscopic and histological changes are strong indicators of monogenic IBD as a group (Figure 1). However, there are clear gene-specific differences in the age of onset. The reported time of onset of IBD-like immunopathology in subgroups with, for example, IL-10 signaling defects, WAS, or IPEX, is infancy and early childhood. However, atypical late onset of IBD has been reported in patients with WAS122,123 as well as IPEX.124–126 The age is variable in neutrophil defects, B-cell defects, and XIAP deficiency. Indeed, XIAP deficiency caused by identical genetic defects within families can be associated with VEOIBD or adult-onset IBD.68,73,127 Other diseases, such as GUCY2C deficiency, typically develop during adulthood (Figure 1). Phenotypes of many monogenic forms of IBD change over time; gastrointestinal problems can present as an initial or a later finding.

Some candidate disorders will be recognized by their pathognomonic symptom combinations. Because there are no specific and fully reliable endoscopic and histological features of monogenic VEOIBD, patients with VEOIBD and multiple other features (listed in Table 3) should be considered to have increased likelihood to carry disease-causing mutations. The degree of suspicion should dictate the extent of functional and genetic exploration for an underlying cause. It is important to emphasize that in the majority of patients with infantile IBD or VEOIBD, no genetic defect has currently been discovered that would explain the immunopathology. This fraction of causative defects will increase as our knowledge expands and with a growing number of patients undergoing whole-exome sequencing (WES). Although young age of IBD onset is a strong indicator, a strong suspicion for a monogenic cause should lead to limited functional or genetics screening irrespective of age.

Laboratory Tests and Functional Screens

Laboratory tests, upper and lower gastrointestinal endoscopy with histological analysis of multiple biopsy specimens, and imaging should be performed for every patient with VEOIBD according to guidelines.13,18–21,128 Histological investigation is paramount not only to differentiate IBD-like features but also to exclude other established pathologies such as eosinophilic or allergic gastrointestinal disease and infection.
Cow’s milk protein allergy is common and can cause severe colitis that resembles UC and even requires hospitalization. It manifests typically within the first 2 to 3 months of exposure to cow’s milk protein. This may be apparent with breast-feeding or only after introducing formula feeding. Colitis resolves after cow’s milk is removed from the diet, so a trial of exclusive feeding with an amino acid–based infant formula is a customary treatment strategy for all VEOIBD diagnosed when the patient is younger than 1 year of age. However, improvement of symptoms or inflammation does not exclude the possibility that a patient could have a monogenic IBD disorder, because food intolerance and allergy can be secondary to the disorder and allergen avoidance by exclusive enteral nutrition with elemental formula could also alleviate the inflammation of classic IBD.

High levels of IgE and/or eosinophilia are also found in patients with monogenic disorders caused by defects in FOXP3, IL2RA, IKBKG, WAS, or DOK8 (Table 2 and Supplementary Table 1). It should also be standard practice to exclude infectious causes such as bacteria (Yersinia spp, Salmonella spp, Shigella spp, Campylobacter spp, Mycobacterium tuberculosis, Clostridium difficile), parasites (Entamoeba histolytica, Giardia lamblia), and viral infections (cytomegalovirus or human immunodeficiency virus), remembering that some infections can mimic IBD. However, most of these pathogens do not cause bloody diarrhea for more than 2 to 3 weeks. In addition, monogenic disorders (such as B- or T-cell defect immunodeficiencies or familial HLH type 5, caused by STXB2 deficiency) predispose patients to intestinal infections. Celiac disease should be considered as a differential diagnosis for patients with suspected autoimmune enteropathy presenting with villous atrophy (such as IPEX or IPEX-like patients).

To detect possible causes of monogenic IBD-like immunopathology, we propose additional laboratory screening for all children diagnosed before 6 years of age. The limited set of laboratory tests includes measurements of IgA, IgE, IgG, and IgM; flow cytometry analysis of lymphocyte subsets (CD3, CD4, CD8, CD19/CD20, NK cells); and analysis of oxidative burst by neutrophils (using the nitro blue tetrazolium test or flow cytometry–based assays such as the dihydroorhodamine fluorescence assay).

When placed in the context of clinical, histopathologic, and radiological data, these tests can guide the diagnosis toward the more prevalent defects of neutrophil, B-cell, or T-cell dysfunction. Further tests are necessary to characterize particular subgroups, such as those who develop the disease when they are younger than 2 years of age, those with excessive autoimmunity, or those with severe perianal disease. Those tests include flow cytometry analysis of XIAP expression by lymphocytes and NK cells or FOXP3 expression in CD4+ T cells, which can diagnose a significant proportion of patients with XLP2 and IPEX. Flow cytometry can detect functional defects in MDP signaling in patients with XIAP deficiency. IL10RA and IL10RB defects can be detected by assays that determine whether exogenous IL-10 will suppress lipopolysaccharide-induced peripheral blood mononuclear cell cytokine secretion or IL-10–induced STAT3 phosphorylation. Increased levels of antibodies against enterocytes can indicate autoimmune enteropathy, in particular in patients with IPEX.

In contrast to measurements of Igs, flow cytometry, and oxidative burst assays (which are largely standardized), other tests such as IL-10–mediated suppression of LPS–induced peripheral blood mononuclear cell activation and detection of antibodies against enterocytes are not routine assays. Similarly, additional tests for extremely rare genetic defects might be appropriate but are only available at specialized laboratories, often as part of research projects. The clinical utility of the algorithm to use a limited set of laboratory tests to differentiate between conventional and monogenic VEOIBD, as suggested in Figure 2, is based on experience, case reports, and case series of individual disorders. It has not been validated in prospective studies of patients with all forms of VEOIBD.

### Diagnosis via Sequencing of Candidate Genes versus Parallel Next-Generation Sequencing

The classic approach to detect monogenic forms of IBD, as described in the preceding text and summarized in Figure 2, is based on careful phenotypic analysis and candidate sequencing to confirm a suspected genetic diagnosis. Due to the increasing number of candidate genes, sequential candidate sequencing can be costly and time consuming. It is therefore not surprising to propose that this strategy of functional screening followed by genetic confirmation will increasingly be complemented by early parallel genetic screening using next-generation sequencing followed by functional confirmation. The US Food and Drug Administration has recently granted marketing authorization for the first next-generation genomic sequencer, which will further pave the way for genome, exome, or other targeted parallel genetic tests in routine practice. WES or even whole-genome sequencing will increasingly become part of the routine analysis of patients with suspected genetic disorders including subtypes of IBD. This has several important implications for selecting candidate gene lists, identification of disease-causing variants, and dealing with a large number of genetic variants of unknown relevance. In research and clinical settings, WES has been shown to reliably detect genetic variants that cause VEOIBD in genes such as XIAP, IL10RA, IL10RB, G6PC3, MEFV, LRBA, FOXP3, and TTC7A.

There are several reasons to propose extended parallel candidate sequencing for patients with suspected monogenic IBD. Immune and gastrointestinal phenotypes of patients evolve over time, whereas the diagnosis needs to be made at the initial presentation to avoid unnecessary tests and treatment. IBD-like immunopathology can be linked to nonclassic phenotypes of known immunodeficiencies, such as hypomorphic genetic defects in SCID patients (in genes such as ZAP70, RAG2, IL2RG, IL14, ADA, DCLRE1C, CD3G, or TTC7A; see Table 2) with residual B- and T-cell development, glucose-6-phosphatase 3 deficiency with lymphopenia, or FOXP3 defects without the classic IPEX phenotype. WES has revealed unexpected known causative variants even after workup in centers with specialized
immunologic and genetic clinical and research facilities. This all demonstrates that current knowledge about the disease phenotype spectrum is incomplete, which means that a pure candidate approach is not reliable and genetic screening may have advantages. The 50 monogenic defects associated with IBD provide an initial filter to identify patients with monogenic disorders.

Because of the greatly reduced costs of next-generation sequencing, it is probably cost effective in many cases to perform multiplex gene sequencing, WES, or whole-genome sequencing rather than sequential Sanger sequencing of multiple genes. A big advantage of WES is the potential to identify novel causal genetic variants once the initial candidate filter list of known disease-causing candidates has been analyzed. The number of gene variants associated with VEOIBD is indeed constantly increasing, largely due to the new sequencing technologies, so data sets derived from WES allow updated analysis of candidates as well as novel genes. Because multiple genetic defects can lead to spontaneous or induced colitis in mice, assuming homology, it is likely that many additional human gene variants will be associated with IBD.

Targeted sequencing of genes of interest is an alternative approach to exome-targeted sequencing. Initial studies to perform targeted next-generation parallel sequencing showed the potential power of this approach. Targeted next-generation sequencing of the 170 primary immunodeficiency (PID)-related genes accurately detected point mutations and exonic deletions. Only 9 of 170 PID-related genes analyzed showed inadequate coverage. Four of 26 patients with PID without an established prescreening genetic diagnosis, despite routine functional and genetic testing, were diagnosed, indicating the advantage of parallel genetic screening. Because a major group of VEOIBD-causing variants is associated with PID-related genes, it is obvious how this approach can be adapted and extended to monogenic IBD genes.

Genetic approaches also offer practical advantages. Specialized functional immune assays are often only available in research laboratories and are not necessarily validated; functional tests often require rapid processing of peripheral blood mononuclear cells or biopsy specimens in specialized laboratories. This means that handling of DNA and sequencing seems far less prone to error or variation.

However, relying solely on genetic screening can be misleading, because computational mutation prediction can fail to detect functional damaging variants. For example, variants in the protein-coding region of the IL10RA gene were misclassified as “tolerated” by certain prediction tools, whereas other prediction tools and functional analysis reported defects in IL-10 signaling. Although most studies report variants in protein-coding regions in monogenic diseases, there could be selection bias. It is indeed far more difficult to establish the biological effects of variants that affect processes such as splicing, gene expression, or messenger RNA stability. It should go without saying that novel genetic variants require appropriate functional validation.

The increased availability of sequencing data sets highlights the role of mutation-specific IBD-causing variants that illustrate the functional balance of gene products affected by gain or loss of function variants as well as gene dosage effects. Inherited gain-of-function mutations in guanylyl cyclase cause diarrhea and increase susceptibility to IBD, whereas loss-of-function mutations lead to intestinal obstruction and meconium ileus. Gain-of-function mutations in STAT1 cause an IPEX-like syndrome with enteropathy, whereas loss-of-function mutations are found in patients with autosomal dominant chronic mucocutaneous candidiasis. Loss of TTC7A activity results in multiple intestinal atresia and SCID, whereas hypomorphic mutations cause VEOIBD. Similarly, loss-of-function variants cause classic SCID defects, whereas hypomorphic variants in the same genes allow residual oligoclonal T-cell activation and are associated with immunopathology, including colitis.

Performing next-generation sequencing exome-wide or genome-wide will identify (in each patient) genetic variants of unknown relevance and, in some patients, known variants that are associated with incomplete penetrance or variable phenotype severity. Increasing use of DNA sequencing technologies will lead to detection of hypomorphic variants that cause milder phenotypes and/or later onset of IBD. The increased availability of genotype-phenotype data sets in databases such as ClinVar (http://www.ncbi.nlm.nih.gov/clinvar) or commercial databases will increase our ability to differentiate variants that cause IBD from those without biological effects. WES analysis of patients with pediatric onset of IBD, including VEOIBD, has revealed multiple rare genetic variants in those IBD susceptibility genes that were discovered by association studies. Similarly, WES analysis of patients with genetically confirmed mevalonate kinase deficiency identified multiple variants in IBD-related genes outside of the MVK gene. It is currently not clear how strongly these rare variants influence the genetic susceptibility to IBD as additive or synergistic factors. In particular, in patients with nonconventional forms of IBD, the identification of variants of unknown relevance can lead to the therapeutic dilemma of whether to wait for the disease to progress or start early treatment. Because some of the disease-specific treatment options have potentially severe adverse effects, careful evaluation of genetic variants is required not only to validate sequence data and statistical association but to provide functional evidence that those variants cause disease.

Conclusion

Rare monogenic disorders that affect intestinal immune and epithelial function can lead to VEOIBD and severe phenotypes. These disorders are diagnosed based on clinical and genetic information. Accurate genetic diagnosis is required for assessing prognosis and proper treatment of patients. We summarized phenotypes and laboratory findings for more than 50 monogenic disorders and suggest a diagnostic strategy to identify these extremely rare diseases, which have large effects on patients and their families.
Supplementary Material
Note: To access the supplementary material accompanying this article, visit the online version of Gastroenterology at www.gastrojournal.org and at http://dx.doi.org/10.1053/j.gastro.2014.07.023.

References
73. Speckmann C, Ehl S. XIAP deficiency is a mendelian cause of late-onset IBD. Gut 2014;63:1031–1032.


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Conflicts of interest
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