Case Study Monograph

Gastroenterology & Hepatology May 2011

Community Perspectives on Serologic and Genetic Testing for Crohn's Disease

Commentator

Marla C. Dubinsky, MD
Cedars Sinai Medical Center
Los Angeles, California

Faculty

Morris A. Barocas, MD
West Knoxville Gastroenterology
Knoxville, Tennessee

Charles J. Loewe, MD
Center for Digestive Diseases
Sarasota, Florida

Chandar Singaram, MD, MBA
University of South Dakota
Midwest Medical Care, P. C.
Sioux Falls, South Dakota

Ronald Soltis, MD
University of Minnesota
Minneapolis, Minnesota

Jeffrey A. Tuvlin, MD
Jewish Physician Group
Jewish Hospital and St. Mary's Health System
Louisville, Kentucky

Abstract

One of the major challenges in managing patients with inflammatory bowel disease (IBD) is predicting the likelihood of rapid disease progression. While some IBD patients have mild disease that can be adequately managed with conservative medical therapy, other patients require more aggressive medical treatment or surgery. In determining the best approach for a particular case, clinicians can benefit from serologic and genetic testing, as various genetic mutations and serologic markers are associated with a higher rate of complications in IBD patients. Specifically, 3 mutations in the *NOD2* gene are associated with a more aggressive course of Crohn's disease: R702W, G908R, and 007fs. Serologic markers that are associated with a more complicated disease course may include antibodies to outer membrane porin C, CBir1, and *Pseudomonas fluorescens*–related protein, as well as anti–*Saccharomyces cerevisiae* antibody. Analyzed together, genetic mutations and serologic markers may provide useful information about the likelihood of rapid disease progression and complications. In this monograph, 5 cases illustrate how serogenetic testing may inform patient management.

Supported through funding from Prometheus Laboratories

GASTROENTEROLOGY & HEPATOLOGY™
The Independent Peer-Reviewed Journal

Table of Contents

Indexed through the National Library of Medicine (PubMed/Medline), PubMed Central (PMC), and EMBASE

Disclaimer
Funding for this case study monograph has been provided from Prometheus Laboratories. Support of this monograph does not imply the supporter's agreement with the views expressed herein. Every effort has been made to ensure that drug usage and other information are presented accurately; however, the ultimate responsibility rests with the prescribing physician. Gastro-Hep Communications, Inc., the supporter, and the participants shall not be held responsible for errors or for any consequences arising from the use of information contained herein. Readers are strongly urged to consult any relevant primary literature. No claims or endorsements are made for any drug or compound at present under clinical investigation.

Introduction

Marla C. Dubinsky, MD

The chronic nature of inflammatory bowel disease (IBD) requires that patients receive long-term therapy, generally throughout their lives. However, the severity of IBD varies among patients, with some individuals requiring only mild treatment strategies to maintain control of their disease, while others require more aggressive approaches. Traditionally, physicians used the patient's current clinical situation to guide therapeutic choices. However, this strategy does not fully address the long-term goal of altering the natural history of the disease. Thus, IBD patients would benefit from a tool that could help to determine prognosis and guide therapeutic choices. In an effort to improve IBD prognosis, several genetic and serologic markers have been investigated that may help to predict disease behavior.

One of the most well-described genes to have been associated with Crohn's disease (CD) is *NOD2*, a component of the innate immune system.[1,2] *NOD2* plays an important role in recognizing bacteria in the intestine and stimulating a protective response against these bacteria.[3,4] A number of mutations within *NOD2* have been described, 3 of which have been particularly associated with CD. Two of these mutations—R702W (single-nucleotide polymorphism [SNP] 8) and G908R (SNP 12)—are missense mutations, while the third—007fs (SNP 13)—is a frameshift mutation.[2,5,6] Importantly, CD patients with *NOD2* mutations have been found to have a more aggressive disease course.

Serologic markers, which were first explored for their possible ability to differentiate between CD and ulcerative colitis (UC), have now been shown to have an important role in determining patient prognosis. Several markers, particularly the anti–*Saccharomyces cerevisiae* antibody (ASCA) and the perinuclear antineutrophil cytoplasmic antibody (pANCA), have emerged as tools that can help to identify CD patients who have a high risk for developing disease complications and will likely require surgery. While higher levels of ASCA—both immunoglobulin (Ig)A and IgG—have been associated with an increased risk of aggressive disease and small bowel disease location, higher levels of pANCA are associated with CD that has a more UC-like phenotype.[7,8]

In addition to ASCA and pANCA, other prognostic serologic markers have also been investigated. For example, elevated levels of the antibody to *Escherichia coli* outer membrane porin C (OmpC) and the antibody to CBir1 (flagellin) have been found to be associated with an increased risk of complicating CD phenotypes.[8,9] Both immune responses have been shown to be associated with stricturing and/or internal penetrating disease. Antibodies to the *Pseudomonas fluorescens*–related protein (I2) have been shown to be correlated with stricturing CD and small bowel surgery.[7]

Genetic and serologic markers have now been suggested to share a relationship, and they can produce a prognostic picture for CD patients when both are considered together. It has been hypothesized that *NOD2* mutations may result in an abrogated innate immune response overcompensated by the adaptive immune system.[10,11] As a result, these patients may show an overproduction of several antibodies to microbial antigens, including ASCA, pANCA, anti-OmpC, anti-CBir1, and anti-I2. Elevated levels of these antibodies are associated with a higher likelihood of rapid disease progression and complications that may eventually require surgical intervention.

These data are now being translated into clinical practice, allowing physicians and patients to take into account the patient's genetic and serologic status when considering a course of therapy. Now, genetic and serologic markers have been incorporated into a prognostic laboratory assay that can be used to assess a CD patient's probability of complications, which should provide an individualized approach to CD management.

References

1. Hugot JP, Chamaillard M, Zouali H, et al. Association of NOD2 leucine-rich repeat variants with susceptibility to Crohn's disease. *Nature.* 2001;411:599-603.
2. Ogura Y, Bonen DK, Inohara N, et al. A frameshift mutation in NOD2 associated with susceptibility to Crohn's disease. *Nature.* 2001;411:603-606.
3. Inohara N, Ogura Y, Nunez G. Nods: a family of cytosolic proteins that regulate the host response to pathogens. *Curr Opin Microbiol.* 2002;5:76-80.
4. Kobayashi KS, Chamaillard M, Ogura Y, et al. Nod2-dependent regulation of innate and adaptive immunity in the intestinal tract. *Science.* 2005;307:731-734.
5. Shaw SH, Hampe J, White R, et al. Stratification by CARD15 variant genotype in a genome-wide search for inflammatory bowel disease susceptibility loci. *Hum Genet.* 2003;113:514-521.
6. Sugimura K, Taylor KD, Lin YC, et al. A novel NOD2/CARD15 haplotype conferring risk for Crohn disease in Ashkenazi Jews. *Am J Hum Genet.* 2003;72:509-518.
7. Mow WS, Vasiliauskas EA, Lin YC, et al. Association of antibody responses to microbial antigens and complications of small bowel Crohn's disease. *Gastroenterology.* 2004;126:414-424.
8. Abreu MT, Vasilliauskas EA, Kam LR, et al. Use of serologic tests in Crohn's disease. *Clin Perspect Gastroenterol.* 2001;4:155-164.
9. Targan SR, Landers CJ, Yang H, et al. Antibodies to CBir1 flagellin define a unique response that is associated independently with complicated Crohn's disease. *Gastroenterology.* 2005;128:2020-2028.
10. Devlin S, Yang H, Ippoliti A, et al. NOD2 variants and antibody response to microbial antigens in Crohn's disease patients and their unaffected relatives. *Gastroenterology.* 2007;132:576-586.
11. Papadakis KA, Yang H, Ippoliti A, et al. Anti-flagellin (CBir1) phenotypic and genetic Crohn's disease associations. *Inflamm Bowel Dis.* 2007;13:524-530.

Management of Crohn's Disease in a Patient with Clinical Findings But Few Symptoms

Jeffrey A. Tuvlin, MD

Case 1

The patient is a 49-year-old male who initially presented in July 2009 after a routine physical examination revealed increased liver function tests. The patient had been previously diagnosed with gastroesophageal reflux disease (GERD) and had a family history of liver cirrhosis and liver cancer. While the clinical suspicion was of fatty liver disease, a 3-phase computed tomography (CT) scan was ordered due to the patient's significant family history. The CT scan revealed normal liver findings, but a slight thickening was noted in the terminal ileum. The patient had no luminal or perianal complaints at this time.

Six months later, the patient developed an abscess in the rectal area that required incision and drainage. During a subsequent follow-up visit, the patient continued to complain of blood and drainage, and he reported having 1–2 bowel movements daily. Examination of the abscess area revealed a fistulous opening with active serosanguinous drainage that required antibiotic treatment. After further consultation with the general surgeon, an examination under anesthesia was performed; a seton was placed during the procedure. Concurrently, a colonoscopy was performed, revealing a perianal fistula, mild erythema of the left colon, and ulcers in the terminal ileum and ileocecal valve. Pathologic analysis of the terminal ileum and ileocecal valve biopsies revealed crypt distortion. Furthermore, a small bowel follow-through performed 1 week later showed that there were skipped areas of involvement with CD, including the terminal ileum. Laboratory tests demonstrated deficiencies in vitamin B_{12} and vitamin D.

Approximately 1 month after the small bowel follow-through, the patient was asymptomatic with the exception of continued drainage from the perianal fistula. Serogenetic testing revealed elevations in ASCA-IgA (>114.1 EU/mL) and ASCA-IgG (147.5 EU/mL) levels, but no *NOD2* mutations were detected. After a discussion of treatment options, the patient chose to initiate therapy with once-daily mesalamine and antibiotics as needed, rather than immunomodulator or biologic therapy. Three months later, a follow-up examination showed that the perianal drainage was improved.

After another 3 months, the patient was seen for another follow-up visit, at which time treatment options were revisited. Due to the patient's recurrent need for antibiotics and the presence of elevated markers indicating the potential for a more progressive disease course, the possible benefit of more aggressive therapy was considered. This benefit was weighed against the patient's current lack of luminal symptoms. To help choose the best treatment strategy, a Prometheus Crohn's Prognostic test was ordered. Results of this test indicated a 58% risk of complications at 5 years; this risk was predicted to increase to 65%, 78%, and 87% by 10, 20, and 30 years, respectively (Figure 1). The patient chose not to increase his therapy at the present time.

Discussion

What made this case particularly challenging and interesting was the fact that the patient presented with findings that we would typically associate with more aggressive disease (ie, fibrostenotic disease and perianal disease) while subjectively reporting relatively minimal symptoms. Indeed, it was almost as if we had accidentally uncovered the disease while working up other issues. Aside from perianal irritation, the patient reported no problems with diarrhea, bleeding, or abdominal pain. In this type of situation, we are forced to ask ourselves: Is it better to "treat the patient and not the x-ray," or is it better to be more aggressive with medical therapy in an effort to prevent future complications and need for surgery? Having learned from our rheumatology colleagues and their management of joint destruction, gastroenterologists have become interested in the idea of using more aggressive therapy earlier in the disease process to try and prevent

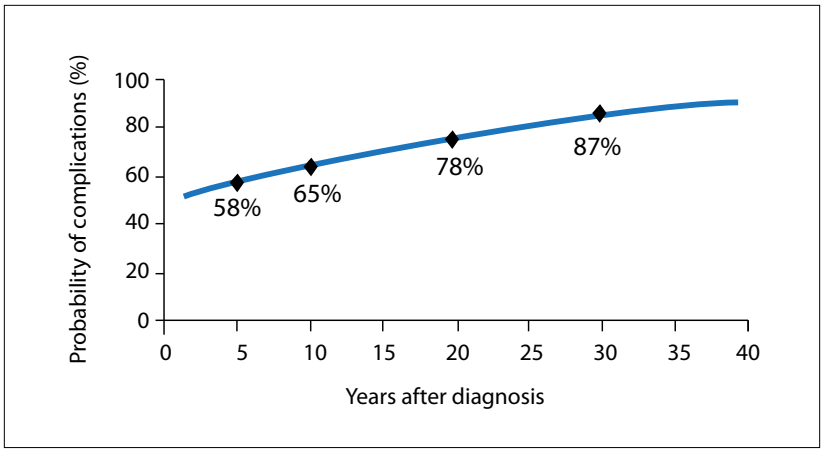

Figure 1. The Crohn's Prognostic test for this patient showed a 58% risk of complications at 5 years after diagnosis.

irreversible damage to the gastrointestinal tract. Obviously, given the potentially significant side effects associated with immunomodulators and anti–tumor necrosis factor (TNF) agents, there is no "free lunch"—whether we should treat earlier or wait is an important question with no clear answer.

While I recommended that this patient increase therapy, the patient opted not to assume the risks of immunomodulatory or biologic therapy at this time, as his only persistent complaint was minimal drainage from the perianal fistula (which was effectively treated with antibiotics). In making a decision about therapy, the patient appreciated having the Crohn's Prognostic test to help him better understand his disease. While acknowledging the limitations of any blood test, the patient felt that the test allowed him to commit to a plan of care and treatment algorithm with more confidence and less worry that he was under- or overtreating his condition. With the Crohn's Prognostic test, he felt confident holding off on more aggressive therapy at the present time, while also being ready to commit to increased therapy if his symptoms changed or worsened. In the treatment of IBD, patients often feel helpless and not in control; thus, it was refreshing that this patient felt very in control of the decision-making process.

In the future, if this patient begins to develop more active or difficult-to-control symptoms, an argument could be made for several treatment options. Immunomodulatory or anti-TNF therapy alone would be reasonable; however, based on newer clinical trials and the fact that this patient is immunomodulator-naïve, combination therapy with an immunomodulator and anti-TNF therapy may be most effective. Of course, the risks and benefits of each of these approaches would need to be discussed.

Diagnosis of Crohn's Disease in a Patient with Recurrent Perianal Abscesses and Fistulae

Charles J. Loewe, MD

Case 2

The patient is a 44-year-old white male who presented to the emergency room with a nondraining and painful perirectal abscess but no fever. He began treatment with ciprofloxacin and metronidazole. The abscess drained spontaneously 1 day later, and the patient experienced significant relief. The patient reports having had intermittent drainage, pain, and abscess formation in the same perianal region over the past 6 years; he also has a history of perianal fistulae, with a fistulotomy performed approximately 5 years earlier. A colonoscopy performed 4 years ago revealed active, moderate, chronic colitis that was not thought to be specific to CD. He has not received any treatment for CD, and his most recent colonoscopy was negative. The patient does not smoke and only drinks occasionally. His family history includes a mother with irritable bowel syndrome and a sister with kidney stones.

At a subsequent follow-up appointment, the patient's physical examination was unrevealing with the exception of a residual perianal abscess in the left lateral rectal area. There was no evidence of fistulae, fissures, or hemorrhoids. A small amount of purulent drainage remained in an indurated and slightly erythematous area. Digital rectal examination revealed mild tenderness but no palpable masses. A fistulogram showed a cutaneous perianal fistula that extended into the distal sigmoid and rectum. Two focal defects were evident in the skin to the left of the anus. The smaller defect was cannulated with a thin catheter and a small balloon that was inflated to occlude the fistulous tract; nonionic contrast was then injected until a thin fistulous tract extended to the distal sigmoid region of the upper rectum. A CT scan of the pelvis with sagittal and coronal reconstructions was also performed; it showed contrast within the distal rectum that was compatible with the prior anal fistula extending to the distal bowel. Mild fibrotic thickening of the distal sigmoid colon was also evident and found to be potentially suggestive of IBD. Possible muscular hypertrophy and fibrosis could not be excluded. Based on these studies, the patient was diagnosed with complex rectal fistula.

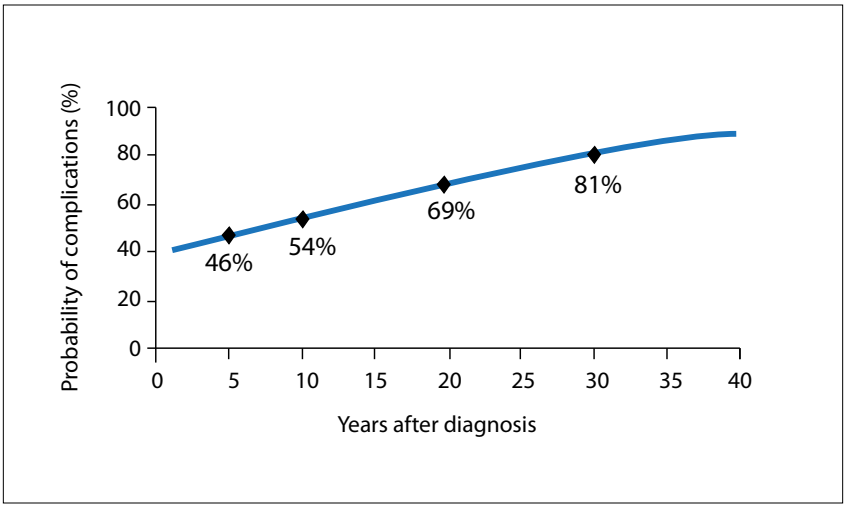

Figure 2. The probability of complications curve for this patient was derived from his serologic profile and genetic result showing that the patient was heterozygous for the SNP 12 mutation.

In order to rule out IBD, a colonoscopy was performed, and multiple biopsies of the distal rectum and ileum were taken to check for evidence of CD. A flat, 0.5-cm polyp was removed in the midsigmoid colon. Biopsies of the ileum and rectum showed no features indicative of IBD and no evidence of dysplasia. A polyp in the sigmoid colon revealed a tiny fragment of small intestinal mucosa that was within normal limits.

Serogenetic testing was also performed; results revealed elevated levels of ASCA-IgA (76.1 EU/mL) and ASCA-IgG (29.8 EU/mL) and a heterozygous mutation at SNP 12 (G908R). Overall, prognostic testing showed that the patient's probability of developing complications at 5 years was 46%; this risk increased to 54%, 69%, and 81% at 10, 20, and 30 years, respectively (Figure 2).

The patient's treatment included ciprofloxacin and metronidazole for 6–8 weeks and the addition of 6-mercaptopurine at a dose of 1.5 mg/kg.

Discussion

Perianal fistulae can be a cause of significant morbidity in patients with CD. In this case, pain, scarring, and fecal incontinence had a significant impact on the patient's quality of life. Previously, perianal CD was treated with surgery; however, with new advances in diagnosis and therapy, a multidisciplinary approach was taken in this case. Successful management of this patient's perianal disease consisted of first establishing the activity of the luminal disease and delineating the fistulizing process. Both endoscopy and magnetic resonance imaging (MRI) were used to assess this patient's perianal fistula. A recent study showed that a combination of either MRI or endorectal ultrasound with examination under anesthesia can allow perianal fistulae to be assessed with near-100% accuracy.[1] This case was especially challenging because the patient was a soldier who had been overseas; he had had recurrent symptoms but no true diagnosis prior to his evaluation at our center.

A Perianal Disease Activity Index was tabulated to quantify this patient's disease activity according to several criteria: discharge, type of perianal disease, degree of induration, pain/restriction of activities, and restriction of sexual activity. Since negligible intraluminal activity was seen on endoscopy, a Prometheus Crohn's Prognostic test was also obtained. The Crohn's Prognostic test showed that the patient carried markers associated with a higher likelihood of disease complications. Current studies and data show that patients with such markers (ie, antibodies and positive *NOD2* SNPs) will have a more complicated disease course and will require more hospitalizations and surgery.[2,3] This finding, along with the patient's presentation, led to more aggressive treatment in this case, including the addition of immunosuppressive medications and consideration of biologic therapy. Overall, the Crohn's Prognostic test added confidence to the decision to take a more aggressive, top-down treatment approach. With further refinement, I hope future tests will also allow me to predict response to therapy, which could help me to further personalize each patient's therapy.

Reference

1. Schwartz DA, Wiersema MJ, Dudiak KM, et al. A comparison of endoscopic ultrasound, magnetic resonance imaging, and exam under anesthesia for evaluation of Crohn's perianal fistulas. *Gastroenterology*. 2001;121:1064-1072.
2. Ippoliti A, Devlin S, Mei L, et al. Combination of innate and adaptive immune alterations increased the likelihood of fibrostenosis in Crohn's disease. *Inflamm Bowel Dis*. 2010;16:1279-1285.
3. Allez M, Lemann M, Bonnet J, Cattan P, Jian R, Modigliani R. Long term outcome of patients with active Crohn's disease exhibiting extensive and deep ulcerations at colonoscopy. *Am J Gastroenterol*. 2002;97:947-953.

Deciding Whether to Continue Long-Term Treatment in a Young Crohn's Disease Patient

Ronald Soltis, MD

Case 3

An 18-year-old undergraduate student was referred to the gastroenterology clinic from the student health service with a 6-week history of intermittent mid-abdominal pain that was worse about 1 hour after eating. This pain was cramping in nature, did not radiate, and was associated with increased intestinal noise. The patient had also noted a change in bowel habits over the past 2 weeks, with 2–3 soft stools per day and occasional dark red blood. He has had night sweats for approximately 1 year. He had no weight loss, and his appetite was good. There was no family history of IBD. Laboratory tests performed at the health service showed a decreased serum albumin level (3.1 mg/dL). The patient's past history was significant only for migraine headaches. The physical examination was normal with the exception of mild right lower quadrant tenderness. The initial impression was probable small bowel CD.

A Prometheus IBD Serology 7 test was obtained, which showed a pattern consistent with CD (Table 1). Colonoscopy demonstrated multiple shallow ulcers throughout the terminal ileum and the entire colon, with rectal sparing and several skip areas. Biopsies demonstrated active IBD consistent with CD. According to the radiologist's interpretation, magnetic resonance (MR) enterography showed "skip areas of bowel wall thickening in the sigmoid/descending colon junction and ascending colon. There is bowel wall thickening in the terminal ileum. Corresponding areas of hyperemic mucosa are present in these same regions. The findings are consistent with active ileitis and colitis. No evidence of penetrating, fistulizing, or fibrostenotic disease."

The patient was initially started on budesonide (9 mg daily). After 6 weeks, his symptoms were unchanged; he was switched to prednisone (30 mg daily), and approval was sought from his insurance company to begin certolizumab pegol. Approval for this drug was obtained, and he was started on 400 mg certolizumab pegol administered subcutaneously at Weeks 0, 2, 4, and every 4 weeks thereafter. Within 6 weeks, he was asymptomatic, and the prednisone was gradually tapered over the next month.

The patient remained asymptomatic while on certolizumab pegol. Six months after the drug was begun, he asked if it was necessary to continue therapy, since he was now in remission. Remission was confirmed by repeat colonoscopy, which showed minor edema of the terminal ileum and no ulcers. To assist with this decision, a Prometheus Crohn's Prognostic test was obtained; it showed a 58% probability of complications at 5 years (Figure 3). Based on this high probability, both the patient and his gastroenterologist agreed that long-term treatment was the proper course.

Discussion

For several years, there has been discussion and debate about the step-up versus step-down approaches for treating patients with CD. Before the advent of anti-TNF agents, there were relatively few treatment choices. Now, the question is whether to start with less aggressive choices (such as budesonide) and progress to anti-TNF agents if this approach fails, or whether to start with more aggressive agents and later "step down" if a response occurs. Until recently, we had very little data to guide us in making these decisions.

When this patient presented to our clinic, we obtained an IBD Serology 7 test that facilitated the diagnosis. The patient failed to respond to budesonide, was treated with a course of prednisone, and then was begun on certolizumab pegol. Subsequent endoscopy showed near-total healing after a relatively short period of time. Was now the time to step down?

Since the time of this patient's diagnosis, Prometheus Laboratories had developed the Crohn's Prognostic test, which measures 3 *NOD2* gene variants plus a series of serologic markers. These results are analyzed by a logistic regression algorithm to quantify the likelihood that a patient will progress to a complicated CD phenotype. The report gives a probability score reflecting the likelihood of disease progression to complications over time.

Recent literature has shown that the frequency and level of these serologic markers could predict the likelihood of future complications, namely fibrostenosing and internal penetrating disease and need for surgery.[1,2] Other studies have shown that the presence of any of the 3 *NOD2* gene variants also predicts a complicated disease course.[3] This patient's probability of complications was

Table 1. Prometheus IBD Serology 7 Results

Assay	ASCA IgA ELISA	ASCA IgG ELISA	Anti-OmpC IgA ELISA	Anti-CBir1 ELISA	IBD-specific pANCA		
					Autoantibody ELISA	IFA perinuclear pattern	DNAse sensitivity
Assay value	106.3 EU/mL	69.1 EU/mL	70.4 EU/mL	9.8 EU/mL	<12.1 EU/mL	Not detected	Not detected
Reference value	<20.0 EU/mL	<40.0 EU/mL	<16.5 EU/mL	<21.0 EU/mL	<12.1 EU/mL	Not detected	Not detected

Patient test results are based on the Smart Diagnostic Algorithm, which interprets patterns among the assay values. Assay and reference values are provided for prognostic interpretation.

ASCA=anti–*Saccharomyces cerevisiae* antibody; CBir1=flagellin; ELISA=enzyme-linked immunosorbent assay; IBD=inflammatory bowel disease; IFA=immunofluorescence antibody; Ig=immunoglobulin; OmpC=*Escherichia coli* outer membrane porin C; pANCA=perinuclear antineutrophil cytoplasmic antibody.

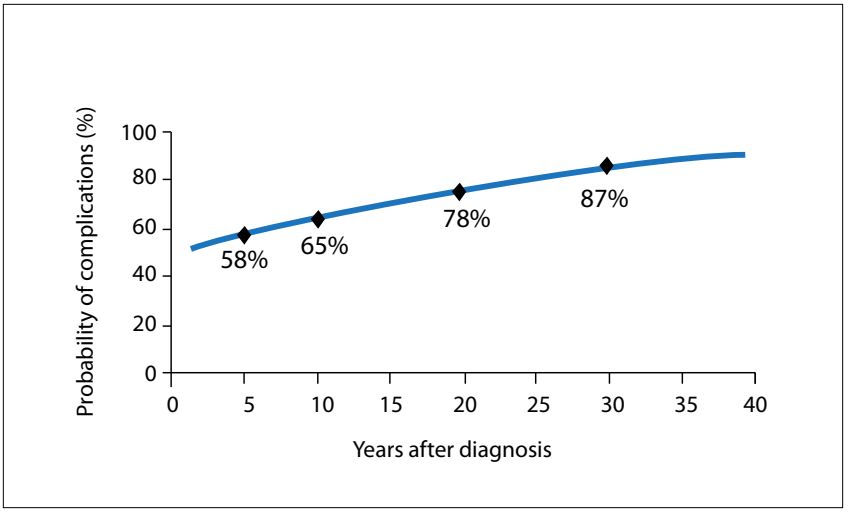

Figure 3. This patient's probability of complications curve indicated a greater-than-50% risk of complications at 5 years and an 87% chance of complications by 30 years after diagnosis.

quite high at 5 years, and since budesonide had not been an effective treatment, it seemed unwise to step down to less aggressive therapy. Had the Crohn's Prognostic test been available at the time of the initial diagnosis, in conjunction with his clinical findings, we likely would have begun therapy with an anti-TNF agent. We are now routinely using this test on all newly diagnosed CD patients, as it is very useful in guiding the management course for the patient.

References

1. Lichtenstein GR. Emerging prognostic markers to determine Crohn's disease natural history and improve management strategies: a review of recent literature. *Gastroenterol Hepatol (N Y)*. 2010;6:99-107.
2. Lichtenstein GR, Targan SR, Dubinsky MC, et al. Combination of genetic and quantitative serological immune markers are associated with complicated Crohn's disease behavior. *Inflamm Bowel Dis*. 2011 Mar 9. Epub ahead of print.
3. Ippoliti A, Devlin S, Mei L, et al. Combination of innate and adaptive immune alterations increased the likelihood of fibrostenosis in Crohn's disease. *Inflamm Bowel Dis*. 2010;16:1279-1285.

Evaluation and Management of a Suspected Crohn's Disease Patient with Diarrhea, Pain, and Weight Loss

Morris A. Barocas, MD

Case 4

This patient is a 32-year-old white female who began to experience unexplained weight loss and diarrhea approximately 6 years prior to her current evaluation. She also developed back and abdominal pain 2 years ago. A CT scan performed during a brief hospitalization at that time revealed thickening of the distal small bowel that was suggestive of ileitis and may have been consistent with CD. No specific treatment was initiated at that time. Small bowel dilation was evident on a radiograph taken 3 years later. The patient remained symptomatic and presented to me earlier this year. She complained of dysphagia for solids and liquids and had a 13-year history of GERD, for which she regularly took antacids. She often has up to 5–8 loose bowel movements daily, which are associated with pain, cramping, and urgency. She also experiences rectal bleeding associated with rectal pain. More recently, her abdominal pain has become somewhat diffuse; occurs regardless of meals; and is associated with distention, nausea, and mild vomiting.

Current medications include carisoprodol, alprazolam, and hydrocodone for pain. Her medical history includes asthma, cervical cancer, a loop electrosurgical excision procedure, a dilation and curettage procedure, tubal ligation, and facial cosmetic surgery. She admits to smoking half a pack of cigarettes daily and occasionally drinks alcohol. Her family history is notable for both an uncle and a son with CD.

At the patient's initial visit, omeprazole was recommended for reflux, and mesalamine and budesonide were initiated based on a suspected diagnosis of CD. To confirm this diagnosis and plan the patient's treatment course, further evaluations included esophagogastroduodenoscopy (EGD), colonoscopy, small bowel biopsy (to rule out celiac disease), thiopurine methyltransferase (TMPT) enzyme activity assay, and a Prometheus Crohn's Prognostic test.

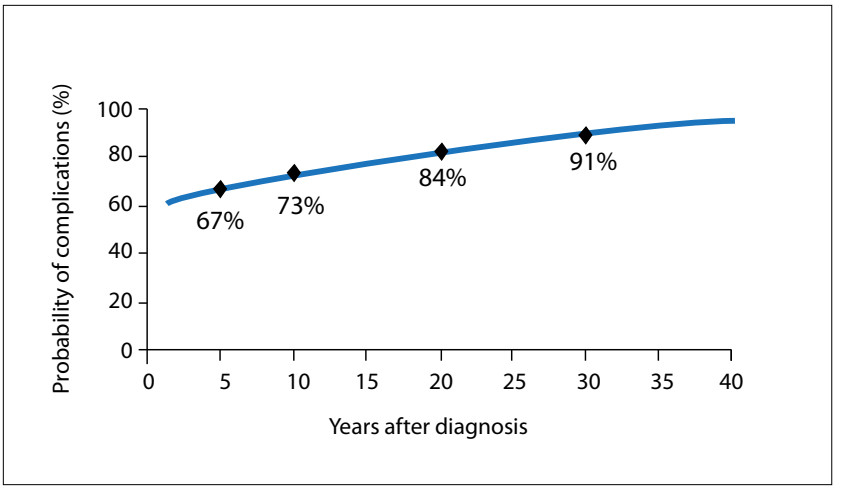

Figure 4. This patient's Prometheus Crohn's Prognostic test indicated a 67% probability of complications at 5 years after diagnosis.

EGD found the esophagus to be normal, without evidence of hiatal hernia, strictures, rings, webs, or Barrett mucosa. Erosive gastritis and hemorrhagic gastritis with submucosal hemorrhage were noted. Biopsies were taken for pathologic analysis; *Campylobacter*-like organism testing was positive for *Helicobacter pylori* infection. Proximal small bowel biopsies demonstrated minimal chronic inflammation. A 58-French Maloney dilator was passed to provide empiric symptomatic relief of dysphagia. Colonoscopy was performed, and biopsies were taken of the terminal ileum and randomly throughout the colon. Terminal ileal biopsies revealed chronic inflammation with architectural distortion. The random colonic biopsies were unremarkable.

Serologic testing showed an elevation in anti-CBir1 antibody (64.0 EU/mL) level, and a heterozygous mutation at SNP 8 (R702W) was also documented. The Prometheus Crohn's Prognostic test predicted a 67% probability of complications at 5 years, which increased to 73%, 84%, and 91% at 10, 20, and 30 years, respectively (Figure 4). The TMPT assay showed normal enzymatic activity (59.8 EU), and TMPT genetic testing identified alleles associated with normal activity.

In response to treatment with omeprazole, mesalamine, and budesonide, the patient's reflux symptoms improved; abdominal pain, nausea, and vomiting resolved; and stool frequency was reduced to 1–2 stools per day. The patient's weight returned to baseline within 3 months.

Discussion

Quantitative risk assessment of complicated small bowel disease—as provided by the Crohn's Prognostic test—can optimize patient management. Rather than categorizing a case as mild, moderate, or severe based on clinical presentation during a symptomatic flare or stratifying risk by clinical parameters (such as young age, small bowel or perianal disease, need for steroids at first flare, and smoking history), the Crohn's Prognostic test uses objective measures. Specifically, it quantifies the number and magnitude of immune responses to various antigens associated with penetrating or internal penetrating disease and need for surgery, as well as the presence of the 3 most common SNPs on the *NOD2/CARD15* gene that are associated with complicated CD.

Management and therapeutic selection is an important means by which patient care is optimized, but clinicians can also improve patient outcomes by recommending shorter intervals between follow-up visits; regularly screening for inflammatory markers such as C-reactive protein (CRP), erythrocyte sedimentation rate, and stool lactoferrin; and performing imaging using CT or MR enterocoloproctography, a capsule camera, and/or endoscopy to identify active asymptomatic complications prior to their clinical manifestation. Earlier detection allows for earlier aggressive intervention and improved prognosis.

Although the initial treatment in this case was commensurate with the patient's clinical severity at presentation, immunomodulatory therapy has since been added. This addition was based directly on the observation of early destructive architectural histologic changes and the results of the Crohn's Prognostic test, both of which suggested an opportunity to proactively intervene and attempt to change the natural history of this patient's illness. Should she develop additional signs or symptoms consistent with acceleration of moderate or severe disease, anti-TNF therapy will be employed as well. She has been encouraged to abstain from smoking and has remained compliant with her follow-up visits and medication regimen, which I feel is a result of her being presented with and understanding her unique complication curve.

Usefulness of Serogenetic Testing in the Management of a Young Patient with Crohn's Disease

Chandar Singaram, MD, MBA

Case 5

The patient is an 11-year-old female referred by a local chiropractor for further management of CD in November 2010. Exactly 2 years earlier, she experienced a viral flu-like illness that included vomiting and diarrhea; she never fully recovered from this episode and continued to have 4–10 loose stools per day.

An examination and barium upper gastrointestinal series at her local hospital demonstrated nodular inflammation of the terminal ileum that was suspicious for CD. The patient received prednisone and experienced some symptom relief. The patient's vaccination history included 3 immunizations; further immunizations were stopped after the age of 6 months due to signs of allergic reactions. She has no other history of medical illness or surgeries, and none of her family members (up to the second generation) have a known diagnosis of CD.

Colonoscopy performed at a tertiary care facility in March 2009 revealed pancolitis and granulomas, including some at the terminal ileum. At this time, the patient was started on budesonide (3 mg/day) and methotrexate injections. However, the patient did not tolerate these treatments well; she experienced significant burning and pain associated with the injections and hyperactivity related to the steroids. In addition, none of the attempted medications were able to decrease the patient's number of stools per day. The patient continued to lose weight and lacked appetite, and she required several emergency room visits for hydration and blood transfusions. She also became increasingly withdrawn.

Clinicians at the tertiary care clinic strongly advised the family to consider beginning infliximab therapy. Given the patient's sensitivity to childhood immunizations and side effects experienced with previous treatments, however, the family was reluctant to begin biologic therapy involving a mouse monoclonal antibody. As a last resort, the patient stopped steroid treatment and began nutritional supplementation under the care of a local functional medicine provider/chiropractor. Although she showed some improvement in energy

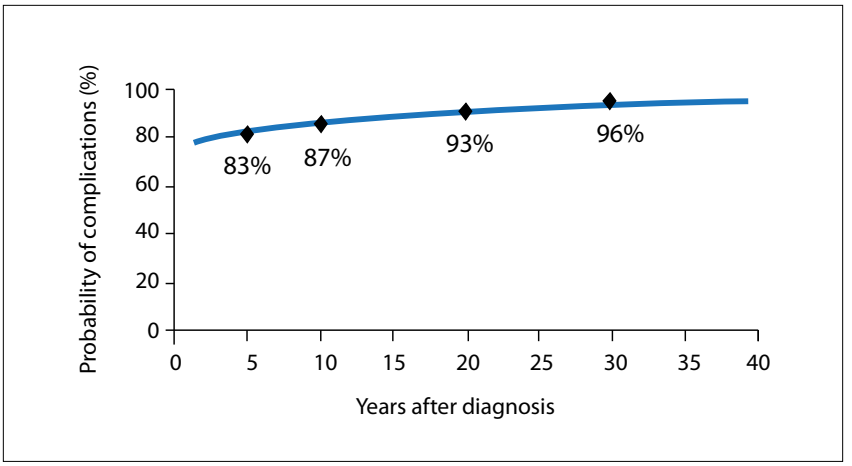

Figure 5. This patient's high probability of complications was calculated based on her elevations in ASCA-IgA and anti-CBir1, positivity for pANCA IFA perinuclear pattern and pANCA DNAse sensitivity, and her heterozygous mutation in SNP 13.

level, she continued to have 6–10 loose stools daily. She was therefore referred to my practice in November 2010 for further management.

At the time of referral, the patient appeared chronically ill, very pale, and very small; she weighed only 49 lbs and had a body mass index of 14.5 kg/m^2. Additionally, she showed evidence of glossitis, dermatitis, and pedal edema, and she had a palpable nodular lesion in the region of the terminal ileum. After counseling the family and carefully reviewing the previous work-up, a Prometheus Crohn's Prognostic test was obtained. This test showed a heterozygote mutation at SNP 13, elevations in ASCA-IgA (34.3 EU/mL) and anti-CBir1 (79.8 EU/mL) levels, and a normal ASCA-IgG level (17.5 EU/mL). The test also showed a pANCA immunofluorescence antibody perinuclear pattern. The results predicted that the patient had a greater-than-80% chance of experiencing a serious complication within the next 5 years (Figure 5).

Due to her prior medication sensitivities, permission to initiate anti-TNF therapy was sought and obtained from her insurance company; this treatment was started in December 2010. Given the degree of clinical deterioration and the patient's poor nutritional status, the patient was also initiated on total parenteral nutrition (TPN) in January 2011. She was closely followed via monthly clinic visits and blood work, supplemented by weekly phone calls. At the patient's most recent follow-up visit, in March 2011, she appeared cheerful, weighed 54 lbs, and was much more active at home. She is still experiencing 2–4 loose stools daily, for which over-the-counter loperamide (taken before bedtime) has been recommended. TPN is being continued.

Discussion

Predicting the possible course of CD at the time of initial presentation has always been a challenge. Clinicians have traditionally used age of onset, requirement of steroids at initial presentation, history of smoking, and other clinical criteria to predict a particular patient's future course. Availability of serogenetic tests that can predict the probability of future complications is a novel development that will aid us in choosing the appropriate management for a particular patient.

In this patient, positivity for the SNP 13 mutation provided additional information to aid in the selection of the most appropriate treatment and helped to convince the family of its immediate need. The patient's sensitivity to other immunizations that contained foreign proteins and the fact that infliximab is the only biologic treatment approved for children made the decision very difficult for the family. With easy-to-understand prognostic data presented as a simple graph, the family could more readily realize the need for an immediate and strong treatment plan. These data also allowed us to more easily discuss these issues with the third-party insurance company and quickly obtain authorization for the use of certolizumab pegol and TPN. With the above measures, it appears that this patient is beginning to do better.

Whether early interventions with biologic therapy in patients with specific genetic mutations will reduce the rate of serious complications needs to be explored using long-term studies. Studies assessing the use of specific biologic therapies for specific mutations and their possible outcomes in CD patients will hopefully allow us to better customize treatments in the future.

Commentary

Marla C. Dubinsky, MD

The cases in this monograph illustrate several different clinical presentations—including ileocecal, ileocolonic, and small bowel–only disease—as well as serologic and genetic variations. As options for CD therapy have expanded, we need to not only be able to differentiate between these phenotypes but also start stratifying patients based on prognosis. Ideally, if we could stratify patients at diagnosis, then we could offer a clear-cut decision-making strategy for management of each case. In this model, everything we know about a patient—their serology and genetic results, CRP level, stool studies, endoscopic appearance, radiologic appearance, clinical symptoms, and other routine laboratory values—would help us to gauge the patient's risk. Given this assessment, we could then make more informed treatment decisions.

Specifically, we need to rapidly step up therapy or consider going straight to more effective interventions, such as biologics, if a patient has a higher-than-moderate risk of developing complications. Since the maximal efficacy of many CD therapies—particularly biologics—is achieved within the first 1–2 years, objective measures that allow us to tell patients why we need to move up the therapeutic spectrum more rapidly can yield better short-term and long-term therapeutic strategies.

Benefits of Serogenetic Testing

Serology and genetic information needs to be viewed in the context of the patient's clinical presentation and other laboratory tests, but sometimes patients like having an objective measure—beyond clinical symptoms or endoscopic appearance—that can help them make a decision about management. In addition to helping patients settle on a course of treatment, serology and genetic testing may also help to decrease variability in clinicians' treatment decisions. By focusing on objective measures and making management decisions based on this information, we should be able to manage patients more consistently than if we rely on clinical symptoms and laboratory tests alone.

Knowing a patient's genetic status is useful because genetics do not change, regardless of treatment or disease activity, and genetics play a major role in causing IBD. The *NOD2* gene, a susceptibility gene for CD, is found in approximately 30% of patients with CD. If a patient is *NOD2*-positive, that information can provide clues about the patient's disease phenotype, which can potentially help guide management decisions; for example, patients who are *NOD2*-positive tend to have more stricturing complications. If a test can detect an objective marker that is predictive of future complications, then I can say, for example, to a patient, "Given your genotype and the literature that supports the role of *NOD2* in CD, I really think you should stay on therapy and stop smoking."

I think that our biggest mistake in managing CD patients is that we often wait too long to decide whether a patient is a complicated case. We can readily react to complications after they occur, but we are not very good at being proactive. The whole point of developing a prognostic test—whether it is based on serogenetic markers, CRP levels, endoscopic or MRI changes, or a combination of these factors—is to get ahead of the curve and prevent complications.

Role of Serogenetic Testing in CD Management

The current Prometheus Crohn's Prognostic test was evaluated in a recent study that analyzed blood bank samples from well-characterized patients and then used logistic regression models to determine the probability of complications occurring over time.[1] Patients generally want to know how their disease will progress over the next 1–2 years, so a major goal of prognostic testing is to focus on the group of patients who are going to progress quickly. If we can identify these individuals, then we might be able to prevent complications by intervening early with effective therapies.

In a patient with very severe CD, clinicians do not need a prognostic test to determine whether the patient has complicating CD; clinical judgment alone is often sufficient to determine that these patients have very severe disease. The situation in which clinicians need serogenetic testing is when a patient appears to have a moderate risk of complications based on their endoscopy and laboratory results. In these cases, the questions the clinician typically asks are: "Did I catch them early in their disease course?" and "Are there any factors that would predict a more complicated course over a 5-year period of time that I would miss by relying on clinical presentation alone?"

In these cases, we want to be able to give patients an estimate of their disease risk based on standard criteria and then tell them how serogenetic testing further influences this risk. If serogenetic testing predicts an increased risk of complications, then we might want to go straight to more effective therapy in order to prevent complications. In general, serogenetic testing is most helpful in cases where the clinician is on the fence: patients who appear to have a moderate risk of complications and who may be candidates for early escalation of therapy but do not definitely require it.

The CD prognostic test includes *NOD2* mutations only at this point. However, in the era of genome-wide association studies, there will likely be additional genes that prove to be important in defining disease phenotypes. These advances will certainly facilitate enhancements to existing serogenetic tests.

Summary

Given the availability of biologic agents and other effective treatments for CD, I think clinicians should begin to be much more proactive in managing CD. With serogenetic testing, we have an opportunity to think more proactively and consider new information when deciding on a course of treatment. As with all our diagnostic tools—endoscopy, CRP measurements, laboratory tests, and stool studies—serogenetic tests need to be interpreted correctly, and the whole puzzle needs to be put together to determine the best course of action.

Reference

1. Lichtenstein GR, Targan SR, Dubinsky MC, et al. Combination of genetic and quantitative serological immune markers are associated with complicated Crohn's disease behavior. *Inflamm Bowel Dis*. 2011. In press.


G&H