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Bancroftian filariasis in an endemic area of Brazil: differences between genders during puberty

Filariose bancroftiana em uma área endêmica do Brasil: diferenças entre os sexos durante a puberdade

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ABSTRACT

Gender differences in susceptibility to infectious diseases have been observed in various studies. A survey was performed in a bancroftian filariasis endemic area in the city of Olinda, Brazil. All residents aged 5 years or older were examined by thick blood film. People aged 9 to 16 years were interviewed and also tested for filarial antigenaemia. Data were analyzed by contingency table methods and regression models. The risk of microfilaraemia for males was significantly higher. Among those aged 9 to 16 years, the analysis of gender and filariasis by age showed that boys from 15 to 16 years had a higher risk of infection than girls. No association was found between menarche and filariasis in girls. The data suggest that variations between gender in filariasis could result, at least in part, from an increase in susceptibility of men. This epidemiologic feature needs to be considered while formulating elimination plans.

Key-words: *Filariasis. Gender differences. Puberty. Risk factors.*

RESUMO

Diferenças entre os sexos quanto à susceptibilidade às doenças infecciosas têm sido observadas em vários estudos. Um inquérito de prevalência foi realizado em uma área endêmica de filariose bancroftiana na cidade de Olinda, Brasil. Todos os residentes com idade ≥ 5 anos foram examinados pela gota espessa. Moradores com idade entre 9 e 16 anos foram entrevistados e testados para a presença de antigenemia filarial. Os dados foram analisados utilizando tabelas de contingência e modelos de regressão. O risco de microfilaraemia nos homens foi significativamente mais elevado. Meninos com idade entre 15 e 16 anos tiveram maior risco de infecção filarial do que as meninas. Os dados sugerem que variações entre os sexos na filariose podem resultar de um aumento na susceptibilidade dos homens a partir da puberdade tardia. Essa característica epidemiológica deve ser considerada ao se formularem os planos de eliminação da endemia.

Palavras-chaves: *Filariose. Diferenças entre sexos. Puberdade. Fatores de risco.*

Gender differences in susceptibility to several diseases have been observed in a number of studies[11 29]. In humans, epidemiological, laboratory and clinical studies have shown variations between gender in parasitic diseases including schistosomiasis, filariasis, leishmaniasis and onchocerciasis, as well as other infections such as rubella, measles, hepatitis B, and tuberculosis[11].

Filarial surveys carried out in Brazil and other endemic countries have observed a greater expression of infection and morbidity in males[4 9 12 15 25]. Usually microfilaraemia and filarial antigenaemia, parasitic burdens and clinical manifestations are more frequently observed among males than females. Different levels of exposure to infected mosquitoes might explain these patterns. However, epidemiologic studies which investigated the influence of socioeconomic factors, occupational exposure, individual protection and type of clothing did not confirm their effect[5].

Differences between gender regarding the occurrence of diseases have also been related to physiological causes,

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particularly hormonal and genetic ones[14 23 24 26 28]. For filariasis, some studies have suggested a possible association between initiation of sexual hormone production and changes in susceptibility to infection. Mavoungou et al[16] compared levels of estrogen, progesterone and testosterone among microfilaraemic and amicrofilaraemic girls aged 14 to 16 years and reported lower levels of sexual hormones in microfilaraemic carriers. Brabin[4], reviewing surveys carried out in different continents, observed that the prevalence of microfilaraemia, and parasite burden, were significantly lower in females than males. The differential sex effect typically started at age 15, corresponding to the beginning of women's reproductive lives. This observation led the authors to conclude that hormonal factors related to pregnancy might limit the fertility of the adult worms, or even make women more resistant to filarial re-infection. Alexander and Grenfell[3] reported a reduction in the parasitic burden among women during the reproductive period in an endemic population of Papua New Guinea, although the comparison between pregnant women and controls demonstrated no evidence of a relationship between parasite burden and pregnancy.

In spite of efforts to clarify the mechanisms behind gender differences in lymphatic filariasis, there is no consensus as to whether the differences are better explained by an environmental or immunological hypothesis, that is, as a consequence of less exposure to the vector, or increased resistance to the parasite[5 7]. This study investigated the associations of biological and environmental factors to bancroftian filariasis between genders during puberty. Among girls, menarche was used as a proxy for estrogen production.

MATERIAL AND METHODS

The study was carried out in Olinda, a city in Northeastern Brazil, where a previous parasitological baseline survey found a spatial cluster of filariasis cases[6]. The area was mapped and data were collected through a door-to-door survey performed between December 1999 and September 2000.

All households were registered and residents aged 5 years and older were invited to be examined by thick film technique. 60μl blood samples were drawn between 9 p.m. and midnight. Participants between the ages of 9 and 16 years were asked to provide an additional blood sample to test filarial antigenaemia and then were given a questionnaire to obtain information about time of residence in the area, use of a bednet, occurrence of menarche, history of pregnancy and use of hormonal contraceptives.

Circulating filarial antigen was checked using either the immunochromatographic card test (AMRAD ICT, New South Wales, Australia) or Og4C3-ELISA (TropBio, Townsville, Australia), which have similar principle and accuracy[17].

The outcome variables were the prevalence of microfilaraemia (mf), the prevalence of circulating filarial antigen (CFA), and microfilarial density. Gender and age-specific prevalence of microfilaraemia and filarial antigenaemia were estimated. The mean microfilarial density was analyzed by negative binomial regression[10]. Logistic regression was used to test the association between gender and filarial infection (mf or CFA) adjusted for co-variables. Finally, logistic regression models were constructed separately for boys and girls. EPI INFO (version 6.0) and STATA (version 6.0) were the statistical programs used.

The project was approved by the Ethical Committee of the Centro de Pesquisas Aggeu Magalhães/Fiocruz Foundation. All participants were informed about the objectives of the study and signed a consent form. Individual data and blood exams for participants under 18 years old were only performed after approval of a parent or guardian.

RESULTS

Epidemiological pattern of bancroftian filariasis in the whole population. Among 5,258 residents that took part in the parasitological survey, 328 (6.2%) were positive by thick film technique. There was a statistically significant association between sex and mf (χ^2_1 = 33.19; p <0.0001). The risk for males was significantly higher than the risk for females, even after adjustment by age group (adjusted OR = 1.94; 95% CI: 1.55-2.43; p<0.0001).

In males, the mf prevalence varied with age; it was 5.4% in the 5 to 9 age group, increased two-and-a-half times in the 15 to 19 age group, remained constant through the 20 to 29 age group and decreased to 4.7% by age 50. These differences were statistically significant (χ^2_6 = 29.99; p <0.0001) (Figure 1). Among females, the mf prevalence that was 3.9% in the 5 to 9 age group rose to 6.0% in the 15 to 19 group and then steadily decreased until reaching 2.8% in those over the age of 50. These differences in female age-specific prevalence were not statistically significant (χ^2_6 = 6.56; p = 0.363) (Figure 1). The test of interaction between sex and age group was performed, but no significant difference was observed (χ^2_6 = 7.62; p = 0.267). The estimated mean microfilaraemic density of 1.63 (95% CI: 1.18-2.27) mf/60ml for males was significantly higher than the value of 0.45 (95% CI: 0.34-0.60) mf/60ml observed in females (χ^2_1 = 30.48; p <0.0001).

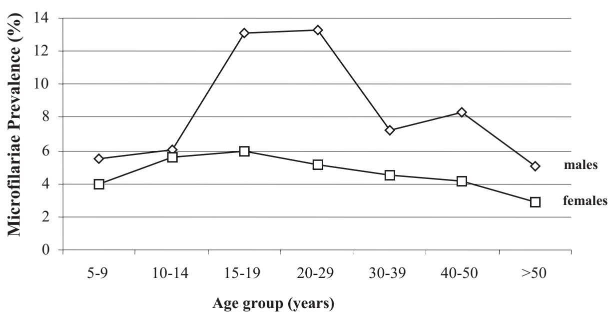
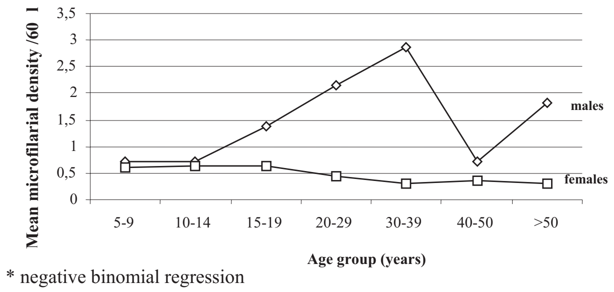


Figure 1 - Microfilaraemia prevalence, according to age and sex among the total study population*.

The mean microfilarial density in males was similar to that in females until the 10-14 age group, at which point male mean density increased sharply, peaking between the ages of 30 and 39 and decreasing from this point onward (χ^2_6 = 12.74; p = 0.04). Among females, the mean microfilarial density remained relatively stable through the 15-19 year age group, after which a slight decline was observed (χ^2_6 = 3.63; p = 0.726) (Figure 2).

Relation between filarial infection, biological factors and bednet use in 9-16 year olds. From the 1,511 boys and girls aged 9 to 16 registered in the area, 1,130 (72.7%) were interviewed and examined by thick film. Of those taking part, 790 (70%) were also tested for CFA by ICT card test (674) or Og4C3-ELISA (116). Those who were tested for CFA did not differ significantly from those who were not tested in age, sex or mf prevalence; this finding ensures the comparability of the two groups (data not shown).



* Number of observations: males = 2,181 and females = 3,071

Figure 2 - Mean microfilarial density, according to age and sex among the total study population.*

Among the 608 girls who provided information about menarche, 303 (49.8%) reported its occurrence, 16 (2.6%) had previously been pregnant and 5 (0.8%) were using hormonal contraceptives.

In the 9 to 16-year-old population, the overall prevalence of microfilaraemia was 6.9% (95% CI: 5.5-9.8) and that of antigenaemia was 25.7% (95% CI: 23.2-30.7).

The crude odds ratio (OR) for the association between mf and sex indicated a higher risk of mf among boys (OR = 1.60; 95% CI: 1.01-2.55). The age-adjusted OR (OR = 1.63; 95% CI: 1.02-2.60) was similar to the crude OR, indicating little, if any, confounding effect. For CFA, the difference was not statistically significant before or after age adjustment. The age-stratified analyses of sex and both mf and CFA showed that boys aged 15 to 16 years had a higher risk of filarial infection than girls of the same age (Table 1).

A statistically significant interaction was observed between age and sex when the outcome was CFA, suggesting that, at least for filarial antigenaemia, age modified the association with sex (χ^2_3 = 9.55; p = 0.02) (Table 1).

Table 1- Odds ration and 95% CI for the association between filarial infection, sex and age group.

	Microfilaraemia (n = 1,130) OR (95% CI) boys versus girls	Antigenaemia (n = 790) OR (95% CI) boys versus girls
Sex	1.60 (1.01-2.55)	1.25 (0.91-1.72)
Age group (years)		
9 – 10	1.80 (0.73-4.37)	1.70 (0.87-3.31)
11 – 12	0.91 (0.37-2.22)	0.96 (0.51-1.81)
13 – 14	1.17 (0.37-3.73)	0.65 (0.33-1.26)
15 – 16	3.21 (1.30-7.96)	2.47 (1.30-4.70)
Age adjusted OR	1.63 (1.02-2.60)	-
	Interaction test:=4.24; p=0.241	*Interaction test =9.65; p=0.022*

In order to examine the potential confounding effect of environmental factors in the association between filarial infection and sex, the effect of bednet use and the presence of a microfilaraemic adult in the household was tested. The magnitude of the adjusted associations between sex and mf (OR = 1.64; 95% CI: 1.02-2.64) and CFA (OR = 1.31; 95% CI: 0.95-1.82) were similar to those obtained prior to adjustment. Bednet use was also tested as an effect modifier in the association between sex and mf, but the result was not statistically significant (χ^2_1 = 2.34; p = 0.12).

Due to the observed interaction between age and sex and also the fact that menarche only occurs in females, the association between filarial infection and co-variables was estimated separately for each sex. No association was found between menarche and mf (OR = 0.65; 95% CI: 0.16-2.68) or CFA (OR = 0.90; 95% CI: 0.31-2.63). Moreover, not using a bednet did not increase risk of mf (OR = 1.64; 95% CI: 0.56-4.81) or CFA (OR = 1.76; CI 95%:0.94-3.31) in girls. In boys, however, not using a bednet was associated with an increased risk of mf (OR = 6.24; 95% CI: 1.47-26.45) and CFA (2.66; 95% CI: 1.38-5.10). The presence of a microfilaraemic adult in the household was a risk of mf for both boys (OR = 3.10; CI 95%:1.46-6.50) and girls (OR = 3.57 95% CI: 1.56-8.15), but was not associated with CFA.

DISCUSSION

In accordance with previous observations[4 9 12 15 25], this study showed a steady increase of both microfilaraemia and parasitic burden in males beginning at age 14. It is known that the sensitivity of thick film increases with higher microfilarial density[17]. Since females usually exhibit lower parasite burdens than males[4], differential misclassification may underestimate the microfilarial prevalence in females and contribute to an apparent gender difference. In this study, analysis of the association between gender and filarial infection during puberty demonstrated that, although the prevalence of microfilaraemia was significantly higher in boys than in girls, this difference was not observed for filarial antigenaemia. This finding is consistent with the lower sensitivity of the thick film technique in females.

Assuming higher validity of the filarial antigenaemia results, the crude data do not suggest a gender difference in the 9-16 age range. Nevertheless, the stratified analysis by age showed a significantly higher risk of filarial infection in 15 to 16-year-old boys. The test for an age-sex interaction reinforced this finding that that the effect of sex on filarial antigenaemia varies by age group. Therefore, it seems that changes in susceptibility to filarial infection might occur in men during the later stages of puberty. This phenomenon could be explained by the influence of either biological or behavioral factors acting in this period of life.

Culex quinquefasciatus is an endophilic mosquito of nocturnal habits that usually breeds in areas around dwellings. Although information concerning its biting behavior in Brazil is lacking, an entomological study in India found that the peak biting period of *Culex* ranged between midnight and 3 a.m.[8]. Considering this, different behavioral patterns between genders, such as current use of bednets, fans or insecticides while sleeping, could be related to the differences between sexes in filarial infection. Nevertheless, the analysis of the association between sex and filarial infection during puberty suggests that, at least for microfilaraemia, the association between these variables occurred independently of the use of bednets and the presence of a microfilaraemic adult in the household. This result is in accordance with a previous epidemiological study carried out in a neighboring city, Recife, in which the filarial infection risk remained twice as high in men, even after adjusting for other exposure variables[1]. Therefore, despite the difficulties of measuring human exposure to infection and in controlling for all the relevant behavioral factors[7], the data suggest that individual protection may not play a decisive role in the gender differences in lymphatic filariasis.

In girls, the occurrence of menarche, used as a proxy for increased estrogen production, was not protective against filarial infection. Moreover, age and non-use of bednet did not constitute risks of infection for this sex. These data suggest that pubescent changes, whether hormonal or behavioral, do not influence female susceptibility to filarial infection**.** These results reproduce, to some extent, experimental studies that investigated the role of sexual hormones on the response to certain infectious diseases in animals. The removal of ovaries in female rats did not produce any effect on the intensity of the parasitism[2 22]. Conversely, the administration of testosterone or removal of the testis in male rats did alter the response to parasitic infection[18 19 20].

Therefore, our data suggest that the observed variations between genders in bancroftian filariasis could result, at least in part, from increased susceptibility, possibly of physiological origin, of men in the late stages of puberty.

Research has shown that men are often more susceptible to infections caused by parasites, fungi, bacteria and viruses[11]. In addition, mortality rates are usually higher in males than in females[21]. Paradoxically, the use of health services is less frequent in the male population[13 27]. These social and biological particularities of genders have not been accounted for in the formulation of public health policies to prevent, control or eliminate diseases. For lymphatic filariasis, males not only exhibit more signs and symptoms of lymphatic damage, they also typically express more microfilaraemia and higher parasite burdens. These features characterize men as an important reservoir of filarial parasites that could maintain and spread disease in the community. Therefore, communication and educational approaches for elimination programs should focus on males by promoting their compliance with the mass chemotherapy for lymphatic filariasis and encouraging those with signs and symptoms to seek early diagnosis in order to prevent more severe forms of filariasis.

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REFERENCES

1. Albuquerque MFM, Marzochi MC, Ximenes RA, Braga MC, Silva MC, Furtado AF. Bancroftian filariasis in two urban areas of Recife, Brazil: the role of individual risk factors. Revista do Instituto de Medicina Tropical de São Paulo 37**:** 225-233, 1995.
2. Alexander J, Stimson W. Sex hormones and the course of parasitic infection. Parasitology Today 4: 189-193, 1988.
3. Alexander N, Grenfell B. The effect of pregnancy on *Wuchereria bancrofti* microfilarial load in humans. Parasitology 119: 151-156, 1999.
4. Brabin L. Sex differentials in susceptibility to lymphatic filariasis and implications for maternal child immunity**.** Epidemiology and Infection 105: 335-353, 1990.
5. Brabin L, Brabin BJ. Parasitic infections in women and their consequences. Advances in Parasitology 31: 1-81, 1992.
6. Braga C, Ximenes RA, Albuquerque M, Souza WV, Miranda J, Brayner F, Alves L, Silva L, Dourado I. Avaliação de um indicador sócio-ambiental utilizado no rastreamento de áreas de transmissão de filariose linfática em espaços urbanos. Cadernos de Saúde Pública 17: 1211-1218, 2001.
7. Bundy D. Gender-dependent patterns of infection and disease. Parasitology Today 4: 196-189, 1998.
8. Chandra G. Short report: peak period of filarial transmission. The American Journal of Tropical Medicine and Hygiene 53: 378-379, 1995.
9. Chanteau S, Glaziou P, Plichart C, Luquiaud P, Moulia-Pelat JP, N'guiyen. *Wuchereria bancrofti* filariasis in French Polynesia: age-specific patterns of microfilaremia, circulating antigen, and specific IgG and IgG4 responses according to transmission level. International Journal of Parasitology 25: 81-85, 1995.
10. Elliot JM. Some methods for statistical analysis of samples of benthic invertebrates. Freshwater Biology Association, Cumbria, 1977.
11. Goble FC, Konopka EA. Sex as a factor in infectious disease. Transactions of the New York Academy of Sciences 35: 325-346, 1973.
12. Itoh M, Weerasooriya MV, Gunawardena NK, Mudalige MP, Samarawickrema WA, Kimura E. Wuchereria bancrofti antigenaemia in Sri Lanka. Tropical Medicine and International Health 4: 207-210, 1999.
13. Kandrack M, Grant KR, Segall A. Sex differences in health related behavior: some unanswered questions. Social Science and Medicine 5: 579-590, 1991.
14. Klein JL. The effects of hormones on sex differences in infection: from genes to behavior. Neuroscience and Biobehavioral Review 24: 627-638, 2000.
15. Maciel A, Rocha A, Marzochi K, Medeiros Z, Carvalho AB, Regis L, Souza W, Lapa T, Furtado A. Epidemiological study of bancroftian filariasis in Recife, northeastern Brazil. Memórias do Instituto Oswaldo Cruz 91: 449- 455, 1996
16. Mavoungou D, Lansoud-Soukate J, Dupont A. Steroid and gonadotropin hormone levels in young African women with filarial infection. Journal of Steroid and Biochemistry 34: 577-580, 1989.
17. McCarthy J. Diagnosis of lymphatic filarial infection. *In*: Nutman TB (ed) Lymphatic Filariasis, Imperial College Press, London, p. 127-150, 2000.
18. Mock BA, Nacy CA. Hormonal modulation of sex differences in resistance to *Leishmania major* systemic infections**.** Infection and Immunity 56**:** 3316-3319, 1988.
19. Nakanishi H, Horii Y, Terashima K, Fujita K. Effect of testosterone on the susceptibility of C57BL/6 mice to infection. Journal of Parasitology 75**:** 455-460, 1989.
20. Nakazawa M, Fantappie MR, Freeman GL, Eloi Santos S, Olsen NJ, Kovacs WJ, Secor WE, Colley DG. Schistosoma mansoni: susceptibility differences between male and female mice can be mediated by testosterone during early infection. Experimental Parasitology 85**:** 233-240, 1997.
21. Owens IPF. Sex differences in mortality rate. Science 297: 2008-2009, 2002.
22. Roberts C, Satoskar A, Alexander J. Sex Steroids, Pregnancy-associated Hormones and Immunity to Parasitic Infection. Parasitology Today 12: 382-388, 1996.
23. Roberts C, Walker W, Alexander J. Sex-associated hormones and immunity to protozoan parasites. Clinical Microbiology Reviews 14: 476-488, 2001.

24. Schuurs AH, Verheul HA. Effects of gender and sex steroids on the immune response. Journal of Steroid and Biochemistry 35: 157-172, 1990.

25. Steel C, Ottesen EA, Weller PF, Nutman TB. Worm burden and host responsiveness in *Wuchereria bancrofti* infection: use of antigen detection to refine earlier assessments from the South Pacific. The American Journal of Tropical Medicine and Hygiene 65: 498-503, 2001.

26. Styrt B, Sugarman B. Estrogens and infection. Reviews of Infection Disease 13: 1139-1150, 1991.

27. Travassos C, Viacava F, Pinheiro R, Brito A. Utilização dos serviços de saúde no Brasil: gênero, características familiares e condição social. Revista Panamericana de Salud Publica 11: 365-373, 2002.

28. Verthelyi D. Sex hormones as immunomodulators in health and disease. International Immunopharmacology 1: 983-993, 2001.

29. Zuk M, McKean KA. Sex differences in parasite infections: Patterns and processes. International Journal of Parasitology 26: 1009-1023, 1996.