CLINICAL STUDIES

Seroprevalence of *Schistosoma mansoni* in Puerto Ricans with Inflammatory Bowel Disease

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Background. **The etiology of Inflammatory Bowel Diseases, Crohn's disease (CD) and ulcerative colitis (UC), is unknown. These diseases have a higher incidence in industrialized countries and their pathogenesis involves an over-reaction of the immune system. A genetic factor is believed to predispose to the development of chronic inflammation in response to an unidentified stimulus. Exposure to infections in childhood may modulate future immune responses. Parasitosis, particularly Schistosomiasis, stimulate Th2 immune responses. It has been hypothesized that the absence of these parasitic infections, as seen in economically developed countries, favors a Th1 response that may result in the clinical appearance of Crohn's disease later in life.**

Objective. **To determine the prevalence of *Schistosoma mansoni* antibodies in Puerto Ricans with Inflammatory Bowel Disease and controls.**

Methods. **Serum from 92 Puerto Ricans with IBD and 106 controls was screened for *S. mansoni* adult microsomal antigens (MAMA) using the FAST™:ELISA assay. Those positive were confirmed with an enzyme-linked immunoelectrotransfer blot test.**

Results. **Seven serum samples (3 UC and 4 controls) were positive for *S. mansoni* antibodies. There was no significant difference between groups in gender, municipality of origin or seroprevalence of Schistosomiasis. The control group was slightly older than the IBD group.**

Conclusions. **Our study did not demonstrate an inverse relation between Schistosomiasis and IBD. However, the decreasing prevalence of Schistosomiasis in the general population of Puerto Rico may account for this result.**

Key words: Schistosomiasis, Crohn's disease, Ulcerative colitis, Inflammatory Bowel Disease (IBD)

The inflammatory bowel diseases (IBD), Crohn's and ulcerative colitis, are chronic diseases of unknown etiology affecting the gastrointestinal tract. Prevalence of these diseases is higher in industrialized countries and the incidence has been observed to increase with the economic development of a population (1-3). Although the cause of IBD is not known, the pathogenesis of the diseases involves an over-reactive gastrointestinal immune system. The predisposition to the conditions is thought to be genetically determined, as suggested by the higher familial incidence. The triggering factor for the inflammatory reaction has not been identified, but bacterial products in the intestinal lumen are strong candidates. Other infectious agents, as well as food products or other environmental factors could also act as stimuli for the immunologic system and it is conceivable that a different stimulus accounts for the development of IBD in individual cases. In Crohn's disease, an increased production of cytokines has been described, in particular Tumor Necrosis Factor (TNF), supporting a cellular immune response of the Th1 class (4-8).

The economic development of underdeveloped countries results in improved sanitation and living conditions and a reduction in the incidence of gastrointestinal infections and parasitosis. Exposure to infections during childhood has been suggested to have a role in the modulation of future immune responses (9-13). Elliott proposed that childhood exposure to parasites

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that stimulate Th2 immune responses, such as helminths, results in conditioning of the gastrointestinal immune system to favor these responses. The absence of these infections would favor a Th1 response. In genetically predisposed individuals, exposure to an unidentified stimulus in the gastrointestinal tract might result in the clinical appearance of Crohn's disease (14).

The prevalence of IBD in Puerto Rico has risen, as suggested by the increase in admissions to the University and San Juan City Hospitals from 1990 to 1995. Hospitalizations for Crohn's disease increased from 5 per 10,000 in 1990 to 10 per 10,000 in 1995. Using the data base for 1966 of the largest health insurer in Puerto Rico, Torres et al described a prevalence of 4.13 per 10000 for Crohn's disease and 6.22 per 10000 for ulcerative colitis, placing Puerto Rico within the middle range for these diseases (15). Infection with *Schistosoma mansoni,* on the other hand, has decreased significantly in Puerto Rico, with a seroprevalence of 10.6% in 1995 as compared to 20% in 1927 (16).Prevalence has been found to favor some well defined geographical areas.

Schistosomiasis infection elicits predominantly a Th2 response (17). If Elliott's hypothesis is correct, the disappearance of this infection in Puerto Rico would remove one of the factors that precondition the immune system to Th2 responses and enhance the possibility of Th1 responses and the manifestation of Crohn's disease in genetically predisposed persons.

The aim of our study was to determine the prevalence of *Schistosoma mansoni* antibodies in Puerto Ricans with IBD and compare it with a control group.

Methods

Stored frozen serum samples from 92 Puerto Ricans with IBD and 106 controls were tested. The samples of IBD were obtained from patients followed in the Inflammatory Bowel Disease clinic of the University Hospital. All the IBD patients had a confirmed diagnosis by clinical, endoscopic, radiologic and/or histologic classic criteria.The controls were healthy volunteers recruited among hospital and university personnel. Demographic characteristics of the population included age, gender, diagnosis, and municipality of residence.

The serum was screened for antibodies to *S. mansoni* adult microsomal antigens (MAMA) using the Falcon assay screening test:enzyme linked immunoabsorbent assay (FAST™:ELISA, Becton Dickinson, Oxnard, CA). Samples yielding a reactivity > 10 units were considered positive. All FAST™:ELISA positive samples were subjected to confirmation by an enzyme-linked immunoelectrotransfer blot test (EITB), in which the presence of species-specific GP30 Kda antigen band is considered diagnostic (18).

S. mansoni prevalence by study group, gender, and municipality was compared using Fisher's exact test. Differences in age were determined by analysis of variance. Data entry and analysis was performed using Epi-Info (v. 6.04). The protocol was approved by the Institutional Review Board of the Medical Sciences Campus.

Results

Ninety two samples with IBD (39 with Crohn's disease and 53 with ulcerative colitis) and 106 controls were tested. Seven samples (3.5%) were positive for *S. mansoni* antibodies, as confirmed by Western Blot, three (5.7%) among the ulcerative colitis group and four (3.8%) controls (Table 1). This difference was not significant (p=1.00).

Table 1. Samples Positive for *S. mansoni* Antibodies

	N	Positive	%
Ulcerative colitis	53	3	5.7%
Crohn's disease	39	0	0%
Controls	106	7	3.8%

The ulcerative colitis group had 36 females (67.9%) and 17 males, with a mean age of 39.9 $\pm$ 16.35 (range 17-87). Prevalence of *S. mansoni* antibodies in this group was 5.67%. There were 23 females (58.9%) and 16 males in the Crohn's disease group, with a mean age of 38.39 $\pm$ 13.34 (range 21-75). None of these subjects had a positive test for *S. mansoni* antibodies. The control group had 58 females (56.9%), 44 males, and 4 subjects had no identifying data. Mean age of controls was 45.8 $\pm$ 16.68 (range 19-84). The prevalence of *S. mansoni* antibodies in the control group was 3.8%. Controls were significantly older than IBD patients (p=.013). No significant differences among the three groups in regard to gender, municipality of residence, or prevalence of *S. mansoni* antibodies were found (p>0.05).

Discussion

Although no *S. mansoni* antibodies were detected in any of the subjects with Crohn's disease, the difference between this group and the controls or the ulcerative colitis subjects was not significant. Because of the low prevalence of *S. mansoni* antibodies found in our control population, an indication of the infrequency of this infection in Puerto Rico, a larger sample would be required to show a statistically significant difference between subjects with

Crohn's disease and controls. Although subjects with ulcerative colitis had a similar prevalence to the control group, the size of the sample is again too small to draw any conclusions.

Schistosomiasis has been associated with specific geographic areas where the water sources are infected, and population studies have shown that the prevalence of antibodies in these areas can be as high as 38.46 % (16). The municipality of residence was similar in our three groups. It would be interesting to repeat the study utilizing controls and subjects with Crohn's disease and ulcerative colitis living in high prevalence areas. However, because IBD is not a common condition in Puerto Rico, recruiting enough subjects to allow for statistical significance may not be plausible.

We chose Schistosomiasis infection for our study not only because it elicits a Th2 response, but also because it is an infection whose previously high prevalence has been shown to decrease along with the economic development of Puerto Rico. There is no published data on the prevalence of other helminth infections in Puerto Rico, but parasitosis is no longer a serious public health problem in our population.

Although our study did not have the statistical power to suggest an inverse association between parasitosis and Crohn's disease as suggested by Elliott, it supports previous work showing that Schistosomiasis is decreasing in the general population of Puerto Rico.

Resumen

La etiología de las Enfermedades Inflamatorias del Intestino (EII), enfermedad de Crohn y colitis ulcerosa, es desconocida. La incidencia de estas enfermedades es más alta en países industrializados. Su patogénesis envuelve una sobre-reacción del sistema inmune. Se cree que un factor genético predispone al desarrollo de la inflamación crónica en respuesta a un estímulo aún no identificado. La exposición a infecciones en la niñez puede modular respuestas inmunes futuras. La parasitosis, en particular la esquistosomiasis, estimula respuestas inmunes Th2. Se ha propuesto la hipótesis de que la ausencia de estas infecciones parasitarias, como sucede en países desarrollados económicamente, favorece una respuesta inmune Th1 que puede resultar en el desarrollo clínico de enfermedad de Crohn más tarde en la vida. El propósito de este estudio fue determinar la prevalencia de anticuerpos a *Schistosoma mansoni* en puertorriqueños con Enfermedad Inflamatoria de Intestino y controles. Se utilizaron sueros de 92 puertorriqueños con EII y 106 controles fue evaluado para anticuerpos adultos microsomales de *S. mansoni* (MAMA) utilizando el ensayo FASTJ:ELISA. Las muestras positivas fueron confirmadas con una prueba de inmunoelectrotransferencia ligada a enzimas. Siete muestras (3 con colitis ulcerosa y 4 controles) fueron positivas para anticuerpos de *S. mansoni*. No hubo diferencias significativas entre los grupos en relación a género, municipio de procedencia, o seroprevalencia de esquistosomiasis. El grupo control era ligeramente mayor en edad que el grupo de EII. Este estudio no demostró una relación inversa entre equistosomiasis y EII. Sin embargo, la prevalencia decreciente de equistosomiasis en la población general de Puerto Rico podría explicar estos resultados.

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