Intestinal parasitoses and clinical manifestations found in 3 to 12 years old schoolchildren in Argentina

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Abstract

We have carried out a study about the frequency of intestinal parasites and the presence of signs and symptoms in 3-12 year-old schoolchildren in the northeast region of the province of Buenos Aires, Argentina, during 2015-2016. For the parasitological study, serial stool analysis and serial anal scraping were done. Through a structured and closed survey applied, the age and sex of each child and the presence of clinical manifestations were recorded. We analized 857 schoolchildren, of which 63.95% were parasitized, with no differences in sex and age; 46.16% monoparasitized, 39% were biparasitized and with three or more parasites 14.84%. The parasitic species detected were: Enterobius vermicularis (50.36%), Giardia intestinalis (28.28%), Blastocystis hominis (61.86%), Ascaris lumbricoides (2.73%) and Trichuiris trichiura (0.91%). Clinical manifestations were registered in 77.59% of schoolchildren, without significant differences between parasitized and non-parasitized. There was a relationship between the number of symptoms/signs and "being parasitized" and between the presence of diarrhea and vomiting with G.intestinalis and B. Hominis. According to our results, we suggest: 1-perform parasitological analysis of children with some clinical manifestation and 2- perform parasitological analysis in asymptomatic children with risk factors associated with intestinal parasitosis.

Keywords: intestinal parasitoses, clinical manifestations, schoolchildren, Argentina.

Introduction

Infectious diseases are a significant cause of morbidity and mortality, and represent one of the major problems in world-wide public health (Gonzalez Montero et al., 2014). Among these diseases, intestinal parasitoses show high frequencies, especially in developing countries (Vieira da Silva et al., 2018).

Despite the global scientific and technical revolution that has taken place, as well as the advance in existing diagnostic, treatment and control measures; intestinal parasites appear as a challenge to modern medicine (Bakon et al., 2012). Though mortality due to these infections is relatively low, complications are common, being responsible for at least 10% of diarrheas and, in some cases, requiring hospitalization (Agudelo Lopez et al., 2008). These intestinal pathologies are closely related to socio-economical development processes, since poor absorption, diarrhea and blood loss reduce the growth of children (Mc Donald V 2003, Minvielle et al. 2016).

Bartolini et al. (2017) think the prevalence of these infections in non-endemic areas is still underestimated and this is due to characteristics of the infections such as mild or unspecific symptoms, an extended incubation period and, in other cases, insufficiency of the available lab methods. Besides, in non endemic areas, clinicians often have limited knowledge of these diseases. In urban and peri-urban populations, presence, persistence and dissemination of intestinal parasites are directly related to the geographical and ecological characteristics specific to the place, as well as to the basic sanitation conditions available and socio-economic and cultural factors. Therefore, controlling them may be a significant socio-political element (Rivero et al., 2017).

About 3,5 billion people are considered to be affected by intestinal protozoa and/or helminths, and approximately 450 million of them show symptoms of these infections. Soil-transmitted helminthiasis, amoebiasis and giardiasis are to be related to poverty (Valverde et al, 2011). According to Gamboa et al. (2014), intestinal protozoan diseases do not seem to restrict to weather conditions, socio-economic groups or geographical areas. B. Hominis and Giardia are considered the most frequent protozoa in the world population, both in symptomatic and asymptomatic people. These parasitoses may occur through environmental infection, ingestion of contaminated food, lack of environmental sanitation, or through ingestion of contaminated water; also, they are zoonotic parasites (Molina et al. 2011, Ciarmela et al. 2016).

In Argentina, intestinal parasitoses occur most frequently among the pediatric population. Some health professionals prescribe a parasitological analysis when they detect a questionable sign or symptom. But most of them remove the parasites in the child without ordering the corresponding parasitological analysis, and such behavior prevents knowing the prevalent species in each region and entails the risk of children developing a resistance to antiparasitic medication (Calentano A, 2012). Miller et al. (2003) have associated abdominal pain to the presence of Ascaris lumbricoides and Blastocystis hominis (Miller et al, 2003), in agreement with El Shazly et a.l (2005). Lotero O (2010) associates diarrhea with the presence of Giardia intestinalis, Uncinarias and –less intensely- B. Hominis. They mention recurrent abdominal pain occurs in cases of G. Intestinalis, A. Lumbricoides and Strongyloides stercoralis. Pruritus ani is associated by these authors to an infection with Enterobious vermicularis. To Gamboa et al. (2014), the presence of some symptoms was also associated to the prevalence of parasites, but only the presence of a symptom (yes or no) was inquired about. Limoncu et al. (2005) consider the association between specific symptoms and parasite species has been inconsistent, since some authors have found no association between symptoms and parasitic infection or have concluded that the presence of symptoms has low predictive value for the presence of pathogenic parasites.

The Program for the Control of Intestinal Parasitoses and Nutrition (PROCOPIN) of the Faculty of Medical Sciences of the National University of La Plata, has been working since 2009 in vulnerable communities in the cities of La Plata and Berisso, province of Buenos Aires, Argentina. This program is developed in four stages: 1. Evaluation of clinical and nutritional status, and detection of intestinal parasites; 2. Therapeutic intervention of children with clinical and nutritional alterations and/or parasites; 3. Educational intervention to prevent returning to the disease state; and 4. Post-intervention control.

In this article we present partial results from the 2015-2016 PROCOPIN first stage, and provide information about detection of intestinal parasitoses and presence of clinical manifestations in 3- to 12-year-old schoolchildren from the towns mentioned.

Materials and Methods

A cross-sectional, descriptive, analytic study was designed. The studied variables were intestinal parasitoses, gender, age, and presence of signs and symptoms in children.

Time and Area of Study: data collection was done during 2015 and 2016 in schoolchildren (ages 3 to 12) from the cities of La Plata (34º 56' 00" S, 57º 57' 00" W) and Berisso (34º 53' 00" S, 57º 54' 00" W), located in the Northeast region of the province of Buenos Aires, Argentina (Figure 1).

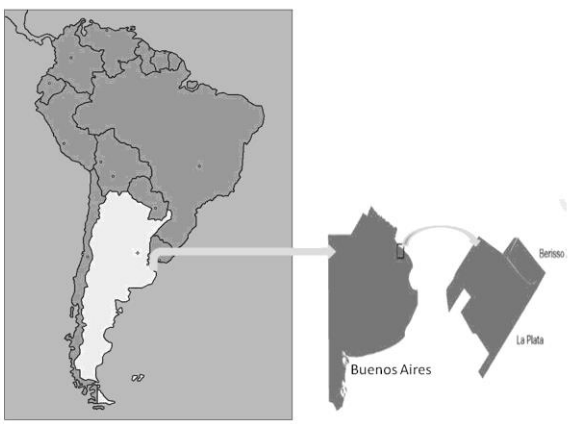

Figure 1. Area of study.

The children studied live in neighborhoods with typically low availability of health, sanitary and economical resources (Figure 2).

Figure 2. Neighborhood photo.

To develop the study, parents/guardians of the children were summoned through the school taking into account the WHO recommendations that suggest entering a community through schools (WHO 1987). Applying a structured and closed survey to each child, age and gender were registered and the presence of the following clinical manifestations was looked into: diarrhea, vomiting, pruritus ani, lack of energy, appetite loss, abdominal pain, and bruxism.

For the parasitological study, serial stool analysis and serial anal scraping were done. Instructions for sample-taking were imparted orally and written instructions were given to parents/guardians. For the serial stool analysis, a daily collection of a portion of stools in a container with preservative for 5 days was indicated. For the anal scraping, each parent/guardian had to dab a folded piece of gauze previously soaked in water around the margins of the child's anus every morning after waking up, for 5 days. The gauze pieces had to be placed in a second container with preservative. Stools were processed by the modified Telemann technique and the obtained pellets were observed through optical microscope (three smears per tube). The serial anal scraping samples were processed by cutting and homogenizing the gauze pieces with the same preservative in the container. After transferring the whole contents to a centrifuge tube, it was concentrated by centrifugation at 1000g for 5 minutes. Finally, three smears per tube were observed through the optical microscope.

Ethical Aspects: parents/guardians were informed orally and in detail about the study in group meetings held at the school. They were requested to give their consent in writing. Protocols developed by our group were approved by the UNLP School of Medical Sciences Ethics Committee. Personal information remained confidential and was obtained in accordance with the Declaration of Helsinki (1964), the Nuremberg Code (1947) and National Act #25,326. Approval of school and municipal authorities in the district was also obtained.

For the statistical analysis, total and specific parasitic infections, frequency of signs or symptoms, and registered variables (gender and age) were estimated. The possible associations were analyzed using the Chi-Square Test. In the associations that turned out to be significant ($p \leq 0.05$), the odds ratio (OR) and 95% confidence interval (CI) were estimated. Statistical analysis was done using the winepi software (http://www.winepi.net) and Epi info[tm] 3.5.4 software tools.

Results:

The study included 857 schoolchildren, 3 to 12 years old, grouped by sex and age. Of them, 454 (52.98%) were male, with 248 aged 3 to 5, 140 aged 6 to 8, and 66 aged 9 to 12. There were 403 females (47.02%), 229 of them were 3 to 5, 120 were 6 to 8, and 54 were 9 to 12 years old.

Of the 857 children, 548 (63.95%) had parasites. Among the males, the prevalence was 64.32% (292/454) and among the females, the prevalence was 63.53% (256/403). As to age ranges, with no sex differentiation, parasites were found in 66.67% (318/477) of the 3- to 5-year-old children, 64.23% (167/260) of the 6- to 8-year-olds, and 52.5% (63/120) of the 9- to 12-year-olds.

The presence of one species of intestinal parasite was detected in 253 (46.16%) children, two parasite species were found in 214 (39%), and 3 or more species in 81 (14.84%) children. The recorded species in parasitized children were Enterobius vermicularis in 276 cases (50.36%), Giardia intestinalis in 155 cases (28.28%), Blastocystis hominis in 339 children (61.86%), Ascaris lumbricoides in 15 (2.73%), and Trichuris trichiura in 5 (0.91%).

The global parasite frequencies in the schoolchildren by sex and age are shown in Table 1. As it can be noted, none of the age ranges showed significant differences in male and female ratios.

Table 1. Frequencies of parasitism by sex and age of schoolchildren.

	Males (n=454)	%	Females (n=403)	%	Difference in ratios ($p \le 0.05$)
Age (years)					
3-5	170/248	68.55	148/229	64.63	0.3642
6-8	92/140	65.72	75/120	62.50	0.5892
9-12	30/66	45.46	33/54	61.12	0.0874
Totals	292/454	64.32	256/403	63.53	0.8100

Taking into account the most frequent means of transmission, the parasite species were grouped in three categories, PI- E. Vermicularis (276/548= 50.36%); direct-contact transmission, PII- G. Intestinalis and/or B. Hominis (418/548= 76.27%); waterborne transmission of protozoa and PIII-A. Lumbricoides and/or T. Trichiura (19/548= 3.47%), geohelminths. Of the 292 males with parasites, 52.4% were infected with PI, 75% with PII, and 3.42% with PIII. Of the 256 females with parasites, 48.04% were infected with PI, 77.73% with PII, and 3.51% with PII. The frequencies of PI, PII and PIII, by sex and age range, are shown in Table 2. No significant differences were observed.

Table 2. Frequencies of PI, PII and PIII by sex and age of schoolchildren, applying statistical analysis of the difference in ratios (p<0.05)

	Males		**Females**		P
Age (years)	**PI**	%	**PI**	%	
3-5	90/170	52.94	70/148	47.29	0.3737
6-8	48/92	52.17	40/75	53.33	0.8976
9-12	15/30	50	13/33	39.39	0.4253
Total	153/292	52.4	123/256	48.04	0.3040
	PII	%	**PII**	%	
3-5	132/170	77.64	119/148	80.40	0.5354
6-8	67/92	72.82	55/75	73.33	0.9411
9-12	20/30	66.66	25/33	75.75	0.4251
Total	219/292	75%	199/256	76.73	0.6709
	PIII	%	**PIII**	%	
3-5	4/170	2.35	3/148	2.02	N/V
6-8	4/92	4.34	5/75	6.66	N/V
9-12	2/30	6.66	1/33	3.03	N/V
Total	10/292	3.42	9/256	3.51	0.9541

PI. E. Vermicularis, PII: G. Intestinalis and/or B. Hominis y PIII: A.lumbricoides and/or T trichiura. N/V: it does not validated

One or more signs or symptoms were recorded in 665 schoolchildren, and 65% of them had parasites, resulting in a significant difference (p<0.001) as compared to those with no parasites. Among the 192 children with no signs or symptoms, 61% had parasites, resulting also in a significant difference (p<0.0004) as compared to those with no parasites. However, the estimated risk between "parasitism" and "presence of signs or symptoms" produced an OR=1.1807 (0.8482<OR<1.6435), both variables turning out to be independent variables (Table 3).

Table 3. Presence of signs and symptoms (S/S) and parasitism.

		Parasites YES	Parasites NO	Total	
S/S	YES	431	234	665	OR: 1.18 (0.84-1.33
	NO	117	75	192	χ^2: 0.97 p>0.05
Total		548	309	857	

Taking into account the number of signs or symptoms per child, of 198 children with just one sign or symptom, 161 with two, and 306 with three or more, 65% in each group had parasites, resulting in significant differences (p<0.001) between children with and without parasites.

The frequencies of each sign or symptom in parasitized and non parasitized children are shown in Table 4, with no significant differences found.

Table 4. Signs and symptoms (S/S) in parasitized and non parasitized children

	S/S in children parasitized (n=548)	S/S in children non parasitized.(n=309)
S/S	%	%
Diarrhea	15.5	16.18
Pruritus ani	41.05	35.27
Lack of energy	18.79	17.47
Vomiting	9.67	8.41
Apetite loss	33.57	29.77
Abdominal pain	42.88	43.36
Bruxism	41.97	42.07
Presence of S/S	78.64	75.72

In 86 schoolchildren only E. Vermicularis was detected and in 175 only G.intestinalis and B. Hominis. One individual showed the association A. Lumbricoides and T. Trichiura only, therefore analyzing the signs and symptoms present was not considered. The frequencies of each sign or symptom in children only parasitized by E. Vermicularis and G.intestinalis and B. Hominis are shown in Table 5. Diarrhea and vomiting significantly predominated in the children infected with both protozoa.

Table 5. Frequency of S/S in schoolchildren only parasitized by E. Vermicularis (EV) and G. Intestinalis and B. Hominis (GB)

	EV (n=86) N (%)	GB (n=175) N (%)	P 0.0184
Diarrhea	7 (8.13)	34 (19.42)	0.0184
Pruritus ani	30 (34.88)	74 (42.28)	0.2511
Lack of energy	12 (13.95)	38 (21.71)	0.1343
Vomiting	2 (2.35%)	20 (11.42)	0.0132
Appetite loss	30 (34.88)	52 (29.71%)	0.3977
Abdominal pain	38 (44.18)	76 (43.43)	0.9074
Bruxism	33 (38.37%)	74 (42.28)	0.5460

Analyzing by sex, signs and symptoms, and parasitism, 347 males were found to show some sign or symptom, 64.85% (225/347) of whom had parasites, while 107 males showed no signs or symptoms, and 62.62% (67/107) of them had parasites. As regards female subjects, 318 showed some sign or symptom, 64.78% (206/318) of them had parasites and 85 of the girls had no signs or symptoms, 58.83% (50/85) had parasites. The estimated risk by sex, signs or symptoms produced an OR= 1.0324 with an IC 95%= 09609 and 1.1093; therefore, those variables are not associated.

Discussion

The research done shows over 60% parasite prevalence in children from vulnerable communities in the northeast province of Buenos Aires. We found protozoa G. Intestinalis and/or B.hominis were present in more than 75% of children with parasites, helminth E. Vermicularis in 50%, and geohelminths A. Lumbricoides and T. Trichiura in less than 4% of them. These findings are in agreement with other studies done in the region, where pathogenic protozoa are more prevalent than geohelminths (Indelman et al. 2011, Pezzani et a.l, 2012, Gamboa et al., 2014, Orden et al., 2014). These findings contrast those found by Zonta et al. (2010) in native communities in the north of Argentina, with high presence of geohelminths, matching a study carried on by Milano et al. (2007) in the Argentinean northeast and a study by Valverde et al. (2011) in an endemic area of the Brazilian Amazon, showing ground pollution is a determinant factor in those areas, though controlled in our region. The percentage of people with mono- and bi-parasitism was important, and the high frequencies of protozoa and E. Vermiculairs would show situations related to water and food pollution, as well as human behavior (Bartolini et al. 2017).

In this study, general parasite frequencies were not different from the sex variable in the population, but were reduced as the schoolchildren's age increased, a result that could be associated to immunity acquired over the years. Nevertheless, 9- to 12-year-old females were found to have more parasites than males in the same age group (61.12% versus 45.46%), a situation probably related to a greater exposition to protozoa infection sources, as observed in Table 2. An increase of the frequencies of E. Vermicularis and geohelminths was found in the older males, which could be related to their poorer personal hygiene and possible contact with contaminated soil. Studies carried out by Gamboa et al. (2014) and Zonta et al. (2010) in the region also agree on that the frequency of parasite infection was not associated with age or sex of hosts. In communities of coastal settlements in Brazil, Vieira da Silva et al. (2018) found no differences in the prevalence of intestinal parasitoses between males and females, but demonstrated a higher prevalence in children over 10 years old. For Valverde et al. (2011), younger children are less aware of the importance of personal hygiene, resulting in schoolchildren being considered a reservoir of intestinal parasites. Within this context, prevalence and intensity of infections are reduced by the time of adolescence, progressively declining in adults.

As already mentioned, pathogenic protozoa were detected in over 75% of the children with parasites. Among them, G. Intestinalis gains significance as a pathogen detrimental for the nutritional status of the infected people. Cross-sectional studies aimed at evaluating the relationship between intestinal parasitism and anthropometric parameters often found an association between giardiasis and low weight for age and low height for age (Matos et al., 2008 and Botero Garces et al., 2009). Besides, they showed that people infected with Giardia are younger, thus indicating the epidemiologic importance of giardiasis in children. B. Hominis is the other protozoon prevalent in this study. It is one of the parasites frequently found in human fecal samples in developed countries (Bartolini et al., 2017). This microorganism was initially considered a commensal, though following observations suggested its pathogenicity. Later, it was discovered that the host specificity and pathogenic potential of the different parasites correlated with the variations of sequence in the SSU-rrna. Based on these variations, members of the genus are classified in several subtypes (ST), which could possibly be referred to as species (Subhash J, 2014). At present, the most convincing but unsatisfactory explanation for the pathogenicity of Blastocystis is the correlation of ST with virulence. (Tan et al, 2010). Initial studies determining the pathogenicity of the subtype reported that ST3 had a strong correlation with the symptomatic disease (Tan et al, 2008). No genotypification was performed in our study, but almost 20% of the people who had G. Intestinalis and/or B. Hominis had diarrhea. Juarez and Rajal (2013) recount that Giardia has been reported throughout the Argentine territory in water samples, home effluents, soil and animal feces, and Blastocystis was found in water from rivers and streams, drinking water, and in ditches and soil, and this would be related to the high prevalences of these protozoa in the studied schoolchildren. It has been argued that massive periodic deworming, done without knowledge of the prevalence rates of enteric protozoa in different areas, could turn protozoan diseases in the predominant enteric parasitoses, and their control programs are a real challenge for public health policy-makers (Valverde et al. 2011).

E. Vermicularis is probably the most common helminth infecting human beings. High prevalence rates have been recorded in the northeast of Europe and the United States. Its wide-world distribution has no association with any socio-economical level, race or culture in particular. A frequency of 11.4% was informed in a study of 35 states in the USA (Burkhart and Burkhart, 2005). For these authors, the infection is common in children, with prevalent rates in this age group in specific regions reporting up to 61% in India, 50% in England, 39% in Thailand, 37% in Sweden and 29% in Denmark. In our country, numerous studies report its detection, mainly among children, ranging between 13.6% and 47% (Juarez and Rajal 2013, Rivero et al. 2017). These percentages were exceeded in our study, going over 50%. Even though this helminth is not considered to be associated with a severe disease, the morbidity associated to its presence, especially in children, is significant. Besides, removing this parasite in a household or institutional group entails important problems due to the interruption of treatment or reinfection (Burkhart and Burkhart, 2005). Juarez and Rajal (2013) report that E. Vermicularis is more prevalent in cold and temperate areas. The area where this study was carried out has moderate winters and hot summers.

As regards clinical manifestations, we observed that in our population there was no association between the presence of signs or symptoms and parasitism when all of the children were analyzed. Besides, no relationship between having parasites and sex and/or age was found. These results match studies carried out by Gamboa et al. (2009, 2014) and by Garbossa et al. (2013) in adult and children living in precarious settlements in our country. Nevertheless, when we analyze the number of signs or symptoms, children in the groups of one, two, or three of them showed association with intestinal parasitoses as compared to those with no parasites. The association between specific symptoms and parasite species has not been consistent in the diverse descriptions. While Limoncu et al. (2005) have not found association between symptoms and parasite infection, Miller et al. (2003) from Venezuela have associated abdominal pain with the presence of A. Lumbricoides and B. Hominis (Miller et al., 2003), in agreement with El Shazly et al. (2005) from Egypt. Lotero J (2010), from Argentina states that intense diarrhea appears with infections with G. Intestinalis and Uncinarias and a less intense diarrhea appears in the cases of infection by B.hominis; recurring abdominal pain appears with G. Intestinalis, A. Lumbricoides and S. Stercorlaris, and pruritus ani appears with E. Vermicularis. Mendez Bustelo et al. (2015) state that epidemiologic studies suggest B. Hominis is associated with a wide range of intestinal and extra-intestinal disorders. For these authors, signs and symptoms associated with this protozoon were: chronic or recurring abdominal pain, abdominal distention, appetite loss, diarrhea, vomiting and pruritus ani. Gamboa et al. (2009) claim that Blastocystis and Giardia are considered the most frequent protozoa in the world population. Both species appear in asymptomatic as well as symptomatic people. Infections due to these intestinal protozoa cannot be restricted to weather conditions, socio-economic groups or geographic areas. Probably, they are not related with sex either, but could be influenced by age, immunologic status and factors related to the hosts' hygiene. These authors found no association between digestive symptoms and these parasites. In our population, only vomiting and abdominal pain were related to the presence of G.intestinalis and B. Hominis.

As mentioned before, most pediatricians in our region performs antiparasitic treatment in children with no prior analysis requested. The control of intestinal parasitoses is based almost exclusively on medication and prophylactic measures of environmental sanitation. For Calentano A (2012), treatment with drugs may have the purpose not only of removing the infection in a patient individually (not an easy task), but also a population-wide decrease of the transmission, geohelminths in particular. But we must be alert about the unnecessary and/or repeated administration of drugs, as in massive prophylactic treatments or in individuals with chronic infections, since they favor the development of resistance. In drugs with "slow" pharmacokinetics, when the substance remains in contact with the parasite in sub lethal doses for extended periods, the parasite may survive and adapt to become resistant. Cross resistance should also be taken into account since its existence could be proposed between drugs with a similar mechanism of action. This hypothesis has an indirect support after having experimentally observed that strains of Trichomonas vaginalis were simultaneously resistant to metronidazole, nitazoxanide and furazolidone. While no resistance to antihelmintics has been observed in humans, it has indeed been observed at veterinary level, where mass treatments are more systematic.

The clinical course of the infection with intestinal parasites may vary from asymptomatic to severe complications, depending on the number, type or intensity of infections, as well as the immune status of the affected person. With respect to the symptoms and signs in the studied population, it can be pointed out that although there was a statistical association between signs and symptoms and the finding of one or more parasites, it was also found in a significant way, parasitized asymptomatic population. Keeping this situation in mind, we can suggest two behaviors should be taken into account, 1) perform the parasitological study for children who have some clinical manifestation mentioned and 2) also perform the parasitological study in asymptomatic children with risk factors associated with intestinal parasitoses, since previous studies have accounted for the existence of soil and water contamination, in humans and pets in our region (Cordoba et al. 2010, Juarez and Rajal 2013, Gamboa et al. 2014, Beretta et al. 2017).

Conclusions:

This study brings data to the controversy regarding the association between parasitoses and clinical conditions in children. The results reflect that in 3 to 12-years-old schoolchildren, no evidence of a relationship between the presence of signs or symptoms typically associated to the presence of intestinal parasites and the detection of parasites in a parasitological analysis can be found. Professionals are encouraged to request a lab study to detect the parasites circulating among the pediatric population so as to apply the appropriate anti-parasitic agent and also record the frequencies of parasites present in each region.

In addition, the results of this study reveal the importance of intestinal parasitoses with direct effect on an individual's health and the fact that they are a sign of precarious sanitary conditions in the community. Therefore, it is necessary to establish specific prevalences in each geographic area, so as to view the situation and undertake the relevant public health and healthcare actions.

Conflict of interest

The authors declare that there is no conflict of interest.

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