Seroepidemiological trend of strongyloidiasis in the Bolivian Chaco (1987–2013) in the absence of disease-specific control measures

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Abstract

OBJECTIVES   The prevalence of *Strongyloides stercoralis* infections is grossly underestimated because infections go mostly undetected, although they can persist for a lifetime due to the auto-infective cycle. In the Bolivian Chaco, the prevalence of soil-transmitted nematodes dropped dramatically in the past 25 years, but the mebendazole used for preventive chemotherapy has no effect on *S. stercoralis*. Meanwhile, the prevalence of intestinal protozoan infections remains unchanged. We compared *S. stercoralis* seroprevalence in rural communities of the Bolivian Chaco from 1987 to 2013.

METHODS   Sera collected during two previous serosurveys, conducted in the Chaco region in 1987 and 2013, were tested for *S. stercoralis* using a commercial assay (Bordier-ELISA, Bordier Affinity Products, Switzerland).

RESULTS   Overall, 355 sera were analysed, 122 from the 1987 survey and 233 from the 2013 survey. Seropositivity for *S. stercoralis* was significantly more prevalent in 1987 (19/122, 16% in 1987 *vs.* 15/233, 6% in 2013, $P = 0.006$), accounted for by a drop from 17% to 3% in people under 26 years of age. Multivariate analysis showed a significant association between seropositivity for *S. stercoralis* and age in the 2013 population (OR 1.03 for each one-year increase, 95%CI 1.00–1.05, $P = 0.04$), but none in 1987.

CONCLUSIONS   The significant reduction in *S. stercoralis* seroprevalence in Bolivian Chaco cannot be explained by preventive chemotherapy or improved social-sanitary conditions. As the drop is seen in younger generations, it is consistent with little transmission occurring. However, the risk of transmission still exists, as prevalence is persistently high in older individuals, who present a potential reservoir due to the lifelong nature of *S. stercoralis* infections.

keywords   *Strongyloides stercoralis*, strongyloidiasis, Bolivian Chaco, Bolivia, ivermectin, soil-transmitted helminths

Introduction

Strongyloides stercoralis is a soil-transmitted helminth widely diffused in tropical and subtropical regions of the world, including South-East Asia, Latin America, sub-Saharan Africa and parts of the south-eastern United States and also in some temperate areas at low endemicity [1]. An estimated 370 million individuals are infected with *S. stercoralis* worldwide [2]. Most infections remain undetected because they are often asymptomatic, or cause mild, unspecific signs and symptoms such as pruritus, abdominal pain, diarrhoea and respiratory disturbances;

eosinophilia may be present. Due to a peculiar auto-infective cycle, the infection perpetuates in the human body indefinitely, if not treated. In the immunocompromised host, the number of larvae can highly increase and spread to any tissue of the body, leading to hyperinfection syndrome and disseminated forms. These are severe life-threatening syndromes, which are often complicated by sepsis or meningitis due to intestinal bacteria carried into the bloodstream by the larvae. A number of severe syndromes have been reported due to concomitant use of immunosuppressant drugs, especially corticosteroids [3].

The global prevalence of the infection has long been underestimated, as data came from studies based on a diagnostic technique with low accuracy [2]. Indeed, direct stool examination and the Kato-Katz method, the most common stool-based parasitological techniques used in surveys on helminths prevalence, have unsatisfactory sensitivity due to the small numbers of *S. stercoralis* larvae in stool. More accurate parasitological techniques, such as specific concentration methods (e.g. the Baermann method), coproculture in agar plates and in-house molecular methods, are not routinely performed, being available only in few reference laboratories. Although serological techniques are more sensitive than stool-based methods, and also sufficiently specific above a given cut-off, there are few seroprevalence surveys for *S. stercoralis* [4].

A recent systematic review of the prevalence of strongyloidiasis in Latin America identified to Argentina, Ecuador, Venezuela, Peru and Brazil as high-prevalence countries (>20%), but reliable data were not available for most of the Latin American countries [5]. Moreover, studies were characterised by large heterogeneity in terms of population target and diagnostic methods used, with a small minority of serological-based surveys. Limited data on strongyloidiasis prevalence are available for Bolivia, all from surveys of soil-transmitted helminths (STH) prevalence, using stool-based methods [6–8]. Given the low sensitivity of these techniques, the reported prevalence of *S. stercoralis*, ranging from 0.9% to 1.8%, is likely to be grossly underestimated. The present study reports the *S. stercoralis* seroprevalence in rural communities of the Bolivian Chaco measured at two time points 26 years apart, in 1987 and 2013. In the period between the two surveys, no large-scale control measures potentially affecting strongyloidiasis were implemented.

Methods

Study area and population

The study populations consisted of people living in the Chaco region, south-eastern Bolivia between longitude

64°30′ and 58°50′ west of the Greenwich meridian and at latitude 17°58′ and 22°20′ south. The former survey was conducted in 1987 in Camiri (Cordillera Province, Department of Santa Cruz, 20°06′ south; 63°32′ west), a municipality of about 30 000 inhabitants situated in the foothills of the Andes, and Javillo, a very small community, isolated in the jungle north-east of Camiri, at an altitude of about 1500 m. The latter survey was conducted in 2013 in Bartolo (municipality of Monteagudo, Hernando Siles Province, Department of Chuquisaca: 16°30′ south; 59°88′ west) and Ivamirapinta (municipality of Gutierrez, Cordillera Province, Department of Santa Cruz: 19°45′ south; 63°30′ west). A map of the study area is provided in Figure 1.

Serologic assay

Sera, obtained from a sample of 5 ml venous blood, were stored at −20 °C in Bolivia, transported within 2–4 weeks to Italy in dry ice and stored at −70 °C until tested. In July 2016, all the samples were tested with a commercial enzyme-linked immunosorbent assay (ELISA) test (Bordier Affinity Products, Switzerland) using *Strongyloides ratti* L3 larvae as antigen, according to the instructions of the manufacturers. A cut-off of 1.1 was used to define positive tests; this threshold showed a good balance between sensitivity (around 90%) and specificity (around 97%) [9].

Statistical analysis

Data were entered into Microsoft Excel 2010 software (Microsoft, Redmond, WA, USA). Statistical analysis of the data was performed with STATA 11.0 (StataCorp, College Statio, TX, USA). Frequencies and percentages with 95% confidence intervals (CI) for categorical variables, medians and interquartile ranges (IQR) for continuous variables were calculated. Two age categories were created: <26 and ≥26 years of age (the time difference between the two surveys, so as to have a group of 2013 subjects who were not yet born when the first survey was conducted).

Differences in sex and age were estimated by proportion test and Mann–Whitney test, respectively. The Cochran-Mantel-Haenszel stratified test was used to compare seroprevalence. To account for differences in the sample size between the two surveys, frequency ratios were calculated overall and by age category and the seropositivity rates recalculated accounting for these differences; a sensitivity analysis was conducted on this recalculated sample.

Multivariate analysis was performed by logistic regression to establish the association of positive ELISA test

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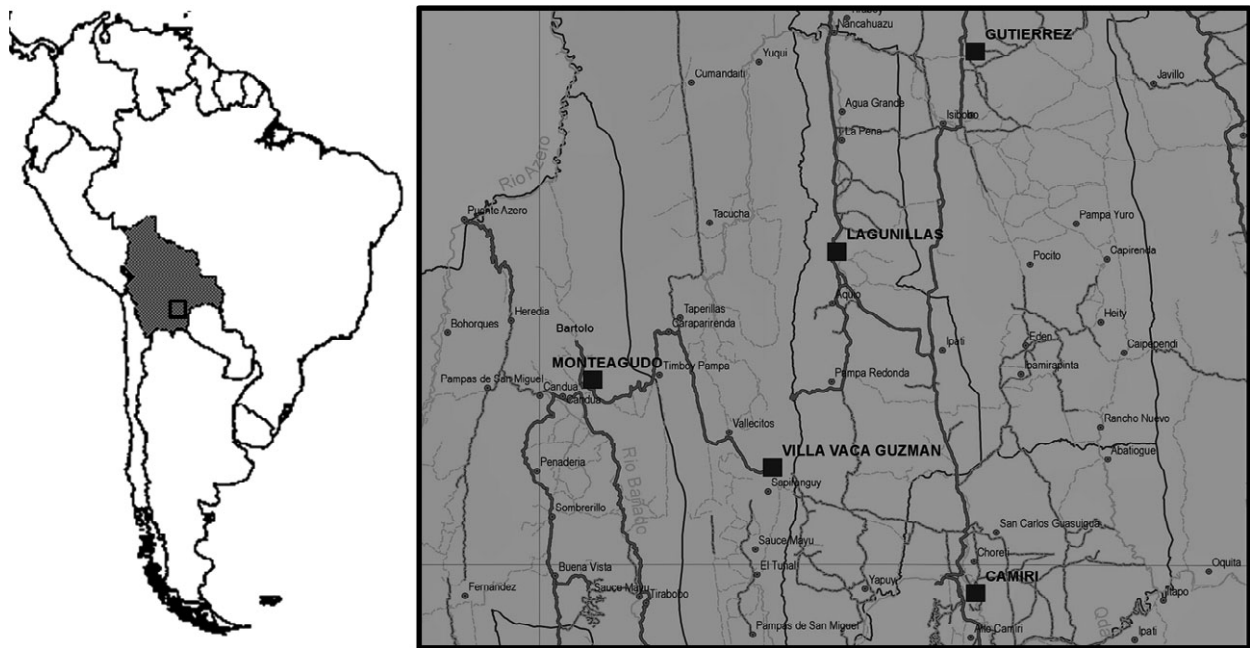
Figure 1 Study area map, within the Bolivian Chaco Region. In 1987, the study was conducted in the municipality of Camiri (Cordillera Province, Department of Santa Cruz) and the community of Javillo (municipality of Gutierrez, Cordillera Province, Department of Santa Cruz) and in 2013 in the communities of Bartolo (municipality of Monteagudo, Hernando Siles Province, Department of Chuquisaca) and Ivamirapinta (municipality of Gutierrez, Cordillera Province, Department of Santa Cruz).

with sex and age. Results were considered significant when the P-value ≤ 0.05.

Ethics

Sera used in the present study were collected during two previous serological surveys focused on other infectious diseases in 1987 and 2013. Sera were completely anonymised. The previous and present studies were programmed and conducted in agreement with the Ministry of Health of the Plurinational State of Bolivia (Convenio Ministerio de Salud y Deportes, Estado Plurinacional de Bolivia/Cátedra de Enfermedades Infecciosas, Universidad de Florencia, Italia) and with the support of the Guaraní political organization (Asamblea del Pueblo Guaraní). Ethical approval for the study was obtained from both institutions.

Results

We analysed a total of 355 sera from subjects aged 1–70 years, of whom 122 came from the 1987 survey in Camiri and Javillo and 233 from the 2013 survey in Bartolo and Ivamirapinta. Demographic characteristics are presented and compared both within and between the

two surveys in Table 1. Within each survey (Camiri *vs.* Javillo, Ivamirapinta *vs.* Bartolo), the study populations were homogenous, except for a higher female frequency in Camiri than in Javillo in 1987.

Significant differences between the two populations were as follows: (i) a higher female frequency among 1987 participants (68.0% *vs.* 50.2% in 2013); (ii) the 2013 population from Bartolo and Ivamirapinta was significantly older than that enrolled in 1987 (mean age 29.3 years *vs.* 17.8).

The *S. stercoralis* seroprevalence was statistically significantly higher in 1987 than in 2013 (19/122, 16% in 1987 *vs.* 15/233, 6% in 2013, Cochran-Mantel-Haenszel test age-stratified $P = 0.006$ on uncorrected data; 0.028 on sensitivity analysis). The overall and age-stratified prevalences are presented in Figure 2. By multivariate analysis, we found a significant association between seropositivity for *S. stercoralis* and age in the 2013 population (OR 1.03 for each one-year increase, 95% CI 1.00–1.05, $P = 0.04$), but none in 1987.

Discussion

We report a significant drop in *S. stercoralis* infections over a 26-year period in rural Bolivia (from 16% to 6%)

Table 1 Demographic data of the human population surveyed in 1987 and 2013 in the Bolivian Chaco

	1987 Survey			2013 Survey		
	Camiri	Javillo	*P*-value	Ivamirapinta	Bartolo	*P*-value
Sex female, N (%, 95% CI)	48 (85.7%, 76.5–95.0)	35 (53.0%, 41.0–65.1)	<0.01	72 (53.3%, 44.9–61.7)	45 (45.9%, 36.1–55.8)	0.26
	83 (68.0%, 59.8–76.3)			117 (50.2%, 43.8–56.6)		<0.01
Median age (IQR)	16 (14–17)	15.5 (10–30)	0.95	23 (12–44)	27 (14–43)	0.40
	16 (11–18)			26 (12–43)		<0.01
Mean age (95% CI)	15.6 (13.9–17.3)	19.7 (16.1–23.1)		28.7 (25.3–32.0)	30.2 (26.4–33.9)	
	17.8 (15.8–19.8)			29.3 (26.8–31.8)		

N, number; IQR, interquartile range; CI, confidence intervals.

in the absence of disease-specific interventions. To the best of our knowledge, this is the first study of this kind. Few serological surveys for *S. stercoralis* have been conducted in Latin America, with disparate results. In a rural community of the Peruvian Amazon region, a study published in 2006 reported a seroprevalence of 72% among 609 sera by ELISA, whereas *S. stercoralis* was identified in only 7.8% of stool samples evaluated by three methods (direct examination, Baermann method and simple sedimentation) [10]. In a Brazilian review, which considered data from 1990 to 2009, the mean seroprevalence for *S. stercoralis* from epidemiological studies in the general population was 21.7% and 29.2%, using immunofluorescence antibody test (IFAT) and ELISA, respectively [11]. In Chile, 0.25% of apparently healthy individuals were infected with *S. stercoralis* by ELISA; infections were found in few areas of the country and in high-risk groups, such as mentally ill hospitalised patients (12.1% of 675 patients from two psychiatric hospitals) [12]. Indirect information about *S. stercoralis* seroprevalence in the Bolivian population can be inferred from few community studies on Bolivian immigrants in European countries. A

serological survey carried out in Bergamo, Italy, between 2009 and 2011, involving 739 Bolivian migrants (mean age 34.7 years) tested with an in-house IFAT, reported a global prevalence of 9.7% [13]. In 2015, among 486 Bolivian migrants in Geneva, Switzerland (mean age 36.5 years), the prevalence was 13.4%, using the Bordier-ELISA test [14]. In that study, which included also migrants from others Latin American countries (such as Brazil, Ecuador, Colombia and Peru), Bolivian nationality and provenance from a rural area were associated with a higher risk to be infected with *S. stercoralis*.

Importantly, the reduction in *S. stercoralis* seroprevalence was accounted for by a drop from 17% to 3% in the under 26, while the prevalence in older subjects remained stable (9% and 10%, respectively). This latter group is likely to be the same generation who were infected 26 years earlier as children and young adults, while the low prevalence in the under 26 in 2013 reflects the cumulative risk of infections in the last quarter of a century.

The association found here in 2013 between age and risk of *S. stercoralis* infection is known, but was not present in 1987, possibly because it was comparatively younger; there was no difference between the two surveys in male representation, another risk factor for *S. stercoralis* infection [10, 14]. Of note, in 2013 we also found 3/8 subjects over 70 to be positive for *S. stercoralis* (data not shown). Considering that *S. stercoralis* infection, when acquired, can potentially last for life because of its auto-infective cycle, older adults will likely present a reservoir in the community; however, they do not seem to be an efficient source of transmission, as infections among the younger generations were now far less common.

The overall reduction of *S. stercoralis* seroprevalence is consistent with the dramatic decrease of STHs evidenced by parasitological surveys conducted in the same areas of the Bolivian Chaco, during this lapse of time: in 1987, the prevalence of hookworm and *T. trichiura* in a rural
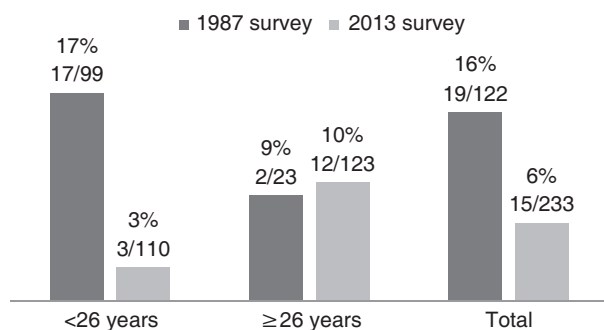
Figure 2 Seroprevalence for *Strongyloides stercoralis* antibodies in the human population surveyed in 1987 and 2013 in the Bolivian Chaco, both overall and age-stratified. Cochran-Mantel-Haenszel *P* value 0.006.

community near Camiri was 64.7% and 25.6%, respectively. In 2013, no STH infections were found among 111 people living in Ivamirapinta, except 1 case of *S. stercoralis* infection, while only 3 *A. lumbricoides* infections (3%) and 2 hookworm infections (2%) were documented among 112 inhabitants of Bartolo [6, 15]. This result may be largely ascribed to the implementation in 1986 of a preventive chemotherapy programme that delivers single-dose mebendazole to two- to nine-year-old children approximately every 6 months. However, given that mebendazole is not likely to affect *S. stercoralis*, other factors must have contributed to the trend. Theoretically, a lower *S. stercoralis* seroprevalence could be attributed to the STH decrease because of serologic cross-reactivity, but this phenomenon was shown to be of limited importance [9, 10]. Another reason could be improved sanitary conditions; however, the persistent high prevalence of intestinal infections caused by water-borne protozoa such as *Entamoeba coli, Giardia intestinalis* and *Blastocystis hominis* documented in the same parasitological studies suggests that general hygienic and sanitary conditions continue to be inadequate in these areas [6, 7, 15]. Other factors, such as climatic change and fewer risky behaviours, e.g. walking barefoot or indiscriminate defecation, are worthy to be further investigated to better understand the trend of *S. stercoralis* infection over the time. Moreover, cross-reactivity of antifilarial antibodies should be considered in areas of endemicity. In 1997, a cross-sectional survey in the Bolivian Chaco showed that 26% of the rural population near Camiri harboured *Mansonella ozzardi* microfilariae [16]; meanwhile, no specific interventions were implemented to reduce its prevalence.

The main limitation of the study is that the two surveys took place in different communities. However, given that the Bolivian Chaco is considered a homogeneous ecological zone and that rural populations share the same hygienic and sanitary conditions, no substantial differences in *S. stercoralis* distribution is expected throughout the region.

In conclusion, despite the observed reduction over the last decades, *S. stercoralis* continues to be endemic within the rural areas of the Bolivian Chaco, with a persistently transmission potential, thus requiring local health-care providers to be alerted about the risk and to implement all the appropriate preventive interventions against *S. stercoralis*.

WHO and its partners consider strongyloidiasis one of the STHs that are particularly neglected. There are theoretical indications and practical experiences showing that large-scale administration of ivermectin can drastically reduce the prevalence and the morbidity due to strongyloidiasis [17–19]. To implement this control strategy at global level, we need more detailed knowledge of the distribution patterns of the infection and sufficient supplies of good-quality and low-cost ivermectin, which is presently missing. WHO, to find a solution for the limited drug availability, is now inviting the submission of dossiers by generic manufacturers for the prequalification of their product [20].

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