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Original paper

Intestinal parasitoses and associated factors in a Brazilian city of German's descendants: a population-based study

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ABSTRACT. Intestinal parasitoses are a recurrent public health problem in developing countries. Their occurrence is usually associated with poor socioeconomic status and environmental risk factors. This study aims to estimate the prevalence of intestinal parasites and associated risk factors in Pomerode, SC. This was a cross-sectional sample with participants from population-based cohort study SHIP-Brazil (n=2,488). Sociodemographic, lifestyle, and environmental variables were selected from the study database. The stool samples were analyzed by Hoffman, Pons, and Janer, and by Faust methods. We estimated the relative frequency of parasites and its association to the study variable was estimated by prevalence ratio (PR) in a Poisson regression model. Among those who provided stool sample (n=797), the prevalence was 10.8% (95% CI 8.6 to 13.4). *Endolimax nana*, 4.8% (95% CI, 3.5–6.7) was the most frequent parasite, followed by *Entamoeba histolytica/dispar*, 1.7% (95% CI, 0.8–3.3), *Urbanorum* spp., 1.6% (95% CI, 1.0–2.7). Men (PR=1.9 95% CI 1.2–2.9), olders (PR=1.7 95% CI 1.0–2.8), non-white (PR=1.9 95% CI 1.2–3.0), living in high-risk dwelling areas (PR=1.8 95% CI 1.4–2.4) were associated with elevated proportions of parasitosis in the adjusted model. The current study found a low frequency of intestinal parasitoses in Pomerode, SC, Brazil. This frequency was higher among males, older, who live alone, non-white, in low SES, and living in high-risk dwelling areas.

Keywords: intestinal parasitosis, epidemiology, public health, socioeconomic factors

Introduction

Intestinal parasitic infections are recurrent problems in underdeveloped and developing countries, mainly in tropical and subtropical regions, affecting populations living in poverty [1]. The occurrence of parasitoses is directly related to the lack of basic sanitation, ineffective water treatment, and insufficient hygiene [2]. Infected water resources result from ineffective water treatment contaminated by the dumping of human and animal waste [3]. Overall, it is estimated that more than a quarter of the world's population is at risk of helminth infections [4].

Since the 80s, Brazil has been experiencing an epidemiological and demographic transition process. While the population life expectancy increased and infant mortality and infectious diseases decreased, the number of chronic diseases, such as hypertension and diabetes mellitus are increasingly common [5]. On the other hand, the country still has a high incidence of infections with intestinal parasites, especially in regions with a lack of information, basic sanitation and water treatment,

and low socioeconomic status [1]. Moreover, distinct regions present epidemiological variations, such as rural and urban areas, presenting uneven prevalence of intestinal parasites [6,7].

The agents that cause these infections may be non-pathogenic, such as *Endolimax nana* and *Entamoeba coli*, or pathogenic, such as *Giardia duodenalis*. This last protozoan is associated with infectious gastrointestinal diseases, anemia, and malnutrition [8]. In association with this transitional scenario, new disease-causing agents have been emerging, such as *Urbanorum* spp. This parasite was first reported in 1994 in Colombia, with the first case in Brazil reported only in 2018 [9,10]. Although the life cycle and forms of tissue invasion are not well-known, it is often associated with acid pH diarrhea, abdominal pain, and vomiting, which can result in malnutrition and dehydration [11].

Pomerode is a small city in Southern Brazil (26º44'27" S and 49º10'33"W) with around 30,000 inhabitants, with a relatively high Human Development Index (HDI) of 0.780 and Gross Domestic Product (GDP) per capita around US$ 17,900.00. Around 58.5% of the households have access to treated water and 91.7% have sewage treatment in the urban area [12]. Despite these numbers, prevalence and mortality due to preventable infectious diseases remain high [8,13].

We aimed to estimate the number of people infected with intestinal parasites, the most frequent and their risk factors in Pomerode, Brazil.

Materials and Methods

The study „Life and Health in Pomerode – SHIP-Brazil" is a population-based cohort, whose baseline examination was conducted between 2014 and 2018. The study included people of both sexes, between the ages of 20 and 79 years, and registered as dwellers in the city of Pomerode for at least 6 months. The exclusion criteria were: 1) people with significant physical or cognitive impairment or mental illness that could not answer the interviews or go to the Examination Center (EC) at the University Hospital (UH); 2) those who did not speak Portuguese. The drawing sample had 12 strata, 2 regarding the sexes and 6 the ages from 20 to 79 years old. The sample size calculation considers the population in each stratum, 50% prevalence of events, precision of 6%, and 95% confidence level, resulting in 3,678 selected participants. The response rate was 67.7%.

Personal interviews were performed with a tested questionnaire with 2,488 participants. All study participants gave informed consent. The study variables were: sex, age (in years), marital status, race/color self-reported, German culture (to speak German at home and to participate in folk clubs), education (college, high school, elementary school, and illiterate), economic class group (A1/A2, B1/B2, C1/C2, D/E) [14], neighborhood, source of treated water (yes/no) and sewage treatment (yes/no).

A three-stratum synthetic variable named Socioeconomic Status (SES) resulted from education and economic class data. Participants were considered in the low stratum if: C1/C2 class groups and illiterate education or; from D/E and the illiterate up to the 4th grade of elementary school. Participants were considered from the high stratum if: A1/A2 class and with undergraduate or high school education; or from B1/B2 class group with undergraduate education. All others were considered to belong to the middle stratum.

Participants were grouped in a dichotomous variable named dwelling area according to the neighborhood frequency of parasites (high or low risk).

Among the 2,488 participants, 802 (32.2%) provided a stool sample, which was collected at the participant's home according to guidelines [15], but 5 (0.6%) samples were excluded due to insufficient material.

The sample was collected in a proper vial with a preservative medium for stool (modified Merthiolate-Iodo-Formol, mMIF). The sample was analyzed at the UH Central Laboratory using two methods: by Hoffman, Pons, and Janer, and by Faust methods. Hoffman, Pons, and Janer method [16,17] uses the principle of spontaneous stool sedimentation. It is prepared with 2 g of stool mixed with 5 ml of water and then supplemented with another 20 ml of water for complete homogenization. The suspension is filtered into a glass through a strainer and surgical gauze. Therefore, the material is filled with water and left to rest for at least 2 hours. A sample is collected and deposited on a slide colored with Lugol and then visualized by an optical microscope. This method was used to detect protozoan cysts and helminth eggs. The Faust method [16,17] uses centrifuge-flotation to detect light protozoan cysts. 10 ml of the suspension previously prepared by the Hoffman method were transferred to a conical tube and centrifuged for 2 minutes at

Table 1. Sample characteristics, SHIP-Brazil, Pomerode, SC, 2014–2018 (n=797)

Variable	%	(95% CI)	Dwelling area		
			High risk	Low risk	*P*
Sex					
Male	45.9	(42.6 – 49.3)	47.1	44.9	
Female	54.1	(50.7 – 57.4)	52.9	55.1	0.589
Age					
Mean	46.2	(46.2 – 47.2)	47.3	45.2	
Marital status					
Married	76.3	(72.6 – 79.6)	76.7	75.9	
Single	14.3	(11.3 – 17.8)	12.5	15.8	
Divorced	4.6	(3.3 – 6.4)	5.1	4.2	
Widower	4.9	(3.8 – 6.2)	4.7	4.1	0.467
Self-reported race/color					
White	92.8	(90.4 – 94.6)	92.2	93.3	
Non-white	7.2	(5.4 – 9.6)	7.8	6.7	0.617
German culture					
Yes	65.2	(61.3 – 69.0)	65.2	65.2	
No	34.8	(31.0 – 38.7)	34.8	34.8	0.994
Education					
Illiterate	1.0	(0.6 – 1.8)	1.2	0.8	
Elementary 1 to 4 series	31.4	(28.3 – 34.6)	35.4	27.8	
Elementary 5 to 8 series	21.0	(18.0 -24.4)	21.4	20.7	
High school	32.0	(28.2 – 36.0)	34.2	30.0	
College	14.6	(11.8 – 17.9)	7.7	20.7	<0.001
Consumption class					
A1/A2	20.8	(17.8 – 24.2)	27.7	14.5	
B1/B2	63.5	(59.7 – 67.1)	54.0	72.1	
C1/C2	15.2	(12.9 – 18.0)	17.5	13.3	
D/E	0.5	(0.0 – 1.0)	0.8	0.2	<0.001
Socioeconomic status					
Better	19.9	(16.6 – 23.6)	17.6	22.0	
Medium	71.0	(67.2 – 74.6)	71.4	70.7	
Worse	9.1	(7.4 – 11.1)	11.3	7.4	0.144
Treated water source					
Yes	73.0	(69.3 – 73.4)	61.8	82.8	
No	27.0	(26.3 – 30.7)	38.2	17.2	<0.001
Sewage treatment					
Yes	84.7	(81.6 – 87.3)	82.2	86.9	
No	15.3	(12.7 – 18.4)	17.8	13.1	0.106

Source: SHIP-Brazil, 2014-2018 CI: Confidence Interval. All analyses are weighted with the product of poststratification weights and inverse probability of participation weights

1,500 rpm. The supernatant was then discarded, and the pellet resuspended. Then, 10 ml of water was added, and the tube was centrifuged again. After the supernatant become clear, it was discarded, and the pellet was resuspended with a 33% zinc sulfate solution, completing the conical tube up to 10 ml. The material was centrifuged for 2 minutes at 1,500 pm, resulting in a film formed on the surface. This film colored with Lugol was analyzed by an optical microscope (10× and 40×).

The main study outcome is the relative frequency of parasites. The secondary outcomes

Table 2. Bivariate analysis of the distribution of parasitic prevalence and the study variables, SHIP-Brazil, Pomerode, SC, 2014–2018 (n=797)

Variable	Parasitosis	Prevalence ratio		
	Yes	No	(95% CI)	*P*
Sex				
Female	91.8	8.2	1	
Male	85.9	14.1	1.7 (1.1 – -2.7)	0.014
Age				
20 to 29	91.5	8.5	1	
30 to 39	92.7	7.3	0.9 (0.3 – 2.6)	
40 to 49	87.8	12.2	1.4 (0.6 – 3.7)	
50 to 59	86.2	13.	1.6 (0.6 – 4.2)	
60 to 69	88.4	11.6	1.4 (0.5 – 3.6)	
70 to 79	84.9	15.1	1.8 (0.7 – 4.6)	0.510
Marital status				
Maried	90.6	9.4	1	
Live alone	85.4	14.6	1.5 (0.9 – 2.6)	0.094
Self-reported race/color				
White	90.0	10.0	1	
Non-white	80.0	20.0	2.0 (1.1 – 3.7)	0.031
German culture				
Yes	89.4	10.6	1	
No	89.1	10.8	1.0 (0.6 – 1.6)	0.928
Education				
Undergraduate	94.0	6.0	1	
High school	90.7	9.3	1.5 (0.5 – 4.4)	
Elementary 5 to 8 series	87.6	12.4	2.1 (0.74 – 5.7)	
Elementary 1 to 4 series	86.9	13.1	2.2 (0.8 – 5.7)	
Illiterate	77.0	23.0	3.8 (1.0 – 14.9)	0.277
Consumption class				
A1/A2	89.8	10.2	1	
B1/B2	89.4	10.6	1.0 (0.6 – 1.9)	
C1/C2	89.1	10.9	1.1 (0.5 – 2.1)	
D/E	49.0	51.0	5.0 (2.0 – 12.8)	0.003
Socioeconomic status				
High	92.7	7.3	1	
Medium	89.1	10.9	1.5 (0.7 – 3.2)	
Low	82.5	17.5	2.4 (1.1 – 5.5)	0.073
Dwelling area				
Low risk	95.0	5.0	1	
High risk	82.6	17.4	1.9 (1.5 – 2.4)	<0.001
Treated water source				
Yes	91.4	8.6	1	
No	83.9	16.1	1.9 (1.2 – 2.9)	0.007
Sewage treatment				
Yes	89.4	10.6	1	
No	88.8	11.2	1.1 (0.6 – 1.9)	0.844

Source: SHIP-Brazil, 2014-2018 CI: Confidence Interval. All analyses are weighted with the product of poststratification weights and inverse probability of participation weights

Table 3. Models of the association between the presence of parasitosis and study variables, SHIP-Brazil, Pomerode, SC, 2014–2018 (n=797)

Variable	PR crude		PR ajusted	
	PR (CI 95)	p	PR (CI 95)	*P*
Dwelling area				
Low risk	1		1	
High risk	1.9 (1.5 – 2.4)	<0.001	1.8 (1.4 – 2.4)	<0.001
Sex				
Female	1		1	
Male	1.7 (1.1 – 2.7)	0.014	1.9 (1.2 – 2.9)	0.004
Age				
Younger	1			
Older	1.4 (0.9 – 2.1)	0.117	1.7 (1.0 – 2.8)	0.032
Marital status				
Married	1		1	
Live alone	1.5 (0.9 – 2.6)	0.094	1.9 (2.0 – 5.9)	<0.001
Self-reported race/color				
White	1		1	
Non-white	2.0 (1.1 – 3.7)	0.031	1.9 (1.2–3.0)	0.004
Treated water source				
Yes	1		1	
No	1.9 (1.2 – 2.9)	0.007	1.4 (0.9 – 2.1)	0.178
Socioeconomic status				
High	1		1	
Medium	1.5 (0.7 – 3.2)	0.258	1.5 (08 – 3.1)	0.242
Low	2.4 (1.1 – 5.5)	<0.001	2.1 (0.9 – 4.6)	0.075

Source: SHIP-Brazil, 2014-2018 PR: prevalence ratio; CI: Confidence Interval. All analyses are weighted with the product of poststratification weights and inverse probability of participation weights

were the amount of parasites and their species.

The questionnaire and exams were processed by double entry using Epidata software. Quality assurance included checking for losses, duplication, and inconsistencies (extreme values, e.g.).

The study sample of SHIP-Brazil differed in age and gender compared to the population of Pomerode. Inverse probability weighting was applied in order to minimize this possible bias [18]. Initially, the probability of being selected for the study was calculated for each participant. Then, the weight, which is the inverse of the selection probability, was computed and included in the analysis. Thus, each selected participant was accounted not only for its value for itself but also for those with characteristics that were not selected. So, the data were described and analyzed with the weightings for 12 strata of gender (2) and age group (6).

The variables were presented using descriptive statistics. The associations of secondary outcomes and the study variables were estimated using the Chi-square test or Fisher's exact test.

The prevalence ratio estimated the association between the main outcome (presence of parasitosis) and the study variables. Those variables with a *P*-value<0.20 in a Poisson regression bivariate analysis were included in the multivariate analysis. A *P*-value<0.05 was considered statistically significant. Statistical analyses were performed using Stata 11.2 (Stata Corporation, College Station, TX, USA).

The study was approved by the Research Ethics Committee of the University of Blumenau (protocol

number 3214522) and complies with the Declaration of Helsinki.

Results

Characteristics of the study participants and their association with sites of risk for parasitosis are provided in table 1. Most of the participants were female (54.1%) with a mean age of 46.2 years. Most subjects were white (92.8%), had German culture (65.2%), elementary education (52.4%) and economic class B1/B2 (63.5%). The participants lived in the following neighborhoods: Testo Rega (33.0%), Centro (14.6%), Ribeirão Areia (14.6%), Testo Central (11.6%), Wunderwald (11.6%), Ribeirão Clara (7.6%), Pomerode Fundo (5.0%) and Testo Alto (2.0%). Housing had sewage treatment and treated water in 84.7% and 73.0%, respectively. There were no statistical differences among dwelling areas regarding sex, age, marital status, ethnicity, presence of German culture or sewage treatment.

The overall relative frequency of parasitoses was 10.8% (95% CI, 8.6 to 13.4) with 9.8% (95% CI, 7.8 to 12.3) of those affected having one parasite and 1.0% (95% CI, 0.5 to 1.9) two or more parasites. The following parasites were detected: *Endolimax nana*, 4.8% (95% CI, 3.5 to 6.7), followed by *Entamoeba histolytica* complex, 1.7% (95% CI, 0.8 to 3.3), *Urbanorum* spp., 1.6% (95% CI, 1.0 to 2.7), *Entamoeba coli*, 1.2% (95% CI, 0.6 to 2.1), *Iodamoeba* spp., 1.2% (95% CI, 0.6 to 2.2), *Giardia duodenalis*, 0.9% (95% CI, 0.3 to 2.0), *Strongyloides stercoralis*, 0.4% (95% CI, 0.1 to 1.1), *Entamoeba hartmanni*, 0.2% (95% CI, 0.0 to 1.0) and *Blastocystis* spp., 0.1% (95% CI, 0.0 to 0.5). There were no associations between these parasites and all the study variables.

The neighborhoods with a higher proportion of parasitoses were Testo Rega (18.4%), Ribeirão Clara (17.0%), Testo Alto (14.5%), and Pomerode Fundos (12.7%). On the other hand, the places with a lower proportion of parasitoses were Testo Central (2.6%), Wunderwald (4.9%), Ribeirão Areia (5.5%), and Centro (6.5%).

Table 2 shows the distribution of the relative frequency of parasitoses and other study variables. The risk of parasitoses was higher among males, nonwhite, in the worst SES, and with no treated water.

Table 3 presents the results of the adjusted multivariate analysis model. Men, of older age, non-white, of low socioeconomic status, and living in high-risk dwelling areas were associated with elevated frequencies of parasitoses in Pomerode.

Discussion

The overall relative frequency of intestinal parasites in this population-based sample from Pomerode-SC was low (10.8%) compared to other regions in Brazil. Most present monoparasitism and protozoans prevail. Our regression model identified a vulnerability profile (men aged 40 to 70, non-white skin color, living alone in high-risk dwelling areas and with low SES) being more affected by the presence of intestinal parasites.

The prevalence of parasitoses varies considering the characteristics of the studied population. For instance, in Southern Brazil schools, the prevalence of parasites can range from 2.74% to 55.44% [19–21]. Regarding the region analyzed, in the west of Santa Catarina the prevalence rate was 12.6%, and the most frequent protozoans had been *E. coli* and *Giardia duodenalis* [22]. The variability of the parasitoses prevalence in certain regions is usually related to socioeconomic and cultural conditions. The low frequencies here observed could be explained, at least in part, due to the city of Pomerode's relatively high HDI and GDP per capita when compared to most Brazilian cities [12]. Also, selection bias cannot be ruled out with persons with a higher socioeconomic class being more prone to participate.

Most of the parasites found in our study were protozoans. They were usually presented in rural areas with a poor water supply and sewage treatment that reinforce its faecal-oral dissemination. Some studies observed the highest prevalence of protozoans among the elderly with no treated water [23,24].

The frequency of *E. nana* in Pomerode is consistent with other Brazilian studies [3,25]. For instance, Santos and Merlini [25] identified *E. coli* in 3.5% of the samples collected in a small city, Maria Helena, of Parana State. However, in the northeastern region of Brazil, Calegar et al. [26] reported *E. coli* as the most prevalent parasitosis (11.3% of the cases). Pathogenic parasites have different prevalence in Brazilian states, such as *Giardia duodenalis*. being the most frequent [3,27].

Few studies provide information about *Urbanorum* spp. such as concerning life cycle stages or infectiveness. In Brazil, a sample pool

study from different regions detected a prevalence of 1.45% of this parasite [28]. On the other hand, Pino [29] found 20.8% of *Urbanorum* spp. in the population of Cajamarca, Peru. This prevalence could be explained, in part, due to the consumption of contaminated water and food [10,28]. Some authors described signs and symptoms such as acid pH stools, without blood mucus or leukocytes, and abdominal pain. However, these symptoms depend on the host's susceptibility and immune status. Notwithstanding, some individuals may be asymptomatic [30].

The relative frequency of parasitoses was associated with some characteristics of participants. Previous studies [23,31] found a higher prevalence of parasites among adult males, range 24.7% to 30%, both in urban and rural areas in different Brazilian regions. In our study, the high prevalence of parasitoses was found among men aged 40 to79 years. These findings might be explained to due men at this age being more exposed to labor and environmental risk conditions [32–34].

We detected a higher proportion of parasites among people living in high-risk areas which presented higher proportions of non-treated water supplies and non-sewage treatment. Some authors [6,7] reported prevalence varying from 7% to 83% of intestinal parasites in rural regions, higher than in urban areas (13% to 31%). These rates were associated with worse sanitary conditions [3]. On the other hand, some authors [7,35] emphasize that protozoan cysts and oocysts were observed even in treated water supplies. These findings might be explained due to a combination of many factors like an inadequate discharge of sewage, non-protected contact with animals (chicken and cattle breeding), lack of waste collection and the consumption of untreated water from natural sources [7,23].

Complementary to those environmental risk factors, our results suggest that low SES, as well as living alone and non-white skin color, were associated with a higher prevalence of intestinal parasites. Kabad et al. [36] reinforced the use of the race/color variable as a proxy of socioeconomic condition. In Brazil, it means that non-white people have a high probability to be poor than whites. Studies suggest that people with low income [37] and low schooling [38] present high rates of parasitoses. Low SES could represent a barrier to medical care and other health services like proper information and preventive measures [39]. Socioeconomically vulnerable people should be put first by public health services to reduce the impact of social inequalities in health.

This study has some limitations. First, this is a cross-sectional study and the associations here observed do not allow causal inferences. In addition, only 32% of participants provided stool samples for analyses. However, there was no statistically significant difference between the participants and non-participants considering sex, age, and place of residence. Finally, our results are based on just one sample of stool, which may have contributed to the low prevalence found.

Our results showed that the frequency of intestinal parasitoses in the city of Pomerode was low (10.8%), like in previous studies in Southern Brazil. The frequency was higher among male, older participants, alone living, non-white, in low SES, and living in high-risk living places. Primary Health Care Services should implement educational and preventive measures focused on this population at risk.

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