Maintaining Intestinal Health: The Genetics and Immunology of Very Early Onset Inflammatory Bowel Disease

Judith R. Kelsen,¹ Robert N. Baldassano,¹ David Artis,² and Gregory F. Sonnenberg²

¹Division of Gastroenterology, Hepatology and Nutrition, Children’s Hospital of Philadelphia, Philadelphia, Pennsylvania; ²Jill Roberts Institute for Research in IBD, Joan and Sanford I. Weill Department of Medicine, Division of Gastroenterology and Hepatology, and Department of Microbiology and Immunology, Weill Cornell Medical College, Cornell University, New York, New York

SUMMARY

Very early onset inflammatory bowel disease (VEO-IBD) is a distinct form of IBD. Here, we review the current knowledge of the genetic and immunologic basis of VEO-IBD, which requires further investigation for improved and individualized care.

Inflammatory bowel disease (IBD) is a multifactorial disease caused by dysregulated immune responses to commensal or pathogenic microbes in the intestine, resulting in chronic intestinal inflammation. An emerging population of patients with IBD younger than 5 years of age represent a unique form of disease, termed very early onset IBD (VEO-IBD), which is phenotypically and genetically distinct from older-onset IBD. VEO-IBD is associated with increased disease severity, aggressive progression, and poor responsiveness to most conventional therapies. Further investigation into the causes and pathogenesis of VEO-IBD will help improve treatment strategies and may lead to a better understanding of the mechanisms that are essential to maintain intestinal health or provoke the development of targeted therapeutic strategies to limit intestinal inflammation and promote tissue repair. Here, we discuss the phenotypic nature of VEO-IBD, the recent identification of novel gene variants associated with disease, and functional immunologic studies interrogating the contribution of specific genetic variants to the development of chronic intestinal inflammation. (Cell Mol Gastroenterol Hepatol 2015;1:462–476; http://dx.doi.org/10.1016/j.jcmgh.2015.06.010)

Keywords: Inflammatory Bowel Disease; Very Early Onset Inflammatory Bowel Disease; Whole Exome Sequencing; Mucosal Immunology.

Inflammatory bowel disease (IBD), comprising Crohn’s disease, ulcerative colitis, and indeterminate colitis, is a multigenetic and environmentally triggered disease resulting in a dysregulated immune response to commensal or pathogenic microbes found in the gastrointestinal tract.¹–⁷ Patients with IBD exhibit local and systemic immune reactivity to various microbes, have significant alterations in the composition of intestinal commensal bacteria, and can become colonized with pathogenic or opportunistic bacteria.⁸–¹³ The multifactorial nature and environmental contribution to IBD is largely responsible for its increased incidence over the last several decades.¹⁴ Added to the complex nature of the disease, host genetics may play a more prominent role in some subpopulations, particularly in very young children (younger than 5 years of age) in whom the disease is termed very early-onset IBD (VEO-IBD).¹⁵,¹⁶ Although this is a heterogeneous population, including some children with mild disease,¹⁷ patients with VEO-IBD can present with a different disease phenotype, including extensive colonic involvement and more severe disease than older children and adults.¹⁷,²⁰,²¹ In addition, due to poor response to conventional therapies, severity of inflammation, and greater duration of disease, there are higher rates of morbidity in this population.²⁰,²²,²³ Because of the aggressive disease phenotype, early age of onset, and strong family history of disease, some measure of VEO-IBD is thought to be a monogenic disease, often involving genes associated with primary immunodeficiencies.¹⁸,²⁰,²⁴ This was elegantly demonstrated with the discovery that several IL10 (interleukin-10),²⁵ IL10RA (interleukin-10 receptor, α), and IL10RB (interleukin-10 receptor, β)²⁶ gene mutations were associated with a phenotype of severe perianal disease and colitis in infants with VEO-IBD. Additional underlying immunodeficiencies or genetic disorders may also present with an intestinal phenotype in

Abbreviations used in this paper: ADAM17, A disintegrin and metalloproteinase domain 17; CGD, chronic granulomatous disease; COL7A1, collagen, type VII, α1; CVID, common variable immunodeficiency; FOXP3, forkhead box protein 3; GUCY2, guanylate cyclase 2; GWAS, genomewide association studies; IBD, inflammatory bowel disease; IL, interleukin; ILC, innate lymphoid cells; ILCS, group 3 innate lymphoid cells; Iga, immunoglobulin A; IKBKG, inhibitor of κ light polypeptide gene enhancer in B cells, kinase of, γ; IFNB, interferon regulatory factor; IFN, interferon; IPEX, immune dysregulation, polyendocrinopathy, and enteropathy, X-linked; MHCII, major histocompatibility complex class II; NEMO, nuclear factor-κB essential modulator; RAG, recombination-activating gene; STAT, signal transducer and activator of transcription; TNF, tumor necrosis factor; Treg, regulatory T cell; TTC7A, tetratricopeptide repeat domain-containing protein 7A; VEO-IBD, very early onset inflammatory bowel disease; WASP, Wiskott-Aldrich syndrome protein; WES, whole exome sequencing; XIAP, X-linked inhibitor of apoptosis protein.

Most current article
© 2015 The Authors. Published by Elsevier Inc. on behalf of the AGA Institute. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/). 2352-345X
http://dx.doi.org/10.1016/j.jcmgh.2015.06.010
patients with VEO-IBD.\textsuperscript{20,24} These include, but are not limited to, common variable immunodeficiency (CVID), Wiskott-Aldrich syndrome (WAS), immunodysregulation, polyendocrinopathy, and enteropathy, X-linked (IPEX) syndrome, and chronic granulomatous disease (CGD).\textsuperscript{17,20,22,26}

Studying consanguinity and targeted genetic sequencing has been an extremely valuable approach to allow the identification and characterization of genetic variants associated with VEO-IBD. However, these approaches alone may not identify novel and rare gene variants. Recent advances in sequencing technology such as whole exome sequencing (WES) have broadened our understanding of the pathogenesis of VEO-IBD and resulted in further discoveries of genes and pathways associated with the disease.\textsuperscript{26-30} The genomic contribution of IBD has been extensively evaluated through genomewide association studies (GWAS), and over 163 IBD-associated risk loci\textsuperscript{31} have been identified.

Several genes located within the IBD-associated loci are critical for regulation of host defense, involving both the innate and adaptive immune responses toward microbes.\textsuperscript{31} However, GWAS studies have been primarily performed in adult-onset IBD and in children 10 years of age and older, whose disease is most frequently a polygenic complex disease. Furthermore, GWAS often do not capture rare variants, specifically those with a minor allele frequency of <5%. In contrast, a proportion of patients with VEO-IBD have a monogenic-driven disease or multigenic disease enriched with rare variants of the same or interacting immunologic pathways.\textsuperscript{32,23} Thus, as in the case of IL10RA and IL10RB defects, the development of intestinal inflammation in VEO-IBD patients can be the direct result of defective immune responses.\textsuperscript{33}

Although WES has revolutionized our ability to study rare variants and to determine the genetic basis of disease, understanding the relevance of the identified variants has remained challenging. The individual patient’s phenotype may be shaped by mode of inheritance, epigenetics, and gene-gene interaction. Environmental modifiers such as the intestinal microbiota, antibiotic exposure, infection, or diet also significantly impact the disease phenotype.\textsuperscript{17,26,32} Because of the clinical presentation, often of severe disease, together with the challenge of identifying the unique pathogenesis of the disease, there is currently no standard of care in the evaluation and treatment for VEO-IBD patients. Identifying the driving forces in patients with particularly severe early-onset disease may lead to group-specific therapeutic approaches. Here we discuss the clinical presentation of VEO-IBD patients, the identification of common gene variants associated with the disease, and functional studies that have demonstrated how these variants may contribute to dysregulated immunologic homeostasis in the intestine.

**Clinical Presentation of Very Early Onset Inflammatory Bowel Disease**

Pediatric IBD has increased in incidence and prevalence, and this phenomenon has included very young children.\textsuperscript{16,33,34} VEO-IBD remains relatively uncommon, approximately 6% to 15% of the pediatric IBD population is younger than 6 years old, and disease in the first year of life is rare.\textsuperscript{16,34} A subset of patients with VEO-IBD present with a phenotype that is distinct from older children and adults, including extensive colonic disease (pancolitis) in which it is frequently difficult to differentiate ulcerative colitis from Crohn’s disease, leading to a diagnosis of indeterminate colitis (Table 1).\textsuperscript{20,34} At diagnosis, patients with VEO-IBD are more commonly diagnosed with ulcerative colitis (35% to 59%) as compared to older onset IBD (children older than 6 years and adults) in which Crohn’s disease is more prevalent (55% to 60%). In contrast, approximately 30% to 35% of VEO-IBD patients are diagnosed with Crohn’s disease. Indeterminate colitis is also diagnosed more often in patients with VEO (11% to 22%) as compared to older onset IBD (4% to 10%).\textsuperscript{19,35-37}

Although formal guidelines or standards of care do not exist, disease evaluation of this population includes

| Table 1. Features of Very Early Onset and Older Onset Inflammatory Bowel Disease |
|---------------------------|---------------------------|
| **Feature**               | **VEO-IBD**               | **Older-Onset IBD** |
| **Disease distribution**  | Predominately colonic     | Ileocolonic         |
|                           | ileal involvement < 20%   | Less extensive disease at presentation |
|                           | Extensive disease at presentation |                        |
| **Disease classification**| CD: 30%–35%               | CD: 55%–60%         |
|                           | UC: 35%–59%               | UC: 40%–45%         |
|                           | IC: 11%–22%               | IC: 4%–10%          |
| **Positive family history**| 40%–50%                   | 10%–20%             |
| **Genetic contribution**  | Increased prevalence monogenic disorders < 2 years | Polygenic inheritance |
| **Surgical intervention** | ~71%                      | ~55%                |
| **Other**                 | Therapeutic response to conventional therapy: decreased |                        |
|                           | Consanguinity             |                     |

CD, Crohn’s disease; IC, indeterminate colitis; UC, ulcerative colitis.
### Table 2. Clinical Presentations and Laboratory Abnormalities of Very Early Onset Inflammatory Bowel Disease Patients

| Type                        | Gastrointestinal Manifestations                                  | Laboratory Abnormalities                                                                 | Pathology                                                                 |
|-----------------------------|-------------------------------------------------------------------|------------------------------------------------------------------------------------------|--------------------------------------------------------------------------|
| B- and T-cell development   | Microthrombocytopenia                                             | Decreased platelets                                                                      | Crohn-like inflammatory process: cobblestone appearance and inflammatory pseudo-polyps<sup>36</sup> |
| WAS (WASP)                  | Moderate-severe eczema                                            | Low marginal B cells, high transitional B cells, elevated IgA, low IgM                  | Ulcerative colitis-like appearance reported: extensive colitis with ulcerations<sup>39</sup> |
|                             | Recurrent or severe infections                                    | Lymphopenia usually present, progressive decline in T cells                             | Neutrophil and T<sub>1</sub>2 lymphocyte infiltration                    |
|                             | Colitis: bloody diarrhea                                          | CD4:CD8 ratio is normal.                                                                 | Lack of plasma cells in lamina propria                                    |
| Hypogammaglobulinemia,      | Diarrhea                                                          | T cells or normal or increased                                                          |                                                                         |
| X-lined (BTK) or AR         | Chronic infectious diarrhea (<i>Giardia lamblia</i>, <i>Salmonella species</i>, <i>Escherichia coli</i>) | CD4: CD8 ratio is normal or decreased                                                   |                                                                         |
|                             | Crohn’s disease appearance                                       | Low IgG, IgA                                                                             |                                                                         |
|                             | Perirectal abscess and fistula                                     | Impaired antibody response                                                               |                                                                         |
|                             |                                                                   | Hypogammaglobulinemia<sup>40</sup>                                                      |                                                                         |
| Hyper IgM (CD40L, CD40,     | Diarrhea, sclerosing cholangitis                                  | Small bowel and colon: acute and chronic inflammation can be seen<sup>41</sup>          |                                                                         |
| AICDA, UNG)                 |                                                                   | Hyper IgE syndrome (STAT3, DOCK8)                                                       | Histology and crypt destruction pattern most resembles infectious agent<sup>42,43</sup> |
| LRBA deficiency             | Abdominal pain, diarrhea                                          | Extremely elevated IgE levels                                                           |                                                                         |
|                             | Hypoalbuminemia may be present                                   | Low IgM                                                                                  |                                                                         |
|                             | Susceptibility to infection (staphylococcal infection)            | Decreased memory B cells                                                                |                                                                         |
|                             | Atopic dermatitis                                                 | Neutropenia can be present                                                              |                                                                         |
|                             | Gastrointestinal manifestations most often secondary to infection | T cell lymphopenia                                                                       |                                                                         |
| Severe combined immunodeficiency (ZAP70, ITK, LCK) | Severe recurrent infection in infancy | B and T cells are decreased in the peripheral blood                                      | Hypocellular lamina propria lacking plasma cells or lymphocytes          |
|                             | Chronic diarrhea, malabsorption, and failure to thrive            | T cells may have immature phenotype                                                     | Graft versus host disease-like process in colon or small bowel can be present |
|                             | Oral and esophageal candidiasis                                   | Variants of severe combined immunodeficiency can have less severe lymphopenia          |                                                                         |
|                             |                                                                   | B cells (CD19-HLA-DR) may be normal or decreased                                       |                                                                         |
|                             |                                                                   | Defective IgG, IgA, normal IgM                                                         |                                                                         |
|                             |                                                                   | Neutropenia                                                                              |                                                                         |
|                             |                                                                   | Absence of T cells and natural killer cells                                             |                                                                         |
|                             |                                                                   | Normal B cells                                                                           |                                                                         |
|                             |                                                                   | Hypereosinophilia and increased IgE                                                     |                                                                         |
|                             |                                                                   | Can have oligoclonal T cells<sup>44</sup> and reduced B cells<sup>45</sup>             |                                                                         |
|                             |                                                                   | Same                                                                                    |                                                                         |
| X-linked severe combined    | Same                                                              | Same                                                                                    |                                                                         |
| immunodeficiency (IL2RG)    |                                                                   |                                                                         |                                                                         |
| Omenn syndrome (RAG1, RAG2, | Diffuse erythroderma                                              | Graft versus host disease: numerous apoptotic crypts cells in colon                    |                                                                         |
| Artemis, IL7Ra, LIG4, ADA,  | Hepatosplenomegaly                                                | Increase in lamina propria eosinophils<sup>46</sup>                                     |                                                                         |
| CHD7)                      | Lymphadenopathy                                                   |                                                                         |                                                                         |
|                             | Chronic diarrhea                                                  |                                                                         |                                                                         |
|                             | Failure to thrive                                                 |                                                                         |                                                                         |
| Type                                                                 | Gastrointestinal Manifestations                                                                 | Laboratory Abnormalities                                                                 | Pathology                                                                 |
|----------------------------------------------------------------------|-------------------------------------------------------------------------------------------------|----------------------------------------------------------------------------------------|---------------------------------------------------------------------------|
| Common variable immunodeficiency (TAC1, ICOS, CD19, CD20, CD21, CD81)| Heterogeneous presentation Recurrent bacterial infection Chronic infectious diarrhea Helicobacter pylori infection common: atrophic gastritis and pernicious anemia Malabsorption Granulomatous enteropathy | Decreased memory B cells Defective plasma C (CD27⁺, CD19, HLA-DR) T cells (CD2, CD3) usually quantitatively normal, T cell abnormalities common CD4:CD8 can be normal or decreased | Antrum: nonspecific increase in lamina propria lymphocytes; apoptotic cells Helicobacter pylori positive: atrophic gastritis Small intestine: villos atrophy, increased epithelial lymphocytosis Plasma cells absent or decreased |
| Epithelial defects                                                   | Present in neonatal period with watery diarrhea that progresses to bloody diarrhea                | Neonatal electrolyte disturbances B-cell activation defects, can have hypogammaglobulinemia Natural killer cell abnormalities Can have low immunoglobulin levels Defect in vaccine response | Hypoplastic crypts in small bowel Enterocolitis with villos atrophy and epithelial shedding |
| Familial diarrhea (GUCY2C) X-lined ectodermal dysplasia and immunodeficiency (IKBKG) | Severe diarrhea Colitis presentation | Defect in vaccine response | Crohn’s disease-like appearance Enterocolitis with villos atrophy and epithelial shedding |
| TTC7A deficiency                                                     | Blistering skin defects and hyperkeratosis of palms and soles                                   | Can have eosinophilia                                                                | Small bowel: villos atrophy Colon: apoptosis Intestinal epithelium: focal detachment of epithelium Ulcerations Colon: apoptosis Severe colitis |
| Klinder syndrome (FERMT1)                                            | Bloody diarrhea                                                                                  |                                                                                        |                                                                           |
| Dystrophic epidermolysis bullosa (COL7A1)                            |                                                                                                 |                                                                                        |                                                                           |
| Phagocyte defects                                                    | Recurrent infection/abscesses Perianal disease/fistula Esophageal and gastric outlet obstruction Colitis | Abnormal respiratory burst | Transmural and discontinuous inflammation, with aphthous or serpiginous ulcers can be seen, can be indistinguishable from Crohn’s disease Intestinal granulomas, increased compared to Crohn’s disease Crohn’s disease-like lesions in the small bowel Fistulas, longitudinal ulcers, stenosis Ulcerative colitis appearance Pigmented macrophages Perianal disease and oral manifestations may appear Colonic disease, adhesions, and strictures may be present |
| Chronic granulomatous disease (CYBB: x-linked, CYBA, NCF1, NCF2, NCF4, RAC1) |                                                                                                 |                                                                                        |                                                                           |
| Glycogen storage disease type 1 (SLC37A4)                           | Crohn’s disease presentation Perianal disease and oral manifestations Defective chemotaxis, phagocytosis, and bacterial killing | Neutropenia Neutrophil dysfunction Increased peripheral granulocytes |                                                                         |
| Leukocyte-adhesion deficiency (ITGB2)                               |                                                                                                 |                                                                                        |                                                                           |
| Genetic variants in the IL-10/IL-10 pathway and regulatory T cells IL-10 ligand and IL10RA and IL10RB | Most frequently neonatal onset of disease, bloody diarrhea Perianal fistula Arthritis Abscesses | Abnormal phosphorylation of STAT3 mediated by IL-10 | Ileal and colonic inflammation, ulcerations, inflammatory infiltrates |
| Type                                                                 | Gastrointestinal Manifestations                                      | Laboratory Abnormalities                                      | Pathology                                                                                           |
|----------------------------------------------------------------------|---------------------------------------------------------------------|----------------------------------------------------------------|-----------------------------------------------------------------------------------------------------|
| X-linked immune dysregulation, polyendocrinopathy, enteropathy (FOXP3, STAT1) | Polyendocrinopathy, Recurrent infection, Severe enteropathy with watery diarrhea, can be bloody | Lack of CD4<sup>+</sup>, CD25<sup>+</sup>, FOXP3<sup>+</sup> regulatory T cells, Elevated IgA and IgE, Normal B cell numbers, Eosinophilia | Extensive villous atrophy, Features of graft versus host disease with apoptosis of epithelial cells, Presence of antienterocyte antibodies along brush border of duodenum<sup>53</sup>, Lymphocytic, neutrophilic and eosinophilic infiltration of crypts and crypt abscesses<sup>53</sup>, Colitis |
| Hyperimmune or autoinflammatory Mevalonate kinase deficiency (MVK)   | Diarrhea, Abdominal pain, emesis, Recurrent fevers                    | Hyperimmunoglobulinemia D, Natural killer cell dysfunction, Elevation of inflammatory markers erythrocyte sedimentation rate, C-reactive protein, Elevated white blood cell count and inflammatory markers | Colon: ulcers, cellular infiltrate, apoptosis<sup>54</sup>, Small bowel inflammation<sup>55</sup>, Changes similar to Crohn’s disease with chronic inflammation and ulceration may appear<sup>56</sup> |
| Familial Mediterranean fever (MEFV)                                 | Recurrent fevers, diarrhea, Abdominal pain, Joint pain, Amyloidosis, Vasculitis, Peritonitis, Recurrent oral aphthae |                                                                 |                                                                                                     |
| X-linked lymphoproliferative syndrome 1 (XLP1) (SH2D1A), defective SLAM | Epstein-Barr virus triggered hemophagocytic lymphohistiocytosis        | Hypogammaglobulinemia, Poor antibody response                   | Unspecified colitis, may have absent plasma cells, Features similar to Crohn’s disease, Apoptosis can be seen |
| X-linked lymphoproliferative syndrome 2 (XLP2, XIAP)                 | Epstein-Barr virus triggered hemophagocytic lymphohistiocytosis, Clinical features similar to observed in Crohn’s disease | Hypogammaglobulinemia, Decreased natural killer cells can be present |                                                                                                     |
| Hermansky-Pudlak syndrome (IBD phenotype involvement: HPS1, HSP4, HSP6) | Oculocutaneous albinism, Easy bruising, Inflammatory bowel disease symptoms of abdominal pain, diarrhea, bloody can be present | Normal platelet count, Prolonged bleeding time, Platelet dysfunction | Broad ulcers, Brown granular pigmentation, Nonnecrotizing granulomas |
laboratory, radiologic, and endoscopic evaluation (Table 2). A diagnosis at a very young age should trigger concern for a monogenic-driven disease, particularly in IBD diagnosed when the patient is younger than 2 years of age. Furthermore, extensive family history, including a history of disease in male family members (such as in X-linked disease) or a history of infection, skin disease, or autoimmunity can help guide the appropriate laboratory screening. The laboratory studies should include not only the routine screening used for IBD diagnosis but also an immunologic evaluation. This includes vaccine titers, immunoglobulin profiles, analyses of B- and T-cell function, and a dihydro-rhodamine flow cytometry assay, and if necessary more targeted phenotyping and functional profiling of the systemic and mucosal immune system. As shown in Table 2, these studies may point to an underlying defect such as neutropenia, which may represent a monogenic disorder causing functional defects in neutrophils such as glycogen storage disease type 1b, leucocyte adhesion deficiency, or congenital neutropenia.

**Genetic Variants Associated With VEO-IBD and Their Immunologic Consequences**

Monogenic diseases that can present with the phenotype of intestinal inflammation include those that cause defects of intestinal epithelial barrier function, phagocyte bacterial killing, development and function of the adaptive immune system, and hyper- or autoimmune-inflammatory disorders. These genetic alterations may differentially influence the development and progression of intestinal inflammation, so these patients will likely exhibit significant heterogeneity in their responsiveness to therapeutic interventions. We will discuss what we have learned from mouse models and translational patient-based studies, which should be considered when developing therapeutic strategies for these unique patient populations.

**Genetic Variants Influencing Intestinal Epithelial Barrier Function**

Mutations in genes associated with maintaining integrity of the intestinal epithelial barrier can present with intestinal inflammation in patients with VEO-IBD. These include loss-of-function mutations in **ADAM17** (A disintegrin and metalloproteinase domain 17) resulting in **ADAM17 deficiency,**57,58 in **IKBKG** (inhibitor of κ light polypeptide gene enhancer in B cells, kinase of; γ; encoding nuclear factor-κB essential modulator: NEMO) resulting in X-linked ectodermal dysplasia and immunodeficiency,**29** in **COL7A1** (collagen, type VII, α1) resulting in dystrophic epidermolysis bullosa,**60** **FERMT1** (Fermitin family homolog 1) resulting in Kindler syndrome,**61–63** and **TTC7A** (tetracopeptide repeat domain-containing protein 7A) or gain-of-function mutations in **GUCY2** (guanylate cyclase 2) resulting in familial diarrhea.**17,64** Mutations in these genes may all lead to an impairment of the intestinal epithelial barrier through distinct pathways, such as limiting epithelial regeneration (**ADAM17**),**65** loss of signaling pathways involved in gene expression (**IKBKG**),**66,67** altered cell adhesion, barrier formation, apoptosis (**COL7A1, FERMT1,** and **TTC7A**),**29,60–63** or impaired bacterial sensing and ion homeostasis (**GUCY2**).**17,64** The intestinal histology of patients with epithelial defects can be helpful in distinguishing the disease from other causes of intestinal inflammation. For example, patients with **IKBKG** (NEMO) defects may have villous atrophy or epithelial cell shedding on pathologic examination.**66** In contrast, histology in patients with **ADAM17** mutations may demonstrate hypoplastic crypts in the small bowel secondary to a low rate of epithelial production, as **ADAM17** is necessary for transforming growth factor-α to be cleaved from the cell membrane.**69,70**

The intestinal barrier is necessary to maintain a physical separation between commensal bacteria and the mammalian immune system, and a breakdown in this barrier through multiple distinct pathways can directly promote chronic intestinal inflammation.**1,4** In addition to the genes we have listed, intestinal barrier function is maintained through a number of physical and biochemical structures, including mucus production, intestinal epithelial cell tight junction proteins, immunoglobulin A (IgA), and antimicrobial peptides (Figure 1). In mice, chemical disruption of the intestinal barrier through administration of dextran sodium sulfate in drinking water results in dissemination of commensal bacteria and activation of the innate immune system.**71** Chronic exposure to dextran sodium sulfate can lead to activation of the adaptive immune system and to the development of proinflammatory, commensal bacteria-specific B- and T-cell responses,**8,72** which are similar to those observed in IBD patients.**8,73**

Intestinal epithelial cells play an important role in directly regulating immunologic homeostasis in the intestine, as mice with intestinal epithelial cell lineage–specific deletion of factors regulating the nuclear factor-κB pathway, including NEMO and IκB kinase-β, have an increased susceptibility to develop chronic intestinal inflammation.**66,67** Although we know that loss of intestinal barrier function can directly cause intestinal inflammation, additional mouse models and translational patient-based approaches are required to further define how mutations in these genes specifically lead to a breakdown in the barrier, and whether we can develop more targeted therapies to restore barrier integrity and limit chronic inflammation.

**Genetic Variants Impairing Development of the Adaptive Immune System**

Several genetic variants can alter the development or function of adaptive immune cells in a cell intrinsic or extrinsic manner.**32** Defects that affect development or function of B cells and T cells occur with loss-of-function mutations in recombination-activating genes (**RAG1** or **RAG2**) or the IL-7R (**IL7R**) causing Omenn syndrome, or the **PTEN** (phosphatase and tensin homolog) gene causing **PTEN** syndrome. Defects in **RAG1, RAG2,** or **IL-7R** can cause cell-intrinsic defects in the development of both T cells and B cells by blocking either early lymphocyte survival or
Defects in B-cell development lead to an absence of circulating mature B cells and antibody production, which have been linked to an IBD phenotype.\textsuperscript{77} This includes agammaglobulinemia, which can also occur in X-linked agammaglobulinemia\textsuperscript{78} and CVID, a complex and heterogeneous disease, with the responsible mutations known for only a minority of cases.\textsuperscript{79} Recently, one candidate gene causing CVID and potentially contributing to intestinal inflammation was identified using WES as a loss-of-function mutation in \textit{LRBA} (lipopolysaccharide-responsive beige-like anchor), resulting in multiple defects in immune cell populations.\textsuperscript{80}

Related to CVID, antibody deficiencies associated with IBD manifestations include IgA deficiency and severe combined immunodeficiency, which can be secondary to multiple variants that influence the development or function of the adaptive immune system (including \textit{RAG1}, \textit{RAG2}, \textit{JAK3}, \textit{CD45}, \textit{CD3G}, \textit{ZAP70}, \textit{ADA}, \textit{DCLRE1C}).\textsuperscript{32,77,80} Omenn syndrome, a recessive form of severe combined immunodeficiency, involves abnormal development of B cells and T cells, and can also be associated with intestinal disease as well as severe eczematous rash.\textsuperscript{80,81} In these patients, laboratory studies indicate increased oligoclonal T cells and reduced B cells, and histologic examination can show an intestinal graft versus host appearance.\textsuperscript{82,83} Conversely, overproduction of specific immunoglobulins, such as hyper IgM, hyper IgE syndrome (resulting from a loss of function mutation in \textit{DOCK8}) can also result in intestinal inflammation and an IBD phenotype.\textsuperscript{84}

It is currently unclear exactly how these selective impairments of the adaptive immune system can manifest in intestinal inflammation. There is a potential involvement of altered regulatory pathways or chronic infections with pathogenic and opportunistic microbes. Therefore, additional lines of study are required to further interrogate the link of these mutations to intestinal inflammation.

Wiskott-Aldrich syndrome results from a loss-of-function mutation in the Wiskott-Aldrich syndrome protein (\textit{WASP}), and patients can exhibit thrombocytopenia, eczema, immune deficiencies, and intestinal inflammation.\textsuperscript{85} The clinical manifestation of VEO-IBD patients with this genetic defect can be pancolitis in addition to other autoimmune processes. \textit{WASP} is a critical cytoskeleton protein expressed in hematopoietic cells, and it is required for the normal development and function of multiple cell types.\textsuperscript{86,87} \textit{WASP} is critical for peripheral B-cell development and function and thus the ability to respond to antigens.\textsuperscript{88,89} Laboratory studies in these patients may show thrombocytopenia, low IgM levels, low marginal B cells, and lymphopenia.\textsuperscript{90} Nguyen et al\textsuperscript{91} identified that intestinal inflammation in \textit{WASP}-deficient mice was critically dependent upon inflammatory T cells, which may result from impaired development of regulatory T cells (Tregs) in the thymus and periphery.\textsuperscript{92} Surprisingly, these defects are likely occurring in a cell-extrinsic manner, as the absence of \textit{WASP} in cells of the innate immune system directly contributed to the development of inflammatory T-cell responses in mice.\textsuperscript{93}

The causes of intestinal inflammation in other similar patient populations are less well understood, but defects in regulatory T cells, IgA, and abnormal selection of T-cell and B-cell specificities likely contribute. Additional immunologic analyses and mouse models, such as those we have described, are needed to define the causes of disease further and to develop potential therapeutic options in these patient populations.

\textbf{Genetic Variants Impairing Regulatory T Cells}

Defects in regulatory T cells can clinically present as colonic disease as well as an enteropathy. IPEX syndrome is most often secondary to mutations of the forkhead box protein 3 (\textit{FOXP3}) gene, a transcription factor that is essential for the development and immunosuppressive
activity of CD4 Foxp3+ Tregs.94,95 There are over 20 mutations in FOXP3 that have been identified in patients with IPEX,96 and patients frequently present with neonatal severe secretory diarrhea, failure to thrive, infection (due to defects in immunoregulation), skin rash, insulin-dependent diabetes, thyroiditis, cytopenias, and other autoimmune disorders.94,95 Tregs are absent or dysfunctional in these patients, and in the intestine histologic analyses may reveal infiltration of inflammatory cells in the lamina propria and submucosa of the small bowel and colon as well as changes in the mucosa of the small bowel.94 Other genetic defects have been found to cause IPEX-like disease, including loss-of-function mutations impacting IL-2/IL-2R interactions, STAT5b (signal transducer and activator of transcription 5b), and ITCH (itchy E3 ubiquitin protein ligase), or gain-of-function mutations in STAT1, all of which critically influence the development and function of Tregs.81 Further, Zeissig et al97 have recently identified in VEO-IBD a novel loss of function mutation in CTLA4 (cytotoxic T lymphocyte-associated protein 4), a surface molecule of regulatory T cells that directly suppresses effector T-cell populations.

The mechanisms by which regulatory T cells limit intestinal inflammation are well characterized in mice. Tregs can develop in the thymus as “natural Tregs” and directly contribute to limiting proinflammatory T cells in the intestine.98 The composition of commensal bacteria influences the repertoire of Tregs,98 and commensal bacteria-specific “induced Tregs” can also be generated in the periphery after sampling of commensal bacteria by dendritic cells in the intestine and migration to the mesenteric lymph node (Figure 2).1,5,99,100 Once generated, Tregs can then promote intestinal homeostasis through direct regulation of innate and adaptive immune cell responses to commensal bacteria, a process that involves cytokine production, direct cell-cell contact (in part through CTLA4), and sequestering of growth factors.1,5,99

Consistent with a major role for Tregs in limiting proinflammatory immune cell responses to commensal bacteria, mice deficient in IL-2 or FoxP3 develop significantly less intestinal inflammation when maintained in germ-free versus conventional housing conditions, but they exhibit comparable levels of systemic autoimmunity.101,102 Recent evidence also suggests that the balance of tissue-specific IL-23 and IL-33 expression in mice is critical for regulating the function of Tregs in the intestine and chronic inflammation,103 although the role of these pathways in human VEO-IBD has not been examined.

Figure 2. Regulatory T cells, interleukin-10 (IL-10) and ILC3 critically limit dysregulated immune responses to commensal bacteria. Regulatory T cells (Treg) can differentiate in the thymus or periphery and limit immune cell responses to intestinal commensal bacteria through multiple mechanisms, including cytokine production, direct cell-to-cell contact and sequestering of growth factors. Further, IL-10 production by multiple cell types can directly promote anti-inflammatory responses from myeloid cells to limit intestinal inflammation. ILC3 can also directly limit proinflammatory T cells through MHCII-dependent interactions. iTreg, inducible regulatory T cell; nTreg, natural regulatory T cell. Identified genetic variants that result in a loss or gain of function mutation and are associated with very early onset inflammatory bowel disease are noted in orange boxes.
Genetic Variants in the Interleukin-10/Interleukin-10 Receptor Pathway and Related Cytokine Family Members

Homozygous loss of function mutations in IL10 ligand and receptors IL10RA and IL10RB are associated with significant intestinal inflammation, particularly in neonatal or infantile VEO-IBD, with a phenotype of severe enterocolitis and perianal disease.112 In addition, compound heterozygote loss of function mutations of IL10RA have been reported with neonatal Crohn’s disease and enterocolitis.104 IL-10 is an anti-inflammatory cytokine secreted by a variety of cells, including dendritic cells, natural killer (NK) cells, eosinophils, mast cells, macrophages, B cells and CD4+ T cell subsets (including Th2 cells, Th1 cells, Th17 cells and Treg).105,106 IL-10 maintains homeostasis through suppression of an excessive proinflammatory response and exerts its effect through binding to the IL-10 receptor, IL-10R, which is a tetrameric complex.107 It is composed of 2 distinct chains, 2 molecules of IL-10R1 (α chain) and 2 molecules of IL-10R2 (β chain).108 IL-10 binding to IL-10R activates the JAK1/STAT3 cascade, which subsequently limits proinflammatory gene expression.109 In addition to intestinal inflammation, IL-10 defects are associated with arthritis, folliculitis, and a predisposition to lymphoma.104,109 Given that the defects in IL-10-IL-10R interactions predominantly influence the immune system, a potential treatment for these patients is successful hematopoietic cell transplantation.110 Although this can be challenging and typically requires an HLA-identical donor, there has been recent success reported with haploidentical stem cell transplantation, however nonengraftment complications can occur.111

An essential role for IL-10 in limiting intestinal inflammation was demonstrated when IL-10 deficient mice were generated and found develop severe spontaneous colitis,112 and subsequent studies by Sartor et al113 identified that the intestinal inflammation in IL-10-deficient mice was entirely dependent upon the presence of commensal bacteria. Therefore, IL-10 plays a critical role in limiting dysregulated immune cell responses to intestinal commensal bacteria (Figure 2). The exact cellular sources and targets of IL-10 that contribute to the maintenance of intestinal homeostasis have been less well defined until the recent development of mice that permit conditional deletion of IL-10 and IL-10R. These critical studies have revealed an essential role of regulatory T cell-intrinsic IL-10 expression in preventing intestinal inflammation in mice.114,115 Further, it was recently demonstrated that IL-10R expression on myeloid cells in mice is critical to elicit anti-inflammatory responses and limit T cell-dependent intestinal inflammation.116,117 Critically, patients with loss-of-function mutations in IL10RA or IL10RB also exhibited an impaired ability to differentiate anti-inflammatory myeloid cells in vitro, and rather exhibited increased proinflammatory properties, such as elevated expression of IL-6, IL-12, tumor necrosis factor-α (TNFα), MHCII (major histocompatibility complex class II), and costimulatory molecules.118 Although mouse models have provided invaluable insight into human health and disease, it should be noted that mice deficient in IL10 do not completely replicate the phenotypes of humans with loss-of-function mutations in IL10, likely due to many confounding factors.

IL-22 is a cytokine that is related to IL-10, shares the IL-10R2 chain with a unique IL-22R1, signals through predominantly STAT3, and also plays a critical role in mediating intestinal homeostasis.110 However, unlike IL-10, the functional IL-22R is restricted to predominantly nonhematopoietic cells; in the intestine, IL-22 acts almost exclusively on intestinal epithelial cells to mediate innate immunity and intestinal barrier function (Figure 1).110 IL-22 can be produced by Th17 cells, and more recently has been identified to be predominantly expressed by a previously unrecognized cell type of the innate immune system, termed group 3 innate lymphoid cells (ILC3).118,119 This breakthrough in immunology has led to the identification of other members of the innate lymphoid cell (ILC) family, including group 1 ILCs that express T-bet and proinflammatory cytokines TNFα and interferon-γ, and group 2 ILCs that express GATA3 and type 2 cytokines IL-4, IL-5, IL-9, IL-13, and amphiregulin.119,120

The ILC family exhibits a heterogeneity comparable to that of differentiated CD4 T-cell subsets, and plays a profound role in regulating intestinal health and disease in mouse models.118-120 Critically, recent reports suggest that ILC3 is a dominant source of IL-22 in the intestine of healthy humans, and that dysregulated ILC responses are observed in adult patients with IBD.121-127 Further, we have also recently identified that ILC3 expresses MHCII, and that selective deletion of MHCII on ILC3 results in dysregulated CD4 T-cell responses and spontaneous intestinal inflammation.122 MHCII+ ILC3 selectively induces cell death of proinflammatory, commensal bacteria-specific CD4 T cells in the intestine; critically, we observed that MHCII was reduced on ILC3 from intestinal biopsy tissues of pediatric IBD patients versus non-IBD controls, and that this was inversely correlated with the level of proinflammatory TNf17 cells.126

Despite these advances, ILC and IL-22 responses have yet to be explored in VEO-IBD. Given the importance of these pathways in mediating intestinal health and disease, it is likely that the genetic variations associated with VEO-IBD, such as IL7/IL7R, may differentially influence ILC responses.

Genetic Variants Influencing Bacterial Recognition and Clearance

Chronic granulomatous disease (CGD) is a result of defective intestinal phagocytes, specifically the granulocytes responsible for bacterial killing and clearance.129 The NADPH oxidase complex is responsible for killing ingested microbes through its production of the respiratory burst. Mutations in any part of the complex molecules (CYBB, CYBA, NCF1, NCF2, NCF4) can result in intestinal inflammation as well as autoimmune disease.130,131 Intestinal inflammation can be observed in as many as 40% of patients with CGD.129,132-134

Several variants have been associated with VEO-IBD, in particular defective NCF2 (neutrophil cytosolic factor 2)
results in altered binding to RAC2 (ras-related C3 botulinum toxin substrate 2). These patients can present in the neonatal or first year of life with colitis, severe fistulizing perianal disease, and stricturing. Histology frequently demonstrates multiple granulomas that may not have associated inflammatory change. Critically, a recent study by Dhillon et al identified that heterozygous loss-of-function mutations in components of the NADPH oxidase complex can determine susceptibility to VEO-IBD, without directly causing overt immunodeficiency. Other neutrophil defects that are associated with VEO-IBD include leukocyte-attachment deficiency due to mutation in ITGB2 (integrin, β2). These patients can present with an IBD phenotype, history of bacterial infection, and laboratory studies remarkable for increased peripheral granulocytes. Glycogen storage disease type 1b, with hallmark features of neutropenia and neutrophil granulocyte dysfunction, can present with intestinal inflammation.

The reasons why CGD and other bacterial processing defects may manifest in intestinal inflammation are poorly understood and warrant additional research. It is possible that the causes include bacterial overgrowth or dysbiosis in the intestine, dysregulated activation of the innate and adaptive immune system, or both. Further, the therapies used to treat such patients need to be carefully considered. For example, anti-TNFα therapy is contraindicated in CGD; although it is effective for intestinal disease, it can increase the risk of severe infections in these patients. Other therapies include leukine, antibiotics, and allogenic hematopoietic stem cell transplantation, which have demonstrated some success. Recent evidence suggests that IL-1R antagonists may be a particularly attractive approach to limit disease in mouse models and patients with CGD by restoring autophagy and directly limiting inflammation.

Hyper- and Autoimmune-Disorders

Several autoimmune diseases have been linked to intestinal inflammation in children with VEO-IBD. These include mevalonate-kinase deficiency, familial Mediterranean fever (FMF), Hermansky-Pudlak syndrome, and X-linked lymphoproliferative syndrome (types 1 and 2). These diseases occur due to loss-of-function mutations in an enzyme critical for metabolism (mevalonate-kinase deficiency), cytoskeletal proteins (familial Mediterranean fever), proteins involved in organelle fusion or biogenesis (Hermansky-Pudlak syndrome), or proteins involved in cell signaling or apoptosis (X-linked lymphoproliferative syndrome). Although there are many additional clinical manifestations in these patients, 20% of patients with X-linked lymphoproliferative syndrome that have a loss-of-function defect in the gene X-linked inhibitor of apoptosis protein (XIAP), present with VEO-IBD.

XIAP is involved in nucleotide-binding oligomerization domain-containing protein 2 (NOD2)-mediated nuclear factor-κB signaling, so these children may have an impaired ability to sense bacteria. In addition, as an inhibitor of apoptosis, it prevents apoptosis of activated T cells, thus allowing for expansion and survival of T cells in response to pathogens. Therefore, in XIAP deficiency, due to the inability to clear pathogens, there is a hyperinflammatory state with increased production of cytokines resulting in an IBD phenotype. Children with these mutations can present with severe colonic and perianal fistulizing disease; of great concern, Epstein-Barr virus infection can result in fatal hemophagocytic lymphohistiocytosis.

This was not an exhaustive description of the rare genomic drivers of VEO-IBD, but it highlights the different components of the immune system, including innate and adaptive responses, involved in this disease. Treatments guided toward the specific defect, such as IL-1 antagonists, colchicine, hematopoietic stem-cell transplantation, or leukine can be used if the defect is determined. Additionally, monitoring for potential complications associated with a genetic defect is essential, such as in XIAP, IL-10 gene variants, and CGD. In addition to these monogenic diseases, VEO-IBD has been shown to have a high degree of genetic heterogeneity. It is therefore likely that there are more pathways involved in VEO-IBD, and the outcome of therapeutic intervention can be improved through further study and identification of the associated variants. Using next-generation sequencing technology such as WES can improve the detection of variants and the diagnosis of disease. Further, there is an urgent need to also directly translate genes to function and to functionally profile the immunologic significance of known genetic variations in intestinal inflammation.

Perspective and Future Directions in Genetic and Immunologic Analyses of Very Early Onset Inflammatory Bowel Disease

To advance our understanding of VEO-IBD, new sequencing technology must be used to completely survey the genetic landscape of this disease. Immunologic studies spanning basic mouse models and translational patient-based approaches are required to determine the contribution of those genetic variations to human disease. Given that dysregulated interactions between the immune system and commensal bacteria underlie the pathogenesis of intestinal inflammation, it is also important to include analyses of the composition and function of the microbiome. Given that these patient populations are studied worldwide and sometimes in small numbers, an international registry containing the genetic, immunologic, and environmental data of VEO-IBD patients could prove beneficial in our goal of better understanding the effects of different variants within known genes and identifying new gene defects that cause IBD through the study of mutations that arise in the same genes of multiple unrelated individuals. With an increased understanding of the disease processes operating in VEO-IBD, we can begin to individualize therapies to the specific patient or patient groups, as well as employ unconventional therapies that are not routinely part of the IBD therapeutic arsenal. These approaches could provide a
roadmap to establishing a standard of care for this disease and improving patient quality of life.

References
1. Maloy KJ, Powrie F. Intestinal homeostasis and its breakdown in inflammatory bowel disease. Nature 2011; 474:298–306.
2. Maynard CL, Elson CO, Hatton RD, et al. Reciprocal interactions of the intestinal microbiota and immune system. Nature 2012;489:231–241.
3. Hooper LV, Macpherson AJ. Immune adaptations that maintain homeostasis with the intestinal microbiota. Nat Rev Immunol 2010;10:159–169.
4. Hooper LV, Littman DR, Macpherson AJ. Interactions between the microbiota and the immune system. Science 2012;336:1268–1273.
5. Belkaid Y, Hand TW. Role of the microbiota in immunity and inflammation. Cell 2014;157:121–141.
6. Khor B, Gardet A, Xavier RJ. Genetics and pathogenesis of intestinal inflammation: implications for the understanding of inflammatory bowel disease. Gut 2006;55:1275–1282.
7. Abraham C, Cho JH. Inflammatory bowel disease. N Engl J Med 2009;361:2066–2078.
8. Lodès MJ, Cong Y, Elson CO, et al. Bacterial flagellin is a dominant antigen in Crohn disease. J Clin Invest 2004;113:1296–1306.
9. Baumgart M, Dogan B, Rishniw M, et al. High prevalence of adherent-invasive Escherichia coli associated with ileal mucosa in Crohn’s disease. Gastroenterology 2004;127:412–421.
10. Daiwadi H, Wei B, Kronenberg M, et al. The Crohn’s disease-associated bacterial protein I2 is a novel enteric T cell superantigen. Immunity 2001;15:149–158.
11. Wilting B, Halfvarson J, Dicksved J, et al. Studies reveal specific imbalances in the mucosa-associated microbiota of patients with ileal Crohn’s disease. Inflamm Bowel Dis 2009;15:653–660.
12. Willing B, Halfvarson J, Dicksved J, et al. A pyrosequencing study in twins shows that gastrointestinal microbial profiles vary with inflammatory bowel disease phenotypes. Gastroenterology 2010;139:1844–1854.e1.
13. Martin HM, Campbell BJ, Hart CA, et al. Enhanced Escherichia coli adherence and invasion in Crohn’s disease and colon cancer. Gastroenterology 2004;127:80–93.
14. Benchimol EI, Guttmann A, Griffiths AM, et al. Increasing incidence of paediatric inflammatory bowel disease in Ontario, Canada: evidence from health administrative data. Gut 2009;58:1490–1497.
15. El Khoury MG, Talbotec C, et al. Characteristic of inflammatory bowel disease with onset during the first year of life. J Pediatr Gastroenterol Nutr 2006;43:603–609.
16. Benchimol EI, Mack DR, Nguyen GC, et al. Incidence, outcomes, and health services burden of very early onset inflammatory bowel disease. Gastroenterology 2014;147:803–813.e7.
17. Uhlig HH. Monogenic diseases associated with intestinal inflammation: implications for the understanding of inflammatory bowel disease. Gut 2013;62:1795–1805.
18. de Ridder L, Weersma RK, Dijkstra G, et al. Genetic susceptibility has a more important role in pediatric-onset Crohn’s disease than in adult-onset Crohn’s disease. Inflamm Bowel Dis 2007;13:1083–1092.
19. Benchimol EI, Mack DR, Nguyen GC, et al. Characteristic of inflammatory bowel disease with onset during the first year of life. J Pediatr Gastroenterol Nutr 2006;43:603–609.
20. Daar E, Grimbacher B. Inflammatory bowel disease: is it a primary immunodeficiency? Cell Mol Life Sci 2012;69:41–48.
21. Ruemmele FM, El Khoury MG, Talbotec C, et al. Characteristic of inflammatory bowel disease with onset during the first year of life. J Pediatr Gastroenterol Nutr 2006;43:603–609.
22. Cannito Z, Berti I, Martelossi S, et al. IBD mimicking enterocolitis in children younger than 2 years of age. Eur J Pediatr 2009;168:149–155.
23. Glocker E, Kotlarz D, Boztug K, et al. Inflammatory bowel disease and mutations affecting the interleukin-10 receptor. N Engl J Med 2009;361:2033–2045.
24. Blank V, Broeckel U, Kugathasan S. Pediatric inflammatory bowel disease: clinical and molecular genetics. Inflamm Bowel Dis 2007;13:1430–1438.
25. Glocker E, Frede N, Perro M, et al. Infant colitis—it’s in the genes. Lancet 2010;376:1272.
26. Agarwal S, Mayer L. Diagnosis and treatment of gastrointestinal disorders in patients with primary immunodeficiency. Clin Gastroenterol Hepatol 2013;11:1050–1063.
27. Mao H, Yang W, Lee PP, et al. Exome sequencing identifies novel compound heterozygous mutations of IL-10 receptor 1 in neonatal-onset Crohn’s disease. Genes Immun 2012;13:437–442.
28. Worthey EA, Mayer AN, Siverson GD, et al. Making a definitive diagnosis: successful clinical application of whole exome sequencing in a child with intractable inflammatory bowel disease. Genet Med 2011;13:255–262.
29. Avitzur Y, Guo C, Mastropaolo LA, et al. Mutations in tetratricopeptide repeat domain 7A result in a severe phenotype. J Med Genet 2014;51:748–755.
30. Kammermeier J, Drury S, James CT, et al. Targeted gene panel sequencing in children with very early onset inflammatory bowel disease—evaluation and prospective analysis. J Med Genet 2014;51:748–755.
31. Ostovar R, Vancket S, Raskova RM, et al. Host-microbe interactions have shaped the genetic architecture of inflammatory bowel disease. Nature 2012;491:119–124.
32. Durandy A, Kracker S, Fischer A. Primary antibody deficiencies. Nat Rev Immunol 2013;13:519–533.
33. Muise AM, Snapper SB, Kugathasan S. The age of gene discovery in very early onset inflammatory bowel disease. Gastroenterology 2012;143:285–288.
34. Uhlig HH, Schwerd T, Koletzko S, et al. The diagnostic approach to monogenic very early onset
inflammatory bowel disease. Gastroenterology 2014; 147:990–1007.e3.

35. Heyman MB, Kirschner BS, Gold BD, et al. Children with early-onset inflammatory bowel disease (IBD): analysis of a pediatric IBD consortium registry. J Pediatr 2005; 146:35–40.

36. Mamula P, Telega GW, Markowitz JE, et al. Inflammatory bowel disease in children 5 years of age and younger. Am J Gastroenterol 2002;97:2005–2010.

37. Aloi M, Lionetti P, Barabino A, et al. Phenotype and disease course of early-onset pediatric inflammatory bowel disease. Inflamm Bowel Dis 2014;20:597–605.

38. Hsieh KH, Chang MH, Lee CY, Wang CY. Wiskott-Aldrich syndrome and inflammatory bowel disease. Ann Allergy 1988;60:429–431.

39. Marks DJ, Seymour CR, Sewell GW, et al. Inflammatory bowel diseases in patients with adaptive and complement immunodeficiency disorders. Inflamm Bowel Dis 2010;16:1984–1992.

40. Alangari A, Alsaltan A, Adly N, et al. LPS-responsive beige-like anchor (LRBA) gene mutation in a family with inflammatory bowel disease and combined immunodeficiency. J Allergy Clin Immunol 2012;130:481–488.e2.

41. Seidel MG. Autoimmune and other cytopenias in primary immunodeficiencies: pathomechanisms, novel differential diagnoses, and treatment. Blood 2014;124: 2337–2344.

42. Zhang Q, Davis JC, Lamborn IT, et al. Combined immunodeficiency associated with DOCK8 mutations. N Engl J Med 2009;361:2046–2055.

43. Randal KL, Lambe T, Johnson AL, et al. Dock8 mutations cripple B cell immunological synapses, germinal centers and long-lived antibody production. Nat Immunol 2009;10:1283–1291.

44. Brooks EG, Filipovich AH, Padgett JW, et al. T-cell receptor analysis in Omenn’s syndrome: evidence for defects in gene rearrangement and assembly. Blood 1999;93:242–250.

45. Kato M, Kimura H, Seki M, et al. Omenn syndrome—review of several phenotypes of Omenn syndrome and RAG1/RAG2 mutations in Japan. Allergol Int 2006; 55:115–119.

46. Schuetz C, Huck K, Gudowius S, et al. An immunodeficiency disease with RAG mutations and granulomas. N Engl J Med 2008;358:2030–2038.

47. Washington K, Stenzel TT, Buckley RH, et al. Gastrointestinal pathology in patients with common variable immunodeficiency and X-linked agammaglobulinemia. Am J Surg Pathol 1996;20:1240–1252.

48. Malamut G, Verkarre V, Suarez F, et al. The enteropathy associated with common variable immunodeficiency: the delineated frontiers with celiac disease. Am J Gastroenterol 2010;105:2262–2275.

49. Cunningham-Rundles C, Bodian C. Common variable immunodeficiency: clinical and immunological features of 248 patients. Clin Immunol 1999;92:34–48.

50. van Zelm MC, Reisli I, van der Burg M, et al. An antibody-deficiency syndrome due to mutations in the CD19 gene. N Engl J Med 2006;354:1901–1912.

51. Zonana J, Elder ME, Schneider LC, et al. A novel X-linked disorder of immune deficiency and hydropidrotic ectodermal dysplasia is allelic to incontinentia pigmenti and due to mutations in IKK-gamma (NEMO). Am J Hum Genet 2000;67:1555–1562.

52. Marks DJ, Miyagi K, Rahman FZ, et al. Inflammatory bowel disease in CGD reproduces the clinicopathological features of Crohn’s disease. Am J Gastroenterol 2009; 104:117–124.

53. Patey-Mariaud de Serre N, Canioni D, Ganousse S, et al. Digestive histopathological presentation of IPEX syndrome. Mod Pathol 2009;22:95–102.

54. Levy M, Aron A, Berrebi D, et al. Severe early-onset colitis revealing mevalonate kinase deficiency. Pediatrics 2013;132:e779–e783.

55. Demir A, Akyuz F, Gokturk S, et al. Small bowel mucosal damage in familial Mediterranean fever: results of capsule endoscopy screening. Scand J Gastroenterol 2014;49:1414–1418.

56. Gurkan OE, Yilmaz G, Aksu AU, et al. Colonic lymphoid nodular hyperplasia in childhood: causes of familial Mediterranean fever need extra attention. J Pediatr Gastroenterol Nutr 2013;57:817–821.

57. Chalaris A, Gewiese J, Paliga K, et al. ADAM17-mediated shedding of the IL6R induces cleavage of the membrane stub by gamma-secretase. Biochim Biophys Acta 2010; 1803:234–245.

58. Blaydon DC, Biancheri P, Di WL, et al. Inflammatory skin and bowel disease linked to ADAM17 deletion. N Engl J Med 2011;365:1502–1508.

59. Karamchandi-Patel G, Hanson EP, Saltzman R, et al. Congenital alterations of NEMO glutamic acid 223 result in hydropidrotic ectodermal dysplasia and immunodeficiency with normal serum IgG levels. Ann Allergy Asthma Immunol 2011;107:50–56.

60. Zimmer KP, Schumann H, Mecklenbeck S, Bruckner-Tuderman L. Esophageal stenosis in childhood: dystrophic epidermolysis bullosa without skin blistering due to collagen VII mutations. Gastroenterology 2002;122: 220–225.

61. Sadler E, Klausergger A, Muss W, et al. Novel KIND1 gene mutation in Kindler syndrome with severe gastrointestinal tract involvement. Arch Dermatol 2006;142:1619–1624.

62. Ussar S, Moser M, Widmaier M, et al. Loss of Kindlin-1 causes skin atrophy and lethal neonatal intestinal epithelial dysfunction. PLoS Genet 2008;4:e1000289.

63. Kern JS, Herz C, Haan E, et al. Chronic colitis due to an epithelial barrier defect: the role of kindlin-1 isoforms. J Pathol 2007;213:462–470.

64. Fiskerstrand T, Arshad N, Haukanes BI, et al. Familial diarrhea syndrome caused by an activating GUCY2C mutation. N Engl J Med 2012;366:1586–1595.

65. Chalaris A, Adam N, Sina C, et al. Critical role of the disintegrin metalloprotease ADAM17 for intestinal inflammation and regeneration in mice. J Exp Med 2010; 207:1617–1624.

66. Nenci A, Becker C, Wullaert A, et al. Epithelial NEMO links innate immunity to chronic intestinal inflammation. Nature 2007;446:557–561.
67. Zaph C, Troy AE, Taylor BC, et al. Epithelial-cell-intrinsic IKK-beta expression regulates intestinal immune homeostasis. Nature 2007;446:552–556.

68. Cheng LE, Kanwar B, Tcheurekdjian H, et al. Persistent systemic inflammation and atypical enterocolitis in patients with NEMO syndrome. Clin Immunol 2009;132:124–131.

69. Luetteke NC, Qiu TH, Peiffer RL, et al. TGF alpha deficiency results in hair follicle and eye abnormalities in targeted and waved-1 mice. Cell 1993;73:263–278.

70. Mann GB, Fowler KJ, Gabriel A, et al. Mice with a null mutation of the TGF alpha gene have abnormal skin architecture, wavy hair, and curly whiskers and often develop cornneal inflammation. Cell 1993;73:249–261.

71. Strober W, Fuss IJ, Blumberg RS. The immunology of mucosal models of inflammation. Annu Rev Immunol 2002;20:495–549.

72. Hand TW, Dos Santos LM, Bouladoux N, et al. Acute gastrointestinal infection induces long-lived microbiota-specific T cell responses. Science 2012;337:1553–1556.

73. Cong Y, Feng T, Fujihashi K, et al. A dominant, coordi- nated T regulatory cell-IgA response to the intestinal microbiota. Proc Natl Acad Sci USA 2009;106:19256–19261.

74. Mombaerts P, Iacomini J, Johnson RS, et al. RAG–1 deficient mice have no mature B and T lymphocytes. Cell 1992;68:869–877.

75. Shinkai Y, Rathbun G, Lam KP, et al. RAG-2-deficient mice lack mature lymphocytes owing to inability to initiate V(D)J rearrangement. Cell 1992;68:855–867.

76. Peschon JJ, Morrissey PJ, Grabstein KH, et al. Early lymphocyte expansion is severely impaired in interleukin 7 receptor-deficient mice. J Exp Med 1994;180:1955–1960.

77. Pieper K, Grimbacher B, Eibel H. B-cell biology and development. J Allergy Clin Immunol 2013;131:959–971.

78. Vorechovsky I, Sideras P, et al. The gene involved in X-linked agammaglobulinaemia is a member of the src family of protein-tyrosine kinases. Nature 1993;361:226–233.

79. Conley ME, Notarangelo LD, Etzioni A. Diagnostic criteria for primary immunodeficiencies. Representing PAGID (Pan-American Group for Immunodeficiency) and ESID (European Society for Immunodeficiencies). Clin Immunol 1999;93:190–197.

80. Pai SY, Cowan MJ. Stem cell transplantation for primary immunodeficiency diseases: the North American experience. Curr Opin Allergy Clin Immunol 2014;14:521–526.

81. Shearer WT, Dunn E, Notarangelo LD, et al. Establishing diagnostic criteria for severe combined immunodeficiency disease (SCID), leaky SCID, and Omenn syndrome: the Primary Immune Deficiency Treatment Consortium experience. J Allergy Clin Immunol 2014;133:1092–1098.

82. Puel A, Ziegler SF, Buckley RH, Leonard WJ. Defective IL7R expression in T+ B+ NK+ severe combined immunodeficiency. Nat Genet 1998;20:394–397.

83. Dadi HK, Simon AJ, Roifman CM. Effect of CD3delta deficiency on maturation of alpha/beta and gamma/delta T-cell lineages in severe combined immunodeficiency. N Engl J Med 2003;349:1821–1828.

84. Nielsen C, Jakobsen MA, Larsen MJ, et al. Immunodeciency associated with a nonsense mutation of IKBKB. J Clin Immunol 2014;34:916–921.

85. Derry JM, Ochs HD, Francke U. Isolation of a novel gene mutated in Wiskott-Aldrich syndrome. 1994;78:635–644.

86. Watanabe Y, Sasahara Y, Ramesh N, et al. T-cell receptor ligation causes Wiskott-Aldrich syndrome protein degradation and F-actin assembly downregulation. J Allergy Clin Immunol 2013;132:648–655.e1.

87. Shimizu M, Kanegane H, Wada T, et al. Aberrant glyco- sylation of IgA in Wiskott-Aldrich syndrome and X-linked thrombocytopenia. J Allergy Clin Immunol 2013;131:587–590, e1–3.

88. Westerberg LS, Dahlberg C, Baptista M, et al. Wiskott- Aldrich syndrome protein (WASP) and N-WASP are critical for peripheral B-cell development and function. Blood 2012;119:3966–3974.

89. Becker-Herman S, Meyer-Bahlburg A, Schwartz MA, et al. WASp-deficient B cells play a critical, cell-intrinsic role in triggering autoimmunity. J Exp Med 2011;208:2033–2042.

90. Lanzi G, Moratto D, Vairo D, et al. A novel primary human immunodeficiency due to deficiency in the WASP-interacting protein WIP. J Exp Med 2012;209:29–34.

91. Nguyen DD, Maillard MH, Cotta-de-Almeida V, et al. Lymphocyte-dependent and Th2 cytokine-associated colitis in mice deficient in Wiskott-Aldrich syndrome protein. Gastroenterology 2007;133:1186–1197.

92. Maillard MH, Cotta-de-Almeida V, Takeshima F, et al. The Wiskott-Aldrich syndrome protein is required for the function of CD4+CD25+ Foxp3+ regulatory T cells. J Exp Med 2007;204:381–391.

93. Nguyen DD, Wurbel MA, Goettel JA, et al. Wiskott- Aldrich syndrome protein deficiency in innate immune cells leads to mucosal immune dysregulation and colitis in mice. Gastroenterology 2012;143:719–729, e1–2.

94. van der Vliet HJ, Nieuwenhuis EE. IPEX as a result of mutations in FOXP3. Clin Dev Immunol 2007;2007:39170.

95. Torgerson TR, Ochs HD. Immune dysregulation, polyendocrinopathy, enteropathy, X-linked syndrome: a paradigm of immunodeficiency due to deficiency in the WASP-interacting protein WIP. J Exp Med 2012;209:29–34.

96. Barzaghi F, Passerini L, Bacchetta R. Immune dysregulation, polyendocrinopathy, enteropathy, X-linked syndrome: a paradigm of immunodeficiency with autoimmunity. Front Immunol 2012;3:211.

97. Zeissig S, Petersen BS, Tomczak M, et al. Early-onset Crohn’s disease and autoimmunity associated with a variant in CTLA-4. Gut 2014, Published online. http://dx.doi.org/10.1136/gutjnl-2014-308541.

98. Cebula A, Seweryn M, Rempala GA, et al. Thymus- derived regulatory T cells contribute to tolerance to commensal microbiota. Nature 2013;497:258–262.

99. Josepowicz SZ, Lu LF, Rudensky AY. Regulatory T cells: mechanisms of differentiation and function. Annu Rev Immunol 2012;30:531–564.

100. Lathrop SK, Bloom SM, Rao SM, et al. Peripheral education of the immune system by colonic commensal microbiota. Nature 2011;478:250–254.
101. Chinen T, Volchkov PY, Chervonsky AV, et al. A critical role for regulatory T cell-mediated control of inflammation in the absence of commensal microbiota. J Exp Med 2010;207:2323–2330.

102. Schultz M, Tonkonogy SL, Sellon RK, et al. IL-2-deficient mice raised under germfree conditions develop delayed mild focal intestinal inflammation. Am J Physiol 1999;276:G1461–G1472.

103. Schiering C, Krausgruber T, Chomka A, et al. The alarmin IL-33 promotes regulatory T-cell function in the intestine. Nature 2014;513:564–568.

104. Shim JO, Hwang S, Yang HR, et al. Interleukin-10 receptor mutations in children with neonatal-onset Crohn’s disease and intractable ulcerating enterocolitis. Eur J Gastroenterol Hepatol 2013;25:1235–1240.

105. Moore KW, de Waal Malefyt R, Coffman RL, et al. Interleukin-10 and the interleukin-10 receptor. Annu Rev Immunol 2001;19:683–765.

106. Hutchins AP, Diez D, Miranda-Saavedra D. The IL-10/IL-10 receptor signaling in innate immune cells regulates anti-inflammatory response: recent developments and future challenges. Brief Funct Genomics 2013;12:489–498.

107. Engelhardt KR, Grimbacher B. IL-10 in humans: lessons from the gut, IL-10/IL-10 receptor deficiencies, and IL-10 polymorphisms. Curr Top Microbiol Immunol 2014;380:1–18.

108. Murray PJ. The primary mechanism of the IL-10-regulated antiinflammatory response is to selectively inhibit transcription. Proc Natl Acad Sci USA 2005;102:8686–8691.

109. Neven B, Mamessier E, Bruneau J, et al. A Mendelian predisposition to B-cell lymphoma caused by IL-10R deficiency. Blood 2013;122:3713–3722.

110. Engelhardt KR, Shah N, Faizura-Yeop I, et al. Clinical outcome in IL-10- and IL-10 receptor-deficient patients with or without hematopoietic stem cell transplantation. J Allergy Clin Immunol 2013;131:825–830.

111. Murugan D, Albert MH, Langemeier J, et al. Very early onset inflammatory bowel disease associated with aberrant trafficking of IL-10R1 and cure by T cell replete haploidentical bone marrow transplantation. J Clin Immunol 2013;34:331–339.

112. Kuhn R, Kohler J, Rennick D, et al. Interleukin-10-deficient mice develop chronic enterocolitis. Cell 1993;75:263–274.

113. Sellon RK, Tonkonogy S, Schultz M, et al. Resident enteric bacteria are necessary for development of spontaneous colitis and immune system activation in interleukin-10-deficient mice. Infect Immun 1998;66:5224–5231.

114. Rubtsov YP, Rasmussen JP, Chi EY, et al. Regulatory T cell-derived interleukin-10 limits inflammation at environmental interfaces. Immunity 2008;28:546–558.

115. Roers A, Siewe L, Strittmatter E, et al. T cell-specific inactivation of the interleukin 10 gene in mice results in enhanced T cell responses but normal innate responses to lipopolysaccharide or skin irritation. J Exp Med 2004;200:1289–1297.

116. Shouval DS, Biswas A, Goettel JA, et al. Interleukin-10 receptor signaling in innate immune cells regulates mucosal immune tolerance and anti-inflammatory macrophage function. Immunity 2014;40:706–719.

117. Zigmond E, Bernshtein B, Friedlander G, et al. Macrophage-restricted interleukin-10 receptor deficiency, but not IL-10 deficiency, causes severe spontaneous colitis. Immunity 2014;40:720–733.

118. Sonnenberg GF, Fouser LA, Artis D. Border patrol: regulation of immunity, inflammation and tissue homeostasis at barrier surfaces by IL-22. Nat Immunol 2011;12:383–390.

119. Sonnenberg GF, Artis D. Innate lymphoid cell interactions with microbiota: implications for intestinal health and disease. Immunity 2012;37:601–610.

120. Spits H, Artis D, Colonna M, et al. Innate lymphoid cells—a proposal for uniform nomenclature. Nat Rev Immunol 2013;13:145–149.

121. Sonnenberg GF, Monticelli LA, Alenghat T, et al. Innate lymphoid cells promote anatomical containment of lymphoid-resident commensal bacteria. Science 2012;336:1321–1325.

122. Hepworth MR, Monticelli LA, Fung TC, et al. Innate lymphoid cells regulate CD4+ T-cell responses to intestinal commensal bacteria. Nature 2013;498:113–117.

123. Bernink JH, Peters CP, Munneke M, et al. Human type 1 innate lymphoid cells accumulate in inflamed mucosal tissues. Nat Immunol 2013;14:221–229.

124. Geremia A, Arancibia-Carcamo CV, Fleming MP, et al. IL-23-responsive innate lymphoid cells are increased in inflammatory bowel disease. J Exp Med 2011;208:1127–1133.

125. Takayama T, Kamada N, Chinen H, et al. imbalance of NKp44+NKp46+ and NKp44–NKp46– natural killer cells in the intestinal mucosa of patients with Crohn’s disease. Gastroenterology 2010;139:882–892, e1–3.

126. Ciccia F, Accardo-Palumbo A, Alessandro R, et al. Interleukin-22 and interleukin-22-producing NKp44+ natural killer cells in subclinical gut inflammation in ankylosing spondylitis. Arthritis Rheum 2012;64:1869–1878.

127. Fuchs A, Vermi W, Lee JS, et al. Intraepithelial type 1 innate lymphoid cells accumulate in inflamed mucosal tissues. Nat Immunol 2013;14:221–229.

128. Hepworth MR, Fung TC, Masur SH, et al. Immune tolerance. Group 3 innate lymphoid cells mediate intestinal selection of commensal bacteria-specific CD4+ T cells. Science 2015;348:1031–1035.

129. Kang EM, Marciano BE, DeRavin S, et al. Chronic granulomatous disease: overview and hematopoietic stem cell transplantation. J Allergy Clin Immunol 2011;127:1319–1326.

130. Abo A, Pick E, Hall A, et al. Activation of the NADPH oxidase involves the small GTP-binding protein p21rac1. Nature 1991;353:668–670.

131. Matute JD, Arias AA, Wright NA, et al. A new genetic subgroup of chronic granulomatous disease with autosomal recessive mutations in p40 phox and selective defects in neutrophil NADPH oxidase activity. Blood 2009;114:3309–3315.

132. Jones LB, McGrogan P, Flood TJ, et al. Special article: chronic granulomatous disease in the United Kingdom.
and Ireland: a comprehensive national patient-based registry. Clin Exp Immunol 2008;152:211–218.

133.Rosenzweig SD. Inflammatory manifestations in chronic granulomatous disease (CGD). J Clin Immunol 2008;28(Suppl 1):S67–S72.

134.Foster CB, Lehnbacher T, Mol F, et al. Host defense molecule polymorphisms influence the risk for immune-mediated complications in chronic granulomatous disease. J Clin Invest 1998;102:2146–2155.

135.Muise AM, Xu W, Guo CH, et al. NADPH oxidase complex and IBD candidate gene studies: identification of a rare variant in NCF2 that results in reduced binding to Rac2. Gut 2012;61:1028–1035.

136.Dhillon SS, Fattouh R, Elkadri A, et al. Variants in nicotinamide adenine dinucleotide phosphate oxidase complex components determine susceptibility to very early onset inflammatory bowel disease. Gastroenterology 2014;147:680–689.e2.

137.Roos D, Law SK. Hematologically important mutations: leukocyte adhesion deficiency. Blood Cells Mol Dis 2001;27:1000–1004.

138.van de Vijver E, Maddalena A, Sanal O, et al. Hematologically important mutations: leukocyte adhesion deficiency (first update). Blood Cells Mol Dis 2012;48:53–61.

139.Schmidt S, Moser M, Sperandio M. The molecular basis of leukocyte recruitment and its deficiencies. Mol Immunol 2013;55:49–58.

140.Davis MK, Rufo PA, Polyak SF, et al. Adalimumab for the treatment of Crohn-like colitis and enteritis in glycogen storage disease type I b. J Inherit Metab Dis 2008;31(Suppl 3):505–509.

141.Uzel G, Orange JS, Poliak N, et al. Complications of tumor necrosis factor-alpha blockade in chronic granulomatous disease-related colitis. Clin Infect Dis 2010;51:1429–1434.

142.Kato K, Kojima Y, Kobayashi C, et al. Successful allogeneic hematopoietic stem cell transplantation for chronic granulomatous disease with inflammatory complications and severe infection. Int J Hematol 2011;94:479–482.

143.de Luca A, Smeekens SP, Casagrande A, et al. IL-1 receptor blockade restores autophagy and reduces inflammation in chronic granulomatous disease in mice and in humans. Proc Natl Acad Sci USA 2014;111:3526–3531.

144.Bianco AM, Girardelli M, Vozzi D, et al. Mevalonate kinase deficiency and IBD: shared genetic background. Gut 2014;63:1367–1368.

145.Kuloglu Z, Kansu A, Ustundag G, et al. An infant with severe refractory Crohn’s disease and homozygous MEFV mutation who dramatically responded to colchicine. Rheumatol Int 2012;32:783–785.

146.Beser OF, Kasapcopur O, Cokugras FC, et al. Association of inflammatory bowel disease with familial Mediterranean fever in Turkish children. J Pediatr Gastroenterol Nutr 2013;56:498–502.

147.Mora AJ, Wolsohn DM. The management of gastrointestinal disease in Hermansky-Pudlak syndrome. J Clin Gastroenterol 2011;45:700–702.

148.Almeida de Jesus A, Goldbach-Mansky R. Monogenic autoimmune diseases: concept and clinical manifestations. Clin Immunol 2013;147:155–174.

149.Specckmann C, Lehmbek G, Albert MH, et al. X-linked inhibitor of apoptosis (XIAP) deficiency: the spectrum of presenting manifestations beyond hemophagocytic lymphohistiocytosis. Clin Immunol 2013;149:133–141.

150.Latour S, Aguilar C. XIAP deficiency syndrome in humans. Semin Cell Dev Biol 2015;39:115–123.

151.Pedersen J, LaCasse EC, Seidelin JB, et al. Inhibitors of apoptosis (IAPs) regulate intestinal immunity and inflammatory bowel disease (IBD) inflammation. Trends Mol Med 2014;20:652–665.

152.Aguilar C, Latour S. X-linked inhibitor of apoptosis protein deficiency: more than an X-linked lymphoproliferative syndrome. J Clin Immunol 2015;35:331–338.

153.Filipovich AH. The expanding spectrum of hemophagocytic lymphohistiocytosis. Curr Opin Allergy Clin Immunol 2011;11:512–516.

Received March 9, 2015. Accepted June 21, 2015.

Correspondence
Address correspondence to: Gregory F. Sonnenberg, PhD, Department of Microbiology and Immunology, Weill Cornell Medical College, Cornell University, 413 East 69th Street, Belfer Research Building 512, Box 190, New York, NY 10021. e-mail: gfsonnenberg@med.cornell.edu; or Judith R. Kelsen, MD, 7NW, Division of Pediatric Gastroenterology, 3400 Civic Center Boulevard, The Children’s Hospital of Philadelphia, Philadelphia, PA 19104. Fax: (215) 590-5326.

Conflicts of interest
The authors disclose no conflicts.

Funding
This study was funded by the National Institutes of Health (K23DK100461-01A1) (to J.R.K.); the National Institutes of Health (DP5OD012116 and R56AI114724), the NIAID Mucosal Immunology Studies Team (MIST) Scholar Award in Mucosal Immunity and the Institute for Translational Medicine, and the Therapeutics Transdisciplinary Program in Translational Medicine and Therapeutics (UL1RR024134 from the US National Center for Research Resources) (to G.F.S. laboratory); the National Institutes of Health (AI061570, AI074878, AI095466, AI095608, AI102942, AI097331 and AI108697), the Burroughs Wellcome Fund Investigator in Pathogenesis of Infectious Disease Award, and the Crohn’s and Colitis Foundation of America (to D.A. laboratory).