Frequency of *Strongyloides stercoralis* Infection in Alcoholics

Luiz Carlos Marques de Oliveira/+, Camila Toffoli Ribeiro, Daniel de Melo Mendes, Tatiana Cunha Oliveira, Julia Maria Costa-Cruz*

Ambulatório de Alcoolismo, Departamento de Clínica Médica, Faculdade de Medicina *Laboratório de Parasitologia, Departamento de Imunologia, Microbiologia e Parasitologia, Instituto de Ciências Biomédicas, Universidade Federal de Uberlândia, Avenida Pará 1720, Campus Umuarama, 38400-902 Uberlândia, MG, Brasil

Several studies have shown that chronic alcoholics have increased susceptibility to infections due to higher exposure to infectious agents as well as breakdown in their immune defenses. As Strongyloides stercoralis *infection is usually more relevant in immunocompromised patients, the aim of this study was to evaluate the frequency of* S. stercoralis *infection in alcoholics. Thus, coproparasitological examination was carried out in 145 subjects, from which 45 were chronic alcoholics (mean age of 45.7 ± 11.0 years), 10 were nonalcoholic cirrhotic patients (mean age of 50.2 ± 13.1 years), and 90 were asymptomatic nonalcoholic subjects (mean age of 46.7 ± 10.1 years), which served as controls. From the alcoholics, 9 had hepatic cirrhosis, 9 had chronic pancreatitis and 27 had neither cirrhosis nor pancreatitis. For the diagnosis of strongyloidiasis, the Baermann-Moraes and Lutz methods were used in three fecal samples from each subject. Samples were collected at alternated days, and three slides of each sample were analyzed for each method, thus totalizing 2,610 slides examined. The frequency of strongyloidiasis in the total alcoholic group (33.3%) and in the subgroups of alcoholics, i.e., patients with hepatic cirrhosis (44.4%), with chronic pancreatitis (33.3%), and those with no cirrhosis or pancreatitis (29.6%) was statistically higher than that found in the control group (5.5%). None of the individuals with nonalcoholic hepatic cirrhosis had* S. stercoralis *infection. Our results showed that the chronic alcoholism itself is an important factor that predisposes to strongyloidiasis.*

Key words: *Strongyloides stercoralis* - alcoholism - hepatic cirrhosis - chronic pancreatitis

Strongyloides stercoralis is widely spread throughout the world, especially in tropical and subtropical regions. This worm has a complex parasitic life cycle and many patients chronically infected with *S. stercoralis* are asymptomatic, and thus, *S. stercoralis* infection can persist for decades. However, in some situations, hyperinfection or disseminated infection has been found usually associated with host immunosupression (Grove 1996).

Infections are more frequent and severe among alcoholics probably due to dulled mental function, breakdown of local protective barriers, aspiration, exposure to pathogens and malnutrition (MacGregor 1986), in addition to alterations of host immune defense mechanisms caused by alcoholism (MacGregor 1986, Jerrells 1991, Cook 1998). Despite the fact that alcoholism is a condition observed in patients with *S. stercoralis* hyperinfection (Grove 1996), no controlled study was found in the available literature showing that strongyloidiasis is more frequent in alcoholic than nonalcoholic patients.

As we have often found patients infected with *S. stercoralis* in our ambulatory for alcoholism treatment, the present study was proposed to investigate the frequency of this infection in these alcoholic patients.

PATIENTS AND METHODS

A total of 145 subjects were evaluated in this study, from which 45 were alcoholics (mean age of 45.7 ± 11.0 years), 10 had nonalcoholic hepatic cirrhosis (mean age of 50.2 ± 13.1 years), and 90 were asymptomatic nonalcoholic subjects (mean age of 46.7 ± 10.1 years), which served as controls. From the alcoholics, 9 had hepatic cirrhosis, 9 had chronic pancreatitis, and 27 had no debilitating disease. All subjects had similar background and current socio-economic conditions. None of them had used corticosteroids or any other immunosupressor drug. Informed consent was obtained from all individuals.

Three fecal samples from each subject were collected at alternated days in plastic recipients without conservatives and stored at 4°C. Parasitological diagnosis was carried out within the first 24 h after the feces have been collected, by the Baermann-Moraes (Baermann 1917, Moraes 1948) method, using approximately 10 g of feces of each samples. Furthermore, 10% formalin solution was added to remaining samples for further analysis by the Lutz (1919) method. Three slides were prepared for the Baermann-Moraes analysis and three for the Lutz analysis for each of the 435 samples. Thus, a total of 2,610 slides were examined. Fisher's exact test was used for statistical analyses.

RESULTS

The frequency of *S. stercoralis* infection in the alcoholic group (33.3%) was higher ($p < 0.001$) than that found in the control group (5.5%). When analyzing the subgroups of alcoholic patients, the frequency of this infection in patients with hepatic cirrhosis (44.4%), chronic pancreatitis (33.3%), and those with no such diseases (29.6%) also were statistically higher as compared to the control group, but with no significant difference among the subgroups. In addition, the frequency of strongyloidiasis in patients with alcoholic hepatic cirrhosis (44.4%) was higher ($p < 0.05$) than that of the patients with nonalcoholic hepatic cirrhosis (0%). This latter group was not included in the control group (Table). No protozoan infection or any other helminthiasis was found in the group of

+Corresponding author. Fax: +55-34-3218.2349. E-mail: oliveiralcm@ufu.br
Received 25 April 2001
Accepted 16 August 2001

alcoholic patients, and out of the four alcoholic patients who had diarrhea none had a positive stool examination for *S. stercoralis*. In the control group, other than five cases of *S. stercoralis* infection, one (1.1%) case of *Giardia lamblia* and one (1.1%) case of hookworm infection were found.

TABLE

Frequency of *Strongyloides stercoralis* infection in alcoholics with and without hepatic cirrhosis (HC) or chronic pancreatitis (CP), in non-alcoholic HC and controls. Statistical analysis by Fisher's exact test

Group	Positive	Negative	Total
Alcoholics	15 (33.3%)	30	45
with HC	4 (44.4%)	5	9
with CP	3 (33.3%)	6	9
without HC or CP	8 (29.6%)	19	27
Non-alcoholic HC	0	10	10
Controls	5 (5.5%)	85	90

$p < 0.001$: alcoholics > controls; $p < 0.01$: alcoholics without HC or CP > controls; alcoholic HC > controls; $p < 0.05$: alcoholic CP > controls; alcoholic HC > non-alcoholic HC

DISCUSSION

Our results showed that the frequency of *S. stercoralis* infection among alcoholics was higher ($p < 0.001$) than that in the control group (33.3% versus 5.5%); such frequency, in the alcoholic group, was higher than that found in a study carried out in Costa Rica (5.7%), in which a single sample of feces was examined (Avendaño et al. 1999), whereas in our study, three samples from each patient were examined. The frequency of strongyloidiasis in alcoholic cirrhotic patients (44.4%) in this study was similar to that found in cirrhotics patients from another region of the State of Minas Gerais (40.2%), which was predominant in alcoholic cirrhosis (Gaburri et al. 1997).

When an individual is infected with *S. stercoralis* there are three possible outcomes that are probably dependent on host immune system: eradication of infection, chronic infection and hyperinfection or disseminated infection (Grove 1996). Thus, at least two hypotheses could be proposed to explain the high prevalence of *S. stercoralis* infection in alcoholics: (1) higher predisposition to infection and/or (2) breakdown of immune defenses to eliminate the parasites, and consequently, greater amount of parasite shed in the feces, thus facilitating the diagnosis.

The first hypothesis can occur, at least in some alcoholic patients, due to poor hygiene conditions, either as a result of higher heteroinfection by walking barefoot in contaminated sites or by autoinfection, in this latter case, rhabditiform larvae present in the perianal region from infected individuals transform in infective filariform larvae and then penetrate (Grove 1996). This infection way also occurs in patients that evacuate in their clothes or due to deficient hygiene following the evacuation (Costa-Cruz 2000).

Concerning the second hypothesis, immunodeficiency would be related to impairment in the ability of the host immune system to eliminate the parasite, which could occur at level of the intestinal mucosa, the cell-mediated and/or humoral immunity, or the complement system.

The defense system at level of intestinal mucosa is related to mast cells, which can act either directly on the parasites or indirectly through their ability to attract and modulate eosinophils; the activation of mast cells can represent an important effector mechanism to contain the initial infection and to protect the host from a disseminated infection (Barrett et al. 1988, Nawa et al. 1994). The findings of reduced number of macrophages in the duodenal mucosa in alcoholics suggest that in these individuals there is a weakening of this part of the nonspecific immune system (Maier et al. 1999). In patients with alcoholic cirrhosis a reduced IgA secretion into the intestinal lumen was also observed, which could be partially responsible for the high incidence of intestinal infection observed in severe cirrhosis (Pelletier et al. 1982).

In the cell-mediated immune response to helminths, there is a predominant Th2 profile from T helper cells with secretion of cytokines such as IL-3, IL-4, IL-5, IL-6, IL-10 and IL-13; IL-3 and IL-4 stimulate basophils and mast cells, IL-4 and IL-13 stimulate B cells to produce IgE antibodies, IL-5 promotes attraction and activation of eosinophils and induction of IgA, and IL-6 promotes stimulation of granulocytes, T and B cells (Finkelman et al. 1999, Roitt et al. 1999, Costa-Cruz 2000). Clinical evidence on the impairment of the cell-mediated immunity includes a higher frequency of tuberculosis and head and neck cancers in alcoholics than in non-alcoholics (MacGregor 1986). The chronic use of ethanol can result in a loss of lymphoid cells from the peripheral blood, spleen, and thymus associated with a loss of lymphocyte function, especially T-cell-dependent immune responses, resulting in an increased incidence of infections, which would be primarily opportunistic infections (Jerrells 1991). In alcoholics the number of circulating T lymphocytes is reduced as is the ability of their lymphocytes to undergo blast transformation in response to mitogenic stimulation (Glassman et al. 1985, MacGregor 1986). Alcohol has been shown to affect production of some cytokines, and changes in their balance have profound effects on the function of the immune cells; experimentally, it has been reported that alcohol exposure shifted the Th1/Th2 balance toward Th2 excess (Cook 1998). The acute and chronic effects of alcohol also prevent the normal delivery of polymorphonuclear cells to sites of bacterial invasion, thus contributing significantly to increase the frequency and the severity of bacterial infections in alcoholics, regardless of concomitant alcoholic liver disease (MacGregor 1986). Several effects on immunity of alcoholics can disappear with the abstinence (Lundy et al. 1975, Maier et al. 1999).

The most of individuals infected by *S. stercoralis* produces specific IgG, IgM, IgA and IgE antibodies, with characteristic increasing of IgE and IgG4 responses in this helminthiasis; IgA, IgE and IgG4 antibodies seem play an important role in the control of the levels of infection by *S. stercoralis* (Costa-Cruz 2000). The effector IgA-mediated immune mechanism modulates the larval output by decreasing the worm fecundity and egg viability, whereas IgE regulates the autoinfection and IgG4 blocks the IgE-mediated responses, thus having a central role for the establishment and persistence of asymptomatic chronic strongyloidiasis (Atkins et al. 1999). Chronic alcoholics often have greatly increased serum immunoglobulin levels. Typically, IgA is elevated both in alcoholics with and without alcoholic liver disease, IgG is elevated in alcoholic liver disease, and IgM is only elevated in alcoholic liver disease

with active disease, such as alcoholic hepatitis (Cook 1998). The changes in the humoral arm of the immune system of patients who chronically ingested alcohol are detectable before overt clinical or biochemical signs of liver damage (Drew et al. 1984). Although the increase of immunoglobulins antibody is usually associated with the development of specific immunity, alcoholic patients with greatly elevated immunoglobulin levels are often immunodeficient (Cook 1998).

Deposition of complement components C1q, C3, C4, C8 and properdin can be found on the larval surface (Messias et al. 1994). *S. stercoralis* filariform larvae and its antigenic preparations activate the complement system by both classical and alternative pathways, and promotes the adhesion of peripheral blood mononuclear and polymorphonuclear cells on the larval surface and these cells can contain enzymes that are lethal to *S. stercoralis*. Thus, the complement system in association with the effector cells plays an important role in the nonspecific immune defense of the host against *S. stercoralis* infection, suggesting the complement system as a first line of host defense (Messias et al. 1994). Regarding the action of the alcohol on the complement system, the data are too conflicting to allow generalization, although serum bactericidal activity may be impaired transiently by acute intoxication, and patients with cirrhosis of the liver tend to have reduced serum complement activity (MacGregor 1986).

Our results showed that the chronic alcoholism itself is an important factor that predisposes to strongyloidiasis, which can be resulting of higher predisposition to infection or due to countless immunodeficiencies described in alcoholics.

ACKNOWLEDGEMENTS

To Maria das Graças Marçal for technical assistance.

REFERENCES

Atkins NS, Conway DJ, Lindo JF, Bailey JW, Bundy DAP 1999. L3 antigen-specific antibody isotype responses in human strongyloidiasis: correlations with larval output. *Parasite Immunol 21*: 517-526.

Avendaño L, Hernandez F, Jimenez F, Avila A, Castro D 1999. *Strongyloides stercoralis* en pacientes alcohólicos. *Parasitol al Día 23*: 91-94.

Baermann G 1917. *Eine Einfache Methode zur Auffindung von Ankylostomum (Nematoden) Larven in Erdproben.* Mededeel mit h. Geneesk. Lab Weltvreden Feestbundel, Batavia, p. 41-47.

Barrett KE, Neva FA, Gam AA, Cicmanec J, London WT, Phillips JM, Metcalfe DD 1988. The immune response to nematode parasites: modulation of mast cell numbers and function during *Strongyloides stercoralis* infections in nonhuman primates. *Am J Trop Med Hyg 38*: 574-581.

Cook RT 1998. Alcohol abuse, alcoholism, and damage to the immune system – a review. *Alcohol Clin Exp Res 22*: 1927-1942.

Costa-Cruz JM 2000. *Strongyloides stercoralis.* In DP Neves, AL Melo, O Genaro, PM Linardi (eds), *Parasitologia Humana*, 10 ed., Atheneu, Rio de Janeiro, p. 247-256.

Drew PA, Clifton PM, LaBrooy JT, Shearman DJC 1984. Polyclonal B cell activation in alcoholic patients with no evidence of liver dysfunction. *Clin Exp Immunol 57*: 479-486.

Finkelman FD, Wynn TA, Donaldson DD, Urban Jr JF 1999. The role of IL-13 in helminth-induced inflammation and protective immunity against nematode infections. *Curr Opin Immunol 11*: 420-426.

Gaburri D, Gaburri AK, Hubner E, Lopes MHM, Ribeiro AMB, Paulo GA, Pace FH, Gaburri PD, Ornellas AT, Ferreira JOD, Chebli JMF, Ferreira LEVVC, Souza AFM 1997. Parasitoses intestinais e cirrose hepática. *Arq Gastroenterol 34*: 7-12.

Glassman AB, Bennett CE, Randall CL 1985. Effects of ethyl alcohol on human peripheral lymphocytes. *Arch Pathol Lab Med 109*: 540-542.

Grove DI 1996. Human strongyloidiasis. *Adv Parasitol 38*: 251-309.

Jerrells TR 1991. Immunodeficiency associated with ethanol abuse. *Adv Exp Med Biol 288*: 229-236.

Lundy J, Raaf JH, Deakins S, Wanebo HJ, Jacobs DA, Lee TD, Jacobowitz D, Spear C, Oettgen HF 1975. The acute and chronic effects of alcohol on the human immune system. *Surg Gynecol Obstet 141*: 212-218.

Lutz AV 1919. *Schistosoma mansoni* e a schistosomose, segundo observações feitas no Brasil. *Mem Inst Oswaldo Cruz 11*: 121-125.

MacGregor RR 1986. Alcohol and immune defense. *JAMA 256*: 1474-1479.

Maier A, Bode C, Fritz P, Bode JC 1999. Effects of chronic alcohol abuse on duodenal mononuclear cells in man. *Dig Dis Sci 44*: 691-696.

Messias IJT, Genta RM, Mohren WD 1994. Adherence of monocytes and polymorphonuclear cells to infective larvae of *Strongyloides stercoralis* after complement activation. *J Parasitol 80*: 267-274.

Moraes RG 1948. Contribuição para o estudo do *Strongyloides stercoralis* e da estrongiloidíase no Brasil. *Rev Saude Publica 1*: 507-624.

Nawa Y, Ishikawa N, Tsuchiya K, Horii Y, Abe T, Khan AI, Shi B, Itoh H, Ide H, Uchiyama F 1994. Selective effector mechanisms for the expulsion of intestinal helminths. *Parasite Immunol 16*: 333-338.

Pelletier G, Briantais MJ, Buffet C, Pillot J, Etienne JP 1982. Serum and intestinal secretory IgA in alcoholic cirrhosis of the liver. *Gut 23*: 475-480.

Roitt I, Brastoff J, Male D 1999. *Imunologia*, 5 ed., Manole, São Paulo, 423 pp.