ARTÍCULOS ORIGINALES

Epidemiological analysis of eosinophilia and elevation of immunoglobulin E as a predictable and relative risk of enteroparasitosis

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ABSTRACT

Introduction: among the intestinal parasites, the helminthiasis occupies a prominent position in Brazil, since it worsens malnutrition and the gives rise to neurovegetative disorders. Helminths like *Ascaris lumbricoides, Enterobius vermicularis,* hookworm, *Trichuris trichiura* and *Strongyloides stercoralis* stand out due to several factors that modulate the immune response of individuals. Among the protozoa are *Giardia lamblia, Entamoeba histolytica/E. dispar*. Parasitic helminth antigens are important to stimulate the production of cytokines such as interleukin-4 and interleukin-5, which act through the induction of IgE synthesis and activation of eosinophils. Eosinophilia is usually detectable in pre-patent period of parasitism, initially linked to B lymphocytes, under the command of Th-2 lymphokines (IL-4 and IL-5), producing IgE in response to initial exposure to an antigen or allergen. Serum IgE high levels occur in tissue migration of larvae or harboring of parasites in tissues. **Objective**: to determine the presence of eosinophils and IgE elevation in children with intestinal parasites. **Methods**: high levels of IgE and eosinophils were observed in groups infected and not infected (allergic) to calculate the relative risk of intestinal parasites presumptive differentiated between protozoa and helminths and check what values of these indicators are observed in the epidemiological profile of the surveyed population. **Results**: the values obtained by calculating the relative risk for eosinophilia, compared with helminths and allergies was 11.71, but when examined by giardiasis compared with other diseases, the relative risk was 0.75. Since the comparison of helminths and giradiase, the relative risk was 27.33. Since IgE and its parameters were appropriate commit Helminth relative risk 0.39; Giardiasis had relative risk 8, 18 and Helminths compared with giardiasis had 0.03. **Conclusion**: in this study it was possible to observe that helminthiasis is connected with cases of eosinophilia with alteration of IgE, which in turn contributes to the presence of IgE eosionofilia and has an effector response against helminths that provides the expulsion of its larvae.

Key words: epidemiology, eosinophilia, intestinal parasites.

INTRODUCTION

Enteroparasitoses are a serious problem in public health, especially in the third world countries,[1] and they provoke frequent malnutrition and diarrhea, as well as physical and mental problems in children.[2] Among the parasitic pathogens more frequently found in human beings, there are the helminths *Ascaris lumbricoides, Enterobius vermiculares, Ancylostoma duodenale, Strongiloides stercoralis* and *Trichuris trichura*,[3] as well as the intestinal protozoa, such as *Giardia lamblia*.[3] Transmitting these parasites to another host depend on complex epidemiological cycles, on a great variety of beings and on the lack of basic sanitation, making it difficult to control such diseases. These factors demand social actions, as well as public health policies.[4] The epidemiological, biological and immunological study of parasite diseases is necessary, in order to contribute more application in early diagnosis and prevention of these diseases. The immunological response caused by a parasite infection is complex and multiple, due to the great metabolic diversity of these beings. Thus,

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there are consequences of Th1 or Th2 responses, depending on the kind of parasite on the host. In accordance with *Machado et al*[5] giardiasis is a kind of parasite infection that causes intense eosinophilia, as well as the helminths mentioned above.[6] However, *Melo-Reis et al*[7] do not agree with this idea and state that the relation between eosinophilia (common in T2 responses) and protozoa is little described on the scientific literature; also, they say it is important to emphasize that not all intestinal parasites —especially those that are on the intestinal light— are able to induce to eosinophilia, what would not make this biological parameter a good indicator of protozoa parasitosis. On the other hand, in helminthiasis, the parasite antigens stimulate an intense Th2 response, with production of inter-leukin-4 and interleukin-5, what induce to the synthesis of Immunoglobulin E (IgE) and eosinophils activation.[8] Eosinophilia is generally detectable in the prepatent period of parasitism, that is, its detection occurs before helminths turn into adults and the occurrence of parasite biological forms that can be detectable.[9] The function of eosinophils is to destroy some helminths through cellular cytotoxicity that depends on the antibody.[10] One of the mechanisms within this process is the reaction started by IgE. Initially, B lymphocytes, directed by Th-2 lymphokine (IL-4 e IL-5), produce IgE in response to the initial exposure to an antigen or allergen.[11] In parasite infections, high IgE serum levels appear on the tissue migration of larvae or parasite housing in tissues.[12]

Eosinophilia and elevation of IgE, however, are not initial indicators of parasitosis, because it occurs in other pathological conditions(ex:allergies). There is no data on literature about serum values of the indicators mentioned, regarding the epidemiological profile of people in our society who suffer from parasitosis, but the occurrence and indication of eosinophilia and IgE high levels in people with such pathologies is wide. Thus, we propose a quantitative analysis on a sample of individuals believed to have enteroparasitosis, of the relative risk (RR) in this condition, as well as of the presence of eosinophilia and elevation of IgE. The aim of this study was to compare eosinophils mean values and elevation of IgE in children with and without parasitosis, obtaining reference values of these biological indicators as a clue of the latency period of enteroparasitosis. Also, intend evaluating the quantitative differences between these immunological indicators regarding helminthiasis, protozoa diseases and other pathologies, analyzing the RR values to the parasitic conditions and eosinophilia, as well as elevation of IgE.

METHOD

This study approaches qualitatively and quantitatively, carried out between June 2009 and June 2010. The sample in this research constituted by 83 exams believed to have enteroparasitosis, performed at the Alpha Clin lab, in São Paulo, Brazil. The variables observed were concentration of eosinophils and of IgE, as well as diagnosis suspect of enteroparasitosis. The data were evaluated through application of descriptive statistics of central tendency, using standard deviation as a measure of dispersion. Besides the description of the values of these indicators, it was performed a test of statistical hypothesis, in which H0 was taken as a null hypothesis (there is no difference in the values of eosinophils and IgE in cases of parasitosis and other pathologies), and H1 was taken as an alternative hypothesis (there are differences between the values observed on the concentration of eosinophils and IgE and the condition of being a carrier of intestinal helminthiasis). The values of eosinophilia and IgE were individually submitted to a test Kruskall-Wallis between the groups with and without parasitic infections. All the statistical tests were carried out considering a significance level of $p < 0.05$, and a confidence level of 95 % was esta-blished as appropriate. In the analyses, we used EPI-INFO 2000 (Centers for Disease Control and Prevention, a software of data tabulation in epidemiology. Available in: http://www.cdc.gov/epiinfo.). The positive exams were diagnosed by sedimentation coproscopy techniques of *Hofman*, *Pons* and *Janner*,[13] and by the *Faust* method.[14] Eosinophils concentration was analyzed by an automated electrical impedance technique, spectrophotometry and optical dispersion (STKS® device), and its reference value in percentage ranged from 1.0 to

4.0.[15] The analysis of IgE concentration was made by using an automated technique based on quimioiluminecense (Acess device®), with reference values from 1.31 to 165.3 UI/mL.[16] Calculating the relative risk between eosinophilia diagnosis and elevation of IgE with enteroparasitosis (exposed), and eosinophilia and elevation of IgE without parasitosis (non-exposed) is going to be made in a 2x2 table, following the models proposed by *Pereira*, as in the table 1 and 2. The relative risk is going to be calculated according to the formula (A/A+B)/(C/C+D), where A are the sick and B are the healthy within the exposed groups; and C (sick) and D (healthy), within the non-exposed group.[17]

Table 1. Incidence of eosinophilia in helminthiasis, giardiasis and allergies

| Analyzed Group | Total | Eosinophilia | |
		Present No. (%)	Absent No. (%)
Helminthiasis	30	29 (96.66)	1 (3.33)
Giardiasis	32	6 (18.75)	26 (81.25)
Allergies	19	6 (31.58)	13 (68.42)

The indexes of eosinophilia was based on *Zago et al*, 2004.[15]

Table 2. Levels of IgE observed in helmintic infections, giardiasis and other allergies

| Analyzed Groups | Total | IgE levels | |
		Elevated No. (%)	Normal No. (%)
Helminthiasis	30	29 (96.66)	1 (3.33)
Giardiasis	32	2 (6.25)	30 (93.75)
Allergies	19	16 (84.21)	3 (15.79)

The indexes of normality and elevation were based on *Soares*, 2002.[16]

The variances in median of concentrations are on table 3.

Information that could identify the patients or the laboratory where the exams were carried out was hidden. This work was approved by the Ethics Committee in Research of the institution where it was carried out, for it was in accordance with the policies stated by Resolution 196/96 of Conselho Nacional de Pesquisa (CNPq/Brasil). Also, for obtaining such results, other causes of eosinophilia and IgE were excluded. Because of random action, there is a variety of disorders associated with eosinophilia,[10] corroborating Melo-Reis *et al*, who suggest there was no association between giardiasis and eosinophilia.[7]

RESULTS

The values obtained calculating RR to eosinophilia, comparing helminthiasis to other pathologies, was 11.71. When comparing giardiasis to allergies, RR was 0.75. Between helminthiasis and giardiasis, RR was 27.33.

Besides, parameters of normality or IgE elevation and their respective RR to the configurations helminths and other pathologies were 0.39. Concerning giardiasis and allergies, RR was 8.18; and when helminthiasis and giardiasis were compared, RR was 0.03.

DISCUSSION

Helminthiasis are well defined when we describe cases of eosinophilia and IgE alteration. Eosinophils and IgE are part of the effector response against helminths, what contributes to release the larvae.[18, 19] Comparing the medians of

Table 3. Comparison of eosinophils values and IgE levels among different studied groups

| | Comparison Groups | | | |
	Helminthiasis	Giardiasis	Allergies	
Eosinophils				
Median	8.00 %	2.00 %	3.00 %	p value Kruskall-Wallis
Range	2.21	1.83	2.06	0.008984
IgE levels				
Median	174 UI/mL	34 UI/mL	42 UI/mL	p value Kruskall-Wallis
Range	23.9	5.66	55	0.013241

parasitosis carriers and non-carriers, Kruskall-Wallis test showed, at the established levels, significantly greater relations between the condition of a carrier and the elevated levels of eosinophils and IgE, with the same parameters, in non-carriers of parasitosis.

A few works demonstrated that *Giardia lamblia* is able to induce an immunological response with elevation of eosinophils.[7,10,11] From the data presented in this work, we observed that patients with giardiasis may present normal values of eosinophils, which confirmed *Melo-Reis*,[7] *Fontenele et al*[10] and *Rosário et al*,[11] but contradicted *Machado et al.*[6]

Analyzing RR, we noticed relevance in eosinophilia as a remarkable factor of association (between the events "pathology" and "elevation of indicators"): in helminthiasis and giardiasis, of 27.33 times; in helminthiasis and other pathologies, RR is 11.71 times; and in giardiasis and in other pathologies, RR is only 0.75, which is higher than 1.0 and, thus, constitute a risk factor, in accordance with *Pereira*.[17] The elevation of IgE did not show relevance between helminthiasis and giardiasis (0.03), between helminths and other pathologies (0.39), but presented risk between giardiasis and other pathologies (8.18). In its turn, IgE proved to be a better indicator to giardiasis than to helminthiasis, partly confirming *Machado et al.*[6]

Regarding the hypotheses, we deny H0 and noticed a more remarkable difference in the RR of these indicators between parasitosis than in other diseases; however, we accept H1, in which there are statistical differences of RR between helminthiasis carriers and the elevation in the mentioned indicators.

Patients with parasitosis vary in their clinical condition, as asymptomatic and symptomatic, altering the polymorphic form of these diseases. They present diarrhea, weight loss, among other manifestation, or they do not present any important symptomatology for a long time.[18] Thus, eosinophilia is a highly important hematological parameter, and it should be considered when approaching and assessing a patient who presents it, for it suggests intestinal parasitosis, specifically pre-patent helminthiasis. Also, the association of this indicator with the condition of the carrier should be observed.

In this study, it was possible to observe that helminthiases are associated with cases of eosinophilia. The values observed can indicate helminth infections (RR) and they are very different from other pathological conditions. This study is highly important for public health, for enteroparasitoses are among its main problems. Their diagnosis depends on a laboratorial structure and experienced professionals with biological information regarding the immunological system. However, we accept that the number of individuals in this study is not sufficient to a good analysis, although it is a good tool for new researches on Immunology, parasite biology and public health.

Análisis epidemiológico de eosinofilia y la elevación de inmunoglobulina E como riesgo relativo y predecible de enteroparasitosis

RESUMEN

Introducción: entre los parásitos intestinales, la helmintiasis ocupa un lugar destacado en Brasil, porque causa malnutrición y la instalación de cuadros de trastornos neurovegetativos. Helmintos como *Ascaris lumbricoides*, *Enterobius vermicularis*, anquilostoma, *Trichuris trichiura* y *Strongyloides stercoralis* se destacan debido a varios factores que modulan la respuesta inmune de los individuos. Entre los protozoos, los más destacados son *Giardia lamblia* y *Entamoeba histolytica/E. dispar*. Los antígenos de helmintos parásitos son importantes para estimular la producción de citocinas como la interleucina-4 e interleucina-5, que actúan a través de la inducción de la síntesis de IgE y la activación de los eosinófilos. La eosinofilia es detectable por lo general en el período pre-patente de parasitismo, inicialmente vinculada a los linfocitos B, bajo el mando de Th-2 linfocinas (IL-4 e IL-5), la producción de IgE en respuesta a la exposición inicial a un antígeno o alergeno. Los niveles séricos de IgE se producen en la migración de las larvas de los tejidos o la acogida de los parásitos en estos. **Objetivo**: determinar la presencia de eosinófilos y la elevación de IgE en niños con parásitos intestinales y cuantificar el riesgo relativo de estos parámetros biológicos. **Methods**: se observaron altos niveles de IgE y de eosinófilos en grupos infectados y no infectados (alergias) a fin de calcular el riesgo relativo (RR) de parásitos intestinales presuntamente diferenciados entre protozoos y helmintos, así como chequear qué valores de estos indicadores se observan en el perfil epidemiológico de la población estudiada. **Resultados**: los valores obtenidos mediante el cálculo del riesgo relativo para la

eosinofilia, en comparación con helmintos y otras enfermedades fue 11,71, pero cuando se examina por la giardiasis en comparación con alergias resultó de 0,75. En la comparación de los helmintos y la giardiasis, el riesgo relativo fue de 27,33. Dado que la IgE y sus parámetros eran adecuados, entonces helmintos RR 0,39; giardiasis RR 8,18; y helmintos en comparación con giardiasis 0,03. **Conclusión**: con este estudio se pudo observar que las helmintiasis están relacionadas con casos de eosinofilia con alteración de la IgE; este último, que a su vez contribuye a la presencia de IgE eosionofilia y a tener una respuesta efectora frente a helmintos que causan la expulsión de sus larvas.

Palabras clave: epidemiología, eosinofilia, parásitos intestinales.

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