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Age of Diagnosis Influences Serologic Responses in Children with Crohn Disease: A Possible Clue to Etiology?

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Abstract

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Crohn disease (CD) is often associated with antibodies to microbial antigens. Differences in immune response may offer clues to the pathogenesis of the disease.

AIM—To examine the influence of age at diagnosis on serologic response in children with CD.

METHODS—Data were drawn from 3 North American multicenter pediatric IBD research consortia. At or shortly after diagnosis, pANCA, ASCA IgA, ASCA IgG, anti-ompC and anti-CBir1 were assayed. Results were compared as a function of age at CD diagnosis (0–7 years vs 8–15 years).

RESULTS—705 children (79 <8 yr of age at diagnosis, 626 ≥8yr) were studied. Small bowel CD was less frequent in the younger group (48.7% vs 72.6%; $p<0.0001$) while colonic involvement was comparable (91.0% vs 86.5%). ASCA IgA and IgG were seen in <20% of those 0–7 yr compared to nearly 40% of those 8–15 yr ($p<0.001$), while anti-CBir1 was more frequent in the younger children (66% vs 54%, $p<0.05$). Anti-CBir1 detected a significant number of children in both age groups who otherwise were serologically negative. Both age at diagnosis and site of CD involvement were independently associated with expression of ASCA and anti-CBir1.

CONCLUSIONS—Compared to children 8–15 yr of age at diagnosis, those 0–7 yr are more likely to express anti-CBir1 but only half as likely to express ASCA. These age-associated differences in antimicrobial seropositivity suggest that there may be different, and as yet unrecognized, genetic, immunologic and/or microbial factors leading to CD in the youngest children.

Keywords

Crohn disease; Children; Serologic responses

INTRODUCTION

While the pathogenesis of Crohn's disease (CD) is thought to be multifactorial, disease expression commonly is characterized by the production of immune responses to a variety of microbial antigens. Detection of the serologic responses associated with CD has the potential to identify subgroups of patients at risk for aggressive or complicated disease (1–3). Identifying differences in immune response among large populations of patients at the time of initial diagnosis may also offer clues to the pathogenesis of the disease.

Previous studies have reported that 50–70% of patients with CD have detectable serologic markers to bacterial and yeast-derived antigens associated with *E. coli* and *Saccharomyces cerevisiae* (2,4). However, a sizable percentage of patients, including most young children, commonly do not have demonstrable abnormal responses to the most common serologic tests (5,6).

Recent studies have identified new serologic markers, including antibody responses to bacterial proteins such as flagellin, that characterize many patients with CD. Antibodies to CBir1 (anti-CBir1), a novel flagellin-like bacterial antigen, appear to be present in a subset of adults with CD, and may be a marker for identifying patients at risk for complicated disease (7–9).

This study was designed to explore the influence of age at diagnosis on serologic response in a population of well-characterized children with CD. In addition, we have used this analysis to better characterize the frequency of anti-CBir1 in the pediatric CD population, and to determine how inclusion of anti-CBir1 in the serologic panel affects the serologic characterization of these children. The data were drawn from the prospectively derived databases of three independent North American pediatric IBD collaborative research

consortia. The observations derived from these cohorts provide a unique opportunity to evaluate the clinical characteristics of children with IBD.

MATERIALS AND METHODS

Subject Inclusion

Data were drawn from the databases of the Pediatric IBD Collaborative Research Group Registry (Eastern US and Canada), the Western Regional Alliance for Pediatric IBD Research (Western US and Canada), and the Wisconsin Pediatric IBD Research Group (Midwest US). All three collaborative groups have prospectively recruited independent cohorts of newly diagnosed children with IBD after obtaining parental or legal guardian consent and the child's assent. Standardized data records including a subject's clinical and demographic characteristics are recorded at diagnosis and prospectively thereafter. All clinical investigators have obtained Institutional Review Board approval for the studies of their respective collaborative groups.

Subjects were selected for inclusion in the current analysis if they were diagnosed with CD before their sixteenth birthday, and had provided blood for serologic assessment at the time of or shortly after the initial diagnosis of CD. Only subjects who had a complete serologic assessment (see below) from their initial serologic specimen were included.

Clinical Assessment

The diagnosis of CD was made by generally accepted radiologic, endoscopic and histologic criteria (10). Each subject's site of CD involvement was categorized according to the following criteria:

1. Small bowel only: disease of the small bowel proximal to the cecum and distal to the ligament of Treitz
2. Large bowel only: any colonic location between cecum and rectum with no small bowel disease
3. Small and large bowel: disease of the small bowel and any location between cecum and rectum

In subsequent analysis, in addition to the 3 site of CD groups above, subjects were reclassified as having "any small bowel" CD by considering those children with "small bowel only" and "small and large bowel" as a single group. Similarly those with "large bowel only" and "small and large bowel" were grouped as "any large bowel" CD. For the purposes of this study, subjects with isolated upper digestive tract or perianal CD were excluded.

Serologic Assessment

A complete serologic assessment included determination of anti-neutrophil cytoplasmic antibody (pANCA), and antibodies directed against the outer membrane porin of *E. coli* (anti-ompC), *Saccharomyces cerevisiae* mannan (ASCA IgA and IgG), and a novel flagellin-like antigen (anti-CBir-1). Serologic analyses were performed by commercial assay (IBD 7 Panel®, Prometheus Laboratories, San Diego, CA) for all centers in the Pediatric IBD Collaborative Research Group or at the Immunobiology Institute at Cedars-Sinai Medical Center (Los Angeles, CA) for all centers in the Western Regional Alliance and Wisconsin Research Group. The same technology was utilized in both laboratories. Antibody levels were expressed as ELISA units (EU/ml). A positive serologic response for each marker was defined as a titer greater than the laboratory reference range for the specific

antibody assay. In cases where subjects were recruited before the availability of anti-CBir1 testing, banked sera from their initial specimen was used for subsequent anti-CBir1 analysis.

Statistical Analysis

To evaluate the effect of age at diagnosis on serologic response, the study population was divided into two groups based on their age at diagnosis: 0–7 years and 8–15 years. The age cut-off between the two groups was selected to maximize the number of subjects in the lower age group while taking into account the small number of very young children diagnosed with CD. Group differences were evaluated using t-tests for continuously-distributed variables and Fisher's Exact Test for categorical variables, including expression of the serologic markers. Odds ratios and 95% confidence intervals for risk factors associated with differences in serologic expression were also determined using a logistic regression. Differences were considered significant for a $p < 0.05$.

RESULTS

Seven hundred and five children had complete serologic assessments performed at the time of or shortly after the diagnosis of CD. Comparable numbers of subjects were drawn from each of the 3 participating consortia (Pediatric IBD Collaborative Group $n=227$, Western Alliance $n=267$, Wisconsin Research Group $n=211$). At diagnosis, 79 children were 0–7 years of age and 626 were 8–15 years of age. The clinical characteristics of the subjects are summarized in Table 1. Of note, the younger children were much more likely to have isolated colonic CD, while the older subjects more commonly had both small and large bowel involvement. The overall differences in site of CD distribution were highly significant between age groups ($p < 0.0001$). Overall, only 48.7% of those < 8 years of age at diagnosis had some degree of small bowel disease, compared to 72.6% of the older subjects ($p < 0.0001$). By contrast, there was no difference in the rate of any large bowel involvement between the two groups (91.0% in 0–7 yr old group, 86.5% in 8–15 yr olds, $p=NS$).

In the overall study population, the individual serologic markers varied widely in their frequencies. Only 16% of all children were positive for pANCA, while 17% were positive for anti-ompC. ASCA IgA and ASCA IgG were both found in 34% of population. By contrast, 56% were positive for anti-CBir1.

When the study population is divided by age at diagnosis, distinct differences between age groups appear (Figure 1). While rates of positive pANCA and anti-ompC remain low and comparable between age groups, both ASCA IgA and IgG antibodies occurred about twice as commonly in the older compared to the younger children ($p < 0.0001$). By contrast, positive anti-CBir1 assays were significantly more frequent in the younger children ($p < 0.05$). The differences in rates of positive ASCA and anti-CBir1 are particularly striking when the data are plotted yearly as a function of age at diagnosis (Figure 2).

The anti-CBir1 assay identified a large number of children who otherwise lacked any detectable serologic response. Fifty two percent of all children in the study population were serologically negative for pANCA, anti-ompC, ASCA IgA and ASCA IgG. The rate of serologically negative subjects fell to only 27% of the entire study population when the results of the anti-CBir1 assay were added to the serologic panel ($p < 0.0001$).

Age at diagnosis appeared to affect this phenomenon, as without the anti-CBir1 assay, the youngest children had the greatest proportion of seronegativity (Figure 3). Seventy percent of the children < 8 years of age at diagnosis were negative for pANCA, anti-ompC, ASCA IgA and ASCA IgG, compared to only 45% of those 8 years of age and older ($p < 0.0001$). The addition of the anti-CBir1 assay to the serologic panel significantly decreased the

number of seronegative children in both age groups, such that only 28% of those <8 years and 26% of those ≥8 yrs remained serologically negative.

A variety of logistic regression models were evaluated to further assess the influence of age at diagnosis as well as a number of additional clinical factors on the likelihood of expressing the different serologic markers. Selected results are summarized in Table 2. Younger age at diagnosis and small bowel site of CD but not gender appear to be independent factors positively influencing expression of anti-CBir1. By contrast, older age at diagnosis and small bowel CD independently increase risk of ASCA IgA and ASCA IgG expression. Age at diagnosis does not appear to increase the odds of expressing pANCA or anti-ompC.

DISCUSSION

This study represents the largest pediatric IBD population reported to date in which serologic responses to microbial antigens in children with Crohn's disease have been examined. Moreover, we have demonstrated that these responses are affected by age at diagnosis with possible implications for understanding the pathogenesis of this complex disease.

In adult populations with known CD, the frequency of ASCA (either IgG or IgA) has been reported to range from 28–59% (11–14), while that of anti-ompC ranges from 37–56% (15,16). Published investigations in children with CD have revealed somewhat higher rates of detectable ASCA (44–76%) (5,17,18) but a lower rate for anti-ompC (24%) (5). In the present study, the overall rates of anti-ompC and ASCA are lower than previous pediatric reports. However, the results are likely affected by the relatively large number of very young children in our study cohort, as the rate of anti-ompC rises to almost 20% and ASCA to 40% in the 8–15 year old subjects. pANCA, an ulcerative colitis associated marker, has been reported to be present in 10–20% of adults with “UC-like” CD (19–21). Our findings reveal a similar, if somewhat greater frequency, with nearly 30% of children of all ages with isolated colonic CD having detectable pANCA (data not shown).

In adult populations, anti-CBir1 antibodies appear to be a specific serologic marker for CD, having been identified in approximately 50% of adult CD patients but very few UC patients (7–9). In our overall population, 56% of children less than 16 years of age at diagnosis had detectable anti-CBir1. At all age groups, anti-CBir1 appears to identify a significant subpopulation of children who otherwise would be characterized as serologically negative. It therefore appears that the addition of the anti-CBir1 assay to a serologic panel should enhance the sensitivity of testing when serology is used as a diagnostic test to identify children likely to have IBD. This speculation must be explored in additional studies, however, as our study population only includes children with documented CD.

For ASCA and anti-CBir1, the age at diagnosis appears to be an important factor affecting the likelihood of identifying positive serologic responses in children with CD. ASCA is rare in the younger children, while anti-CBir1 is particularly common. These observations may be particularly important clues toward understanding the pathogenesis and early immunologic dysfunction that leads to the development of CD.

CD appears to arise from a dysregulated immune response to the endogenous enteric microflora in a genetically susceptible host (22). Normal innate immune responses recognize microbial antigens such as muramyl dipeptide (a component of lipopolysaccharide), resulting in controlled responses by gut associated immunoreactive cells. Defective innate immune responses, such as those resulting from allelic variants of NOD2/CARD15 or various toll-like receptor (TLR) genes, cause a cascade of events that ultimately are expressed as chronic intestinal inflammation (22,23).

The CBir1 antigen has been shown to have a high degree of homology to bacterial flagellins, resembling known flagellins from enteric microbiota including *Butyrivibrio*, *Roseburia*, *Thermotoga*, and *Clostridium* species (7). Antibodies against the CBir1 antigen have been identified in a number of different mouse models of IBD, including one whose underlying genetic predisposition for IBD causes defective epithelial barrier function and a number of others characterized by different defective regulatory T-cell functions (7).

The serologic patterns identified in the different age groups reported in the present investigation strongly suggest that the host microbial interactions underlying the development of CD at varying ages are different. As a group, children less than 8 years of age at diagnosis are predominantly anti-CBir1 positive, ASCA and anti-ompC negative, while those older than 8 years of age are more commonly both ASCA and anti-CBir1 positive. These observations suggest that there may be different underlying genetic defects in children who present at different ages. Which specific genes might be most important in the youngest children requires additional study. Our data suggest that the Toll-like receptor (TLR) gene family may be critically important to investigate, as TLR-5 has been reported to mediate the normal innate immune response to bacterial flagellin (24). In addition, a recently described linkage between a specific haplotype of the NFkB1 gene and expression of anti-CBir1 identifies another potential gene of particular interest in this population (25).

Another potentially fertile area for additional investigation arises from the alternative hypothesis that the enteric microbes central to the development of CD differ in younger compared to older children. The high frequency of anti-CBir1 antibodies in the children less than 8 years of age at diagnosis suggests that microbes such as *Clostridia* may play an important role in precipitating CD in the youngest children, whereas yeast and *E. coli*, the organisms whose antigens are associated with ASCA and anti-ompC, may play a more important role in children who present at an older age.

In summary, our data demonstrate that the anti-CBir1 assay identifies a subgroup of children with CD who would otherwise not be characterized serologically. This appears especially important in the youngest children with CD, in whom anti-CBir1 appears to be particularly common. These data suggest, but do not prove, that the addition of the anti-CBir1 assay to a panel of markers including ASCA, anti-ompC and pANCA may increase the sensitivity of serology as a screening tool for the diagnosis of CD in children. The age-associated differences in the patterns of antimicrobial seropositivity demonstrated in the present study also suggest that there may be different, and as yet unrecognized, genetic, immunologic and/or microbial factors leading to CD in the youngest children.

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References

1. Ferrante M, Henckaerts L, Joossens M, et al. New serological markers in inflammatory bowel disease are associated with complicated disease behaviour. *Gut*. 2007 Oct; 56(10):1394–403. [PubMed: 17456509]
2. Papp M, Altorjay I, Norman GL, et al. Seroreactivity to microbial components in Crohn's disease is associated with ileal involvement, noninflammatory disease behavior and NOD2/CARD15

- genotype, but not with risk for surgery in a Hungarian cohort of IBD patients. *Inflamm Bowel Dis*. 2007 Aug; 13(8):984–92. [PubMed: 17417801]
3. Dubinsky MC, Lin YC, Dutridge D, et al. Serum immune responses predict rapid disease progression among children with Crohn's disease: immune responses predict disease progression. *Am J Gastroenterol*. 2006; 101(2):360–7. [PubMed: 16454844]
 4. Israeli E, Grotto I, Gilburd B, et al. Anti-Saccharomyces cerevisiae and antineutrophil cytoplasmic antibodies as predictors of inflammatory bowel disease. *Gut*. 2005 Sep; 54(9):1232–6. [PubMed: 16099791]
 5. Zhuludev A, Zurkowski D, Young W, et al. Serologic testing with ANCA, ASCA, and anti-ompC in children and young adults with Crohn's disease and ulcerative colitis: diagnostic value and correlation with disease phenotype. *Am J Gastroenterol*. 2004; 99(11):2235–41. [PubMed: 15555007]
 6. Gupta SK, Fitzgerald JF, Croffie JM, et al. Comparison of serological markers of inflammatory bowel disease with clinical diagnosis in children. *Inflamm Bowel Dis*. 2004 May; 10(3):240–4. [PubMed: 15290918]
 7. Lodes MJ, Cong Y, Elson CO, et al. Bacterial flagellin is a dominant antigen in Crohn disease. *J Clin Invest*. 2004; 113:1296–1306. [PubMed: 15124021]
 8. Papadakis KA, Yang H, Ippolitti A, et al. Anti-flagellin (CBir1) phenotypic and genetic Crohn's disease associations. *Inflamm Bowel Dis*. 2007; 13(5):524–530. [PubMed: 17260364]
 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. *Gastroenterol*. 2005; 128(7):2020–2028.
 10. Bousvaros A, et al. North American Society for Pediatric Gastroenterology, Hepatology, and Nutrition; Crohn's and Colitis Foundation of America. Differentiating ulcerative colitis from Crohn disease in children and young adults: report of a working group of the North American Society for Pediatric Gastroenterology, Hepatology, and Nutrition and the Crohn's and Colitis Foundation of America. *J Pediatr Gastroenterol Nutr*. 2007 May; 44(5):653–74. [PubMed: 17460505]
 11. Riis L, Vind I, Vermeire S, et al. The prevalence of genetic and serologic markers in an unselected European population-based cohort of IBD patients. *Inflamm Bowel Dis*. 2007; 13(1):24–32. [PubMed: 17206636]
 12. Desplat-Jego S, Johanet C, Escande A, et al. Update on anti-Saccharomyces cerevisiae antibodies, anti-nuclear associated anti-neutrophil antibodies and antibodies to exocrine pancreas detected by indirect immunofluorescence as biomarkers of chronic inflammatory bowel disease: results of a multicenter study. *World J Gastroenterol*. 2007; 13(16):2312–8. [PubMed: 17511029]
 13. Saibeni S, Folli C, de Franchis R, et al. Diagnostic role and clinical correlates of anti-Saccharomyces cerevisiae antibodies (ASCA) and anti-neutrophil cytoplasmic antibodies (ANCA) in Italian patients with inflammatory bowel disease. *Dig Liver Dis*. 2003; 35(12):862–8. [PubMed: 14703881]
 14. Sandborn WJ, Loftus EV, Colombel JF, et al. Evaluation of serologic disease markers in a population-based cohort of patients with ulcerative colitis and Crohn's disease. *Inflamm Bowel Dis*. 2001; 7(3):192–210. [PubMed: 11515844]
 15. Arnott ID, Landers CJ, Nimmo EJ, et al. Sero-positivity to microbial components in Crohn's disease is associated with disease severity and progression, but not NOD2/CARD15 genotype. *Am J Gastroenterol*. 2004; 99(12):2376–84. [PubMed: 15571586]
 16. Landers CJ, Cohavy O, Misra R, et al. Selected loss of tolerance evidenced by Crohn's disease-associated immune responses to auto- and microbial antigens. *Gastroenterol*. 2002; 123(3):689–99.
 17. Khan K, Schwarzenberg SJ, Sharp H, et al. Role of serology and routine laboratory tests in childhood inflammatory bowel disease. *Inflamm Bowel Dis*. 2002; 8(5):325–9. [PubMed: 12479647]
 18. Bartunkova J, Kolarova I, Sediva A, Holzelova E. Antineutrophil cytoplasmic antibodies, anti-Saccharomyces cerevisiae antibodies, and specific IgE to food allergens in children with inflammatory bowel diseases. *Clin Immunol*. 2002; 102(2):162–8. [PubMed: 11846458]

19. Vasiliauskas EA, Plevy SE, Landers CJ, et al. Perinuclear antineutrophil cytoplasmic antibodies in patients with Crohn's disease define a clinical subgroup. *Gastroenterology*. 1996; 110:1810–9. [PubMed: 8964407]
20. Preda CM, Vermeire S, Rutgeerts P, et al. Prevalence and significance of perinuclear anti-neutrophil antibodies (pANCA) in Romanian patients with Crohn's disease and ulcerative colitis. *Rom J Gastroenterol*. 2005 Dec; 14(4):357–60. [PubMed: 16400351]
21. Freeman H, Roeck B, Devine D, Carter C. Prospective evaluation of neutrophil autoantibodies in 500 consecutive patients with inflammatory bowel disease. *Can J Gastroenterol*. 1997 Apr; 11(3): 203–7. [PubMed: 9167026]
22. Strober W, Fuss I, Mannon P. The fundamental basis of inflammatory bowel disease. *J Clin Invest*. 2007; 117:514–521. [PubMed: 17332878]
23. Begue B, Dumant C, Bambou JC, et al. Microbial induction of CARD15 expression in intestinal epithelial cells via toll-like receptor 5 triggers an antibacterial response loop. *J Cell Physiol*. 2006 Nov; 209(2):241–52. [PubMed: 16897777]
24. Hayashi F, Smith KD, Qzinsky A, et al. The innate immune response to bacterial flagellin is mediated by Toll-like receptor 5. *Nature*. 2001; 410(6832):1099–1103. [PubMed: 11323673]
25. Takedatsu H, Taylor KD, Mei L, et al. Linkage of CD-related serological phenotypes: NFKB1 haplotypes are associated with anti-CBir1 & ASCA, and show reduced NF-kB activation. *Gut*. 2008 Oct 2. [Epub ahead of print].

Figure 1.
 Effect of Age at Diagnosis on Percent of Subjects with Positive Serologic Responses
 * $p < 0.0001$
 # $p < 0.05$

Figure 2.
Percent of Subjects with Positive ASCA IgA and anti-CBir1 by Year of Age at Diagnosis

Figure 3.

Effect of anti-CBir1 Assay on the Rate of Seronegativity

A vs B: $p < 0.0001$

C vs D: $p = \text{NS}$

A vs C: $p < 0.0001$

B vs D: $p < 0.0001$

Table 1

Characteristics of the Study Population

	Overall Study Population	0–7 yrs	8–15 yrs
n	705	79	626
% male	59%	58%	59%
Mean Age $\pm$ SD (yrs)	11 $\pm$ 3	5 $\pm$ 2*	12 $\pm$ 2*
Median Age (yrs)	12	6	12
Site of CD: n [^] (%)			
Small bowel only	91 (13.1%)	7 (9.0%) [#]	84 (13.6%) [#]
Large bowel only	210 (30.1%)	40 (51.3%) [#]	170 (27.5%) [#]
Small and Large bowel	396 (56.8%)	31 (39.7%) [#]	365 (59.0%) [#]
Any small bowel	487 (69.9%)	38 (48.7%) ^{**}	449 (72.6%) ^{**}
Any large bowel	606 (86.9%)	71 (91.0%) [@]	535 (86.5%) [@]

[^] Site of CD involvement missing in 8 subjects

* 0–7 yrs vs 8–15 yrs: $p < 0.0001$

[#] Overall distribution of site of CD (0–7 yrs vs 8–15 yrs): $p < 0.0001$

^{**} Any small bowel compared to large bowel only (0–7 yrs vs 8–15 yrs): $p < 0.0001$

[@] Any large bowel compared to small bowel only (0–7 yrs vs 8–15 yrs): $p = \text{NS}$

Table 2Selected results of logistic regression, estimate of odds ratio[^] (95% confidence interval)

Outcome	Age Group (0–7 vs 8–15 yrs) [^]	Site of CD	Site risk group [^]	Gender (female vs male) [^]
pANCA	1.31 (0.74–2.33)	3.31 (1.41–7.78) ^{**}	Any large bowel vs small bowel (C+IC vs I)	1.92 (1.30–2.83) ^{**}
ASCA IgA	0.30 (0.15–0.59) ^{**}	2.53 (1.73–3.70) ^{**}	Any small bowel vs large bowel (I+IC vs C)	0.96 (0.70–1.33)
ASCA IgG	0.40 (0.21–0.75) ^{**}	3.20 (2.15–4.77) ^{**}	Any small bowel vs large bowel (I+IC vs C)	1.17 (0.85–1.63)
OmpC	0.64 (0.32–1.28)	1.21 (0.78–1.87)	Any small bowel vs large bowel (I+IC vs C)	1.27 (0.87–1.87)
CBir1	1.94 (1.17–3.23) [*]	1.75 (1.25–2.45) ^{**}	Any small bowel vs large bowel (I+IC vs C)	1.13 (0.83–1.54)

*
p<.05;**
p<.01; patients with missing data excluded[^]
odds of positive serology for risk group vs reference group as noted

C = colon only

I = small bowel only

IC = both small and large bowel