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Human Antichimeric Antibody in Children and Young Adults with Inflammatory Bowel Disease Receiving Infliximab

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ABSTRACT

Introduction: Pediatric studies on immunogenicity of infliximab have not been published. The aim of the study was to evaluate the prevalence of human antichimeric antibody (HACA), relationship to infusion reactions (IR), and the role of concomitant immunomodulatory therapies.

Methods: An inflammatory bowel disease (IBD) database was queried, and a retrospective review of patients who had HACA performed was undertaken.

Results: HACA was conclusively determined in 34 patients with IBD (14 male, Crohn disease/ulcerative colitis: 30/4), median age 14.8 years (range, 6.4–22.5 years). Twenty-nine (85.3%) patients were receiving immunomodulatory therapy. A total of 234 infliximab infusions were administered (mean, 6.9; range, 1–26). HACA was detected in 12 (35.3%) patients. IR occurred in 8 (23.5%) patients. HACA-positive patients had a higher proportion of infusions associated with IR than did HACA-negative patients ($P < 0.01$). HACA levels $\geq$ 8.0 μg/mL were more likely to be associated with IR ($P = 0.03$). Levels of $\geq$ 8.0 μg/mL were more common in patients who had an average interval between infliximab infusions of 8 weeks or less ($P = 0.04$). Concomitant immunomodulatory therapy was associated with a lower risk of developing HACA ($P = 0.02$) and lower titer of HACA ($P = 0.04$). Patients did not have HACA at a greater rate when there was an extended interval (more than 12 weeks) between infliximab infusions ($P = 0.89$).

Conclusions: In children and adolescents with IBD, HACA formation is related to IR and to the duration of response to treatment. Immunomodulatory agents seem to have a protective role against development of HACA or high titers of antibodies. The interval between infusions does not influence the development of HACA. ***JPGN 38:502–508, 2004.*** **Key Words:** Human antichimeric antibody—Inflammatory bowel disease—Infliximab. © 2004 Lippincott Williams & Wilkins

Inflammatory bowel disease (IBD) is the most common chronic gastrointestinal illness in children and adolescents. The true incidence and prevalence of IBD in children is unknown, but 25% to 30% of all patients with Crohn disease (CD) and 20% of those with ulcerative colitis (UC) experience the disorders before the age of 20 years (1). The age-specific incidence rates in North America for 10- to 19-year-olds are approximately 2/100,000 for UC and 3.5/100,000 for CD (1).

The standard therapies for CD and UC are similar and in general can be classified as anti-inflammatory or immunomodulatory therapy. Five-aminosalicylic acid compounds, antibiotics, and nutritional therapy usually are considered as anti-inflammatory, whereas steroids, 6-mercaptopurine, azathioprine, cyclosporine, and methotrexate have immunomodulatory properties.

Furthermore, newer biologic agents have been developed that target specific cytokines in the immune system. Tumor necrosis factor (TNFα) is a proinflammatory cytokine that is proximal in the cytokine cascade and mediates the production of other inflammatory cytokines (2). Infliximab is a genetically engineered monoclonal antibody against TNF-α. It is a chimeric immunoglobulin, 75% human and 25% murine origin (2). Infliximab has been approved for the treatment of moderately to severely active CD in patients who have an inadequate response to conventional therapy, and for therapy of fistulizing CD (2). Recent studies also suggest effectiveness of infliximab in the treatment of UC, including medically refractory severe disease (3,4).

The safety and effectiveness of infliximab for pediatric patients with CD has been investigated by several authors (5–7). In our experience, infliximab was well tolerated for multiple infusions, and infusion reactions (IR) were seen in 5.3% of 432 infusions, or 14.6% of 82

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patients (5). This corresponds to reports in adult patients, in whom IR were seen in 4% to 13% of infusions and in 17% to 27% of the study populations (8–10).

A recent study in adult patients with IBD showed that IR are associated with the formation of human antichimeric antibody (HACA), which may shorten the duration of effect of repeated infliximab treatments (8,10). HACA were detected in 61% of adult patients receiving infliximab on "demand schedule" (10). This and several other studies also suggested that treatment with immunomodulatory agents can prevent IR and help maintain clinical efficacy (8,10,11). To evaluate the efficacy of corticosteroids to prevent HACA formation, a randomized controlled trial demonstrated that intravenous hydrocortisone premedication significantly reduces HACA levels but does not significantly reduce HACA formation or IR (12).

Pediatric studies on immunogenicity of infliximab therapy for IBD have not yet been published. The aim of our study was to investigate the prevalence of HACA, the relationship between HACA and infusion reactions, and the role of concomitant immunosuppressive therapies in the formation of HACA in children and young adults with IBD receiving infliximab therapy.

METHODS

An IBD database was queried to identify patients receiving infliximab therapy. The database is maintained within the Center for Pediatric Inflammatory Bowel Disease and has been approved by the Institutional Review Board of The Children's Hospital of Philadelphia. A retrospective review of all records of patients who had HACA performed between January 1, 2002, and May 1, 2003, was undertaken. Additional information was obtained through review of computerized medical records and hard copy patient charts and by contacting the physician of record. Data gathered included demographic information, infliximab and HACA level, number and dose of infliximab infusions, side effects (including IR), and the use of concomitant medications. Documented delayed hypersensitivity reactions and infections were also recorded. Treatments administered for IR (and premedication) were recorded. An IR was defined as any significant adverse event that occurred during the infusion or within 2 hours afterward. A serious IR was defined as any reaction that required infliximab to be discontinued (12). When an IR occurred, the infusion was stopped and restarted at a slower rate. If the symptoms recurred, intravenous corticosteroids and/or intravenous diphenhydramine and/or oral acetaminophen were given. This regimen was then given prophylactically 30 minutes before each subsequent infusion.

We defined concurrent immunomodulatory therapy by use of either prednisone $\geq$1 mg/kg/day or azathioprine (1.5–2.5 mg/kg/day), 6 mercaptopurine (1–1.5 mg/kg/day), or methotrexate (15–25 mg/m^2/weekly) for at least 3 months at the time of HACA evaluation.

Infliximab was periodically administered with intravenous 5 mg/kg infusions, as previously described (5). A dose of 10 mg/kg of infliximab was given to patients with loss of efficacy after multiple doses of 5 mg/kg. Indications for the use and the decision to re-treat with infliximab were made at the discretion of the prescribing physicians and did not follow a set infusion schedule.

Quantitative HACA and infliximab enzyme-linked immunosorbent assays on serum (Prometheus Laboratories, San Diego, CA, U.S.A.) were measured in duplicate in a blinded fashion as described previously (10). HACA were reported as negative when the concentration was less than 1.69 μg/mL and the serum infliximab concentration was less than 1.40 μg/mL; and as indeterminate when the concentration was less than 1.69 μg/mL but the infliximab concentration was 1.40 μg/mL or greater because the presence of infliximab interferes with the HACA assay. The HACA test was considered to be positive when the concentration exceeded 1.69 μg/mL and the infliximab concentration was less than 1.40 μg/mL (10). In this series, concentrations of infliximab and HACA typically were evaluated before the infusion of infliximab.

Statistical Analysis

Comparisons between groups were made using the χ^2 test and Fisher exact test for categorical variables and the Student's *t* test for continuous variables. When there was concern regarding the normality of a continuous variable, the Mann-Whitney *U* test for nonparametric data was used. Statistical significance was defined as a *P* value of 0.05 or less. Statistical analyses were made using the software package Stata 7 (Stata Corporation, College Station, TX, U.S.A.).

RESULTS

During the study period, 132 patients with IBD received a total of 621 infusions. Fifty-six patients had HACA assays sent. Twenty-two patients with CD had indeterminate assays because of detectable circulating infliximab and were excluded. HACA was conclusively determined in the remaining 34 patients. Patient and disease characteristics are summarized in Table 1. Patients who had HACA sent were similar to those who did not have HACA sent during this time period with regard to demographic, disease, and treatment, with the exception of immunomodulator use, which was slightly more prevalent in those who did not have HACA performed (96% *v* 85%; $P = 0.03$).

HACA were detected in 12 (35%) patients (M/F: 4/8; CD/UC: 10/2). There was a trend toward a lower rate of HACA in patients younger than 14 years of age (2/12 *v* 10/22; $P = 0.13$) (Fig. 1). A total of 18 IR occurred in 8 (23.5%) patients. A higher proportion of patients with HACA had IR (4/12) than did those without HACA (4/22), but this finding did not meet statistical significance ($P = 0.40$). However, IR occurred in a higher proportion of infusions given to patients with positive HACA than patients with negative HACA (13.8% of 94 infusions *v* 3.6% of 140 infusions, respectively; $P < 0.01$).

The serum concentrations of HACA had two clusters that could be separated with the use of 8.0 μg/mL as a

TABLE 1. *Patient and disease characteristics of 34 children and young adult patients with inflammatory bowel disease at the time of HACA evaluation*

Characteristic	Value
Age (years)	
Median	14.8
Range	6.4–22.5
Gender, no. of patients (%)	
Male	14 (41.2)
Female	20 (58.8)
Type of disease, no. of patients (%)	
Crohn disease (CD)	30 (88.2)
Ulcerative colitis (UC)	4 (11.8)
Disease duration (years)	
Median	4.6
Range	2–17.6
Involved intestinal areas in 4 patients with UC, no. of patients (%)	
Left colon	0 (0)
Pancolitis	4 (100)
Involved intestinal areas in 30 patients with CD, no. of patients (%)	
Ileum	3 (10)
Colon	10 (33.3)
Ileum and colon	17 (56.7)
Gastroduodenum	4 (13.3)
Concomitant medications, no. of patients (%)	
Corticosteroids	8 (23.5)
Prednisone	5 (62.5)
Budesonide	3 (37.5)
Immunomodulatory agents	29 (85.3)
Azathioprine or 6-mercaptopurine	26 (89.7)
Methotrexate	2 (6.9)
Cyclosporine	1 (3.4)
Mesalamine	27 (79.4)
Antibiotics	8 (23.5)
Metronidazole	5 (62.5)
Ciprofloxacin	3 (37.5)
Probiotics	3 (8.8)
None	1 (2.9)
No. of infliximab infusions	234
Mean infusions per patient	6.9
Range	1–26

cutoff value, as reported in adult subjects (10). Patients with HACA levels 8.0 μg/mL or higher were more likely to experience IR (relative risk, 3.9; 95% confidence interval, 1.3 to 11.7; $P = 0.04$) (Fig. 2). A trend toward higher HACA levels (8.0 μg/mL or higher) was seen in patients with an interval between infliximab infusions of 8 weeks or less ($P = 0.08$) (Fig. 3).

Although patients with IR received pretreatment with a combination of diphenhydramine, acetaminophen, and/or corticosteroids before subsequent infusions, recurrent IR recurred nine times in one HACA-positive patient and one time in an HACA-negative patient. The concentration of antibodies in the HACA-positive subject with recurrent IR was higher than 26.33 μg/mL, which is the maximum value detectable by the described method. The characteristics of IR are reported in Table 2. Reduced infusion rates were used to complete doses when reactions occurred. The rate used in these situations was one step lower than the rate being used when the reaction occurred. Treatments administered for these reactions were diphenhydramine (75%), acetaminophen (62.5%), and intravenous corticosteroid (37.5%). The same regimen was then given prophylactically 30 minutes before each subsequent infusion. No serious adverse events, infections, malignancies, or delayed hypersensitivity reactions were documented in this group of patients. However, it should be mentioned that patients were not routinely contacted to detect delayed hypersensitivity reactions, and the lack of reported delayed reactions does not conclusively demonstrate that none occurred.

Twenty-nine subjects (85.3%) were receiving immunomodulatory therapy (Table 1). Concomitant immunomodulatory therapy was associated with a lower risk of developing HACA (relative risk: 0.34; 95% confidence interval, 0.17–0.72; $P = 0.02$) (Fig. 4). Immunomodulatory agents also protected against high titer of antibodies. Among patients taking immunomodulatory agents, the mean concentration of antibodies was 9.7 μg/mL (95% confidence interval, 0.9–18.5) as compared with 23 μg/mL (95% confidence interval, 17.6–28.4) in patients who were not taking immunomodulatory medicines ($P = 0.04$) (Fig. 5).

Patients did not develop HACA at a greater rate when there was an extended interval (more than 12 weeks) between infliximab infusions ($P = 0.89$).

DISCUSSION

Infliximab is an immunoglobulin (IgG1) monoclonal chimeric antibody to TNF-α, a cytokine thought to be one of the principal mediators of the inflammatory response in patients with CD. Approved for open label use by Food and Drug Administration in 1998, infliximab is now being used by a number of pediatric centers as an alternative to corticosteroids in children with moderate to severe CD, and most recently in children with UC (4,6). This agent has found application in children who are corticosteroid dependent or have experienced corticosteroid toxicity and for children with disease refractory to, or developing complications from, standard immunomodulatory regimens (5,6). To date, clinical experience with infliximab has been reassuring regarding safety in pediatric and adult patients, and clinical response rates have mirrored the efficacy reported in controlled trials (6,9,13).

Immunogenicity is an emerging issue, which has been demonstrated in adult patients to significantly limit the long-term efficacy of infliximab and to be related to IR through the formation of HACA (10,12). In adult patients receiving infliximab therapy, the true incidence of HACA formation remains unknown. Although the large ATTRACT trial in rheumatoid arthritis and the ACCENT1 trial in CD reported an overall incidence of

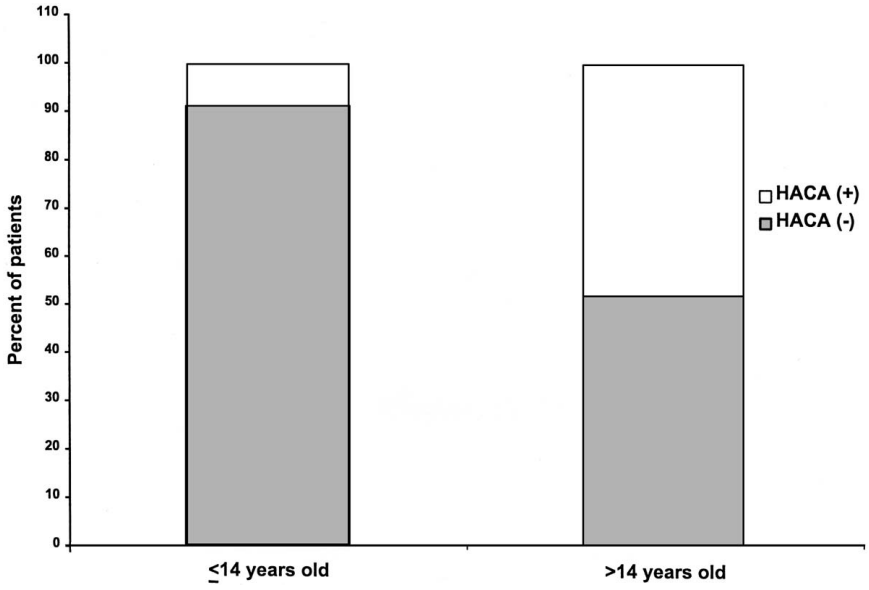

FIG. 1. Ratio of HACA in patients with inflammatory bowel disease older and younger than 14 years of age.

HACA formation in 8% to 14% of patients, Baert et al. (10) reported HACA formation in 68% of a cohort of adult patients with CD who received multiple infliximab infusions on a "demand schedule" (8,14). Most recently, in a prospective observational study, 36% of adult patients with CD receiving infliximab developed HACA (12).

For the first time, we evaluated the prevalence of HACA and the relationship between HACA and IR, as well as the role of concomitant immunomodulatory therapies in a cohort of children and young adults with IBD receiving infliximab periodically, to prevent the exacerbation of the disease. Although HACA analysis in

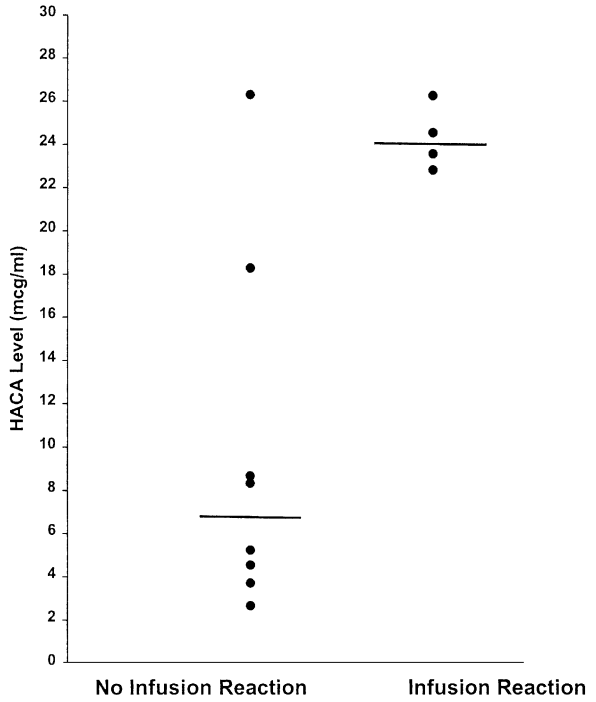

FIG. 2. Relationship between HACA concentration and infusion reaction (lines represent the median value). Higher concentrations of HACA are related to infusion reaction ($P = 0.04$)

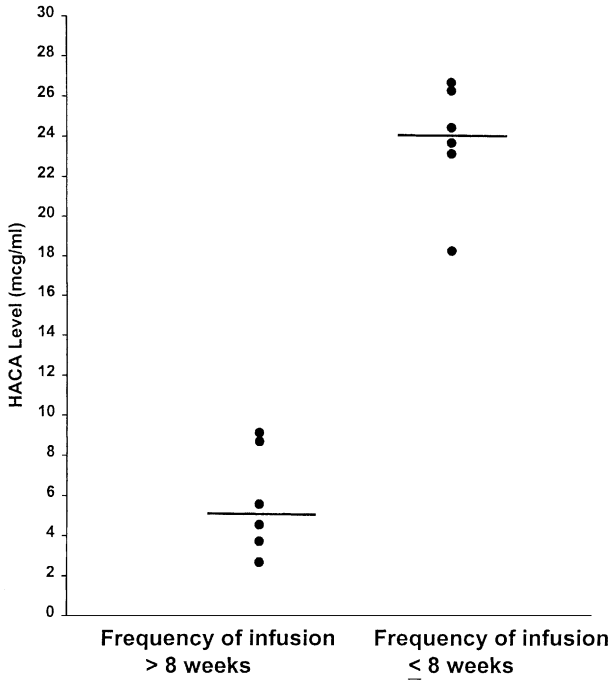

FIG. 3. Relationship between HACA concentration and frequency of infliximab infusion (the lines represent the median value). A tendency toward higher concentrations of HACA are seen with a shorter interval between infliximab infusions ($P = 0.08$).

TABLE 2. *Type of infusion reactions in 8 patients with inflammatory bowel disease receiving infliximab*

Symptoms	No. of Patients
Dyspnea	4
Flushing	4
Chest pain	3
Nausea	3
Hypotension	2
Urticaria/Pruritus	2
Headache	1
Abdominal pain	1
Fever	1

this group was not performed by protocol, we believe the similarities between the patients who received HACA analysis during this time period and those who did not implies that the cohort is representative of our general IBD population.

The overall prevalence of HACA in our series of patients was 35.3%, similar to that reported by Farrell et al. (12). However, we observed a trend toward a lower prevalence of HACA in patients younger than 14 years of age ($P = 0.13$). Because the relationship between HACA and IR has been well described in adult patients (10,12), our data may be supported by a previous report by Kugathasan et al. (15), in which adult patients, rather than pediatric patients younger than 17 years, experienced the majority of severe adverse reactions. Based on our findings, an explanation could be a lower prevalence of HACA in children. However, this hypothesis should be prospectively studied on a larger number of patients to define a possible protective role of young age on the development of HACA.

As reported in adult patients (10), we found an increased risk of IR in HACA-positive children and young adults, which correlated with antibody concentration. Our data show that prophylactic premedication with a combination of intravenous antihistamine and corticosteroid seems to be effective in preventing IR in HACA-positive patients with a low level of antibodies. So far, there are no data from randomized controlled trials to establish the optimal strategy for minimizing HACA formation. Several adult studies suggest that an optimal strategy is to use three-dose induction therapy at 0, 2, and 6 weeks, followed by scheduled (every 8 weeks) maintenance therapy (8,16,17). Our findings suggest that this strategy may be not necessary in pediatric and young adult patients. In fact, a long interval between infusions does not influence the development of HACA in our study population. In addition, in the mentioned study of Kugathasan et al. (15), children with IBD seemed to tolerate episodic infliximab re-treatment without developing IR.

In adult patients it has been demonstrated that a loss of initial response is strongly related to HACA formation and HACA level (8,10). We found that high levels of HACA were more common in patients who had shorter intervals of infusion. We can postulate that higher HACA levels may lead to more rapid clearance of infliximab, necessitating more frequent infusions. However, it is also possible that more frequent infusions result in higher HACA titers. It should also be noted that more frequent infliximab dosing could increase the likelihood of detecting circulating infliximab at the time of measurement, leading to an indeterminate assay.

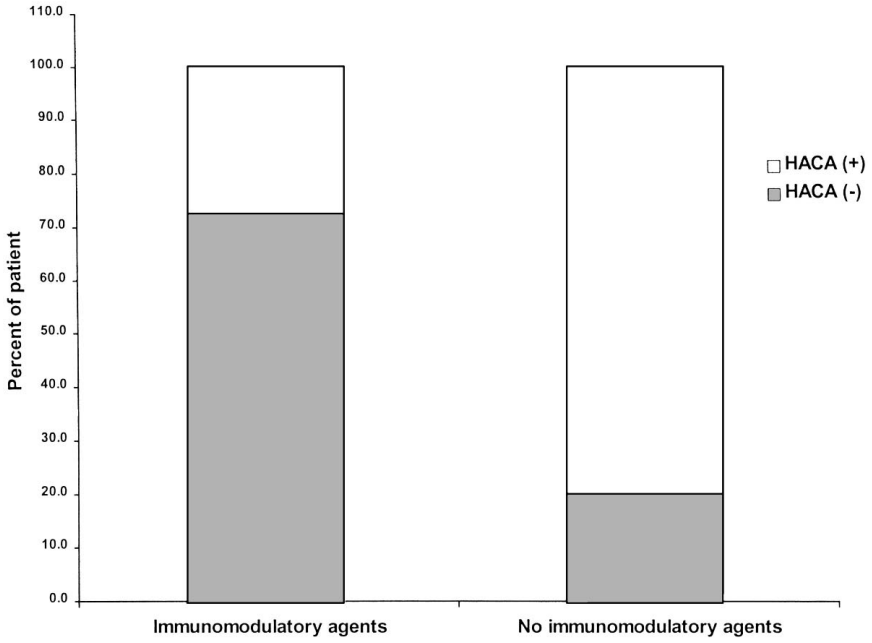

FIG. 4. Relationship between prevalence of HACA and the use of immunomodulatory therapy. The prevalence of HACA was significantly lower ($P = 0.02$) among patients who were taking immunomodulatory therapy.

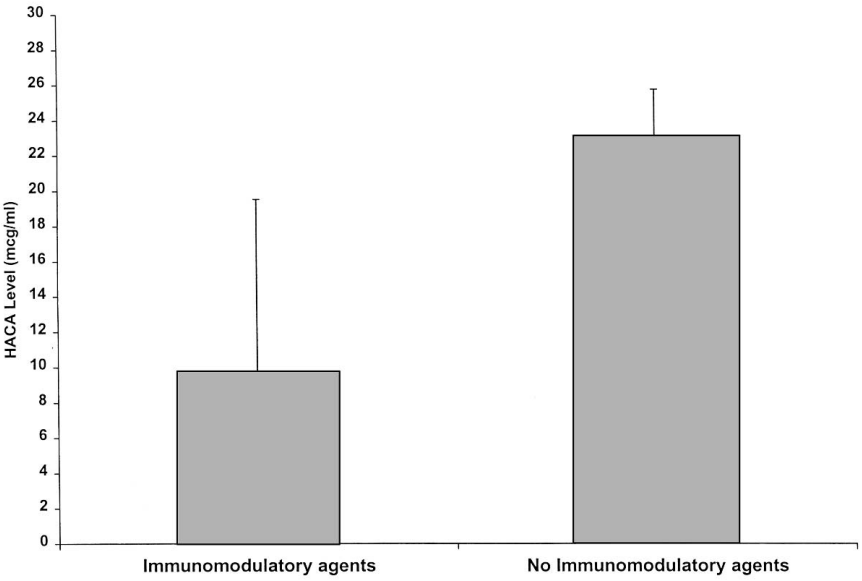

FIG. 5. Relationship between mean HACA concentration and the use of immunomodulatory therapy. The mean concentration of HACA was significantly lower (P = 0.04) among patients who were taking immunomodulatory therapy (the T bars represent the standard deviation from the mean).

The concurrent use of immunomodulatory therapy has been shown to maintain a favorable clinical response to infliximab therapy and to prevent HACA formation in adult patients (8,10,12,16). Based on our data, immunomodulatory agents seem to have a protective role against development of HACA, or at least against high titers of antibodies, in children and young adults. When possible, we advise immunomodulatory treatment for a clinically relevant period of time with azathioprine or 6 mercaptopurine (2–3 months) or methotrexate (1.5–2 months) before initiation of infliximab therapy and subsequent long-term continuation of the same immunomodulatory agent as concomitant therapy with infliximab. Additional studies need to determine which is the best immunomodulatory agent for pediatric patients. In a recent retrospective study, azathioprine has been demonstrated a safe and well-tolerated maintenance therapy for children with IBD (18). So far, there are no data on the interaction between azathioprine and infliximab. However, a drug interaction between methotrexate and infliximab has been proposed in adult patients with rheumatoid arthritis, for which methotrexate has been shown to reduce the clearance of infliximab (19).

In summary, our study offers new information regarding the influence of immunogenicity on infliximab treatment in children and young adults with IBD. Young age may be protective against the development of HACA. HACA formation is related to the IR and to the duration of response to treatment, as seen in adult patients. Immunomodulatory agents seem to have a protective role against development of HACA or high titers of antibodies. The use of premedication before the infliximab infusion seems to be effective to avoid IR in patients with low HACA levels. In contrast to the adult experience, the interval between infusions does not influence the development of HACA in the pediatric and young adult population with IBD, and frequent treatments with infliximab for this reason might not be necessary. In addition, prospective studies need to verify if other newer humanized or fully human therapeutic antibodies may be less immunogenic and if a young age or different protocols of premedication can be protective against the development of antibodies.

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