

# Persistence of *Trypanosoma cruzi* vector-borne transmission among school-age children in the Bolivian Chaco documented by 24-month longitudinal serosurveillance

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**Background:** Chagas disease represents a major public health concern in several Latin American countries, including Bolivia.

**Methods:** We present a longitudinal serosurvey for *Trypanosoma cruzi* antibodies among a cohort of 120 school-age children from rural communities in the Bolivian Chaco at three time points between 2017 and 2019. Serum samples extracted from dry blood spots collected on filter paper were tested for *T. cruzi* antibodies by enzyme-linked immunosorbent assay and rapid diagnostic test.

**Results:** *T. cruzi* antibodies were detected in 7/120 (5.8%), 8/120 (6.7%) and 11/120 (9.2%) samples in 2017, 2018 and 2019, respectively. An average incidence of 1.76 per 100 person-years was observed.

**Conclusions:** Our findings support the persistence of vector-borne *T. cruzi* transmission in this area, highlighting the need for strengthening multidisciplinary efforts against Chagas disease.

**Keywords:** Bolivia, Chagas disease, prevalence, serologic tests, serology

## Introduction

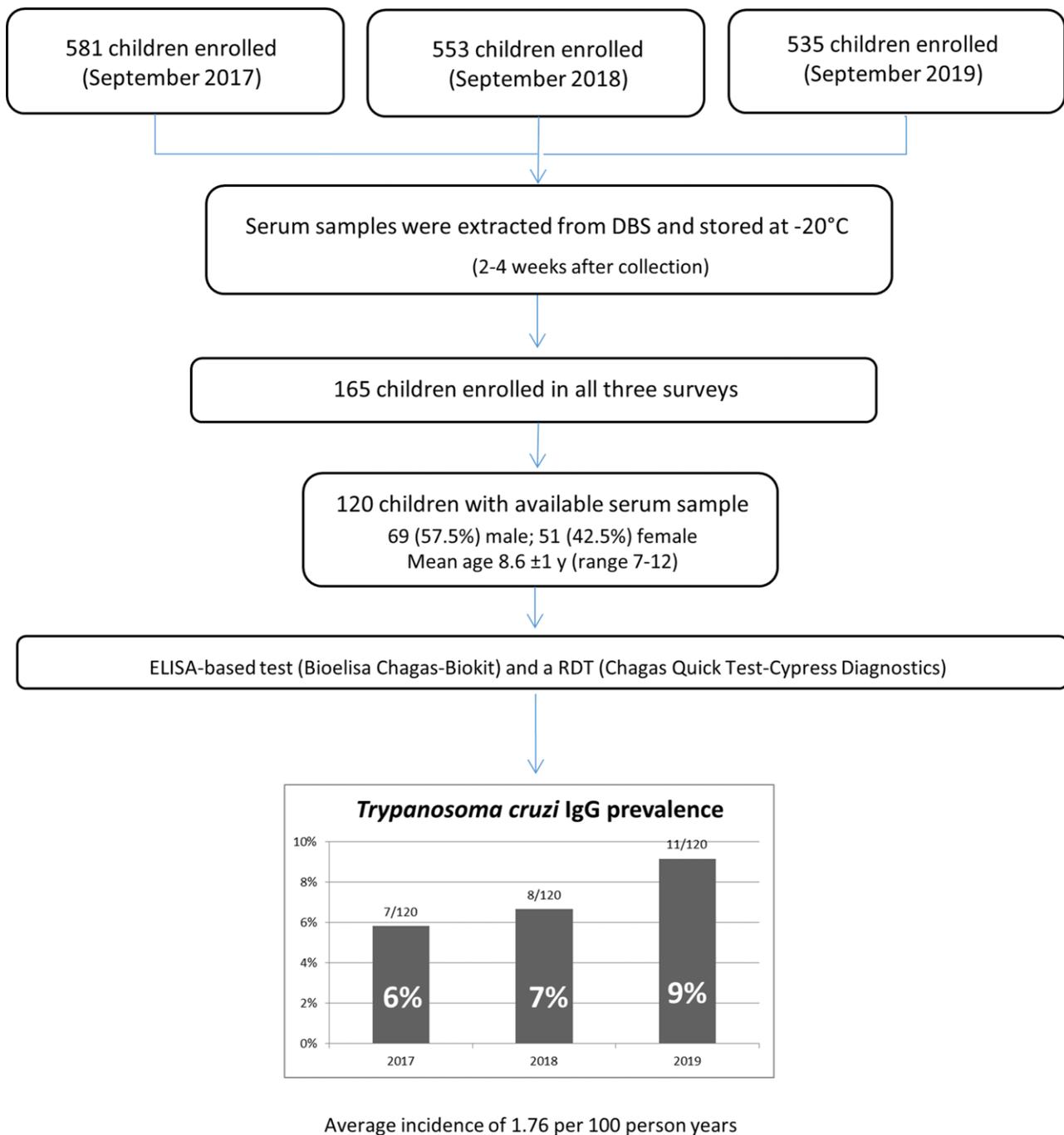
Chagas disease (CD) is caused by infection with the protozoan parasite *Trypanosoma cruzi*, naturally transmitted by triatomine bugs. Although good progress in vector control has been achieved in the recent decades, CD still represents a major public health concern in several Latin American countries. Bolivia is the country with the highest prevalence of CD worldwide (6.1% of the entire population), with an endemic area covering 60% of the country.<sup>1</sup> Following vector control campaigns promoted by the National Program for Chagas Disease Control, a considerable reduction in the proportion of infested houses was achieved in recent decades at the national level, from an estimated 47.7% infestation rate (IR) in 1999 to 1.9% in 2016. However, in the Bolivian Chaco, in southeast Bolivia, control interventions against *Triatoma infestans* were less successful and the house IR is still >3% in that area.<sup>1</sup> In this study we present a longitudinal serosurvey of *T.*

*cruzi* antibodies among a cohort of 120 school-age children living in rural communities in the Bolivian Chaco at three time points 12 months apart in the period 2017–2019.

## Methods

The Bolivian Chaco is a semi-arid and sparsely populated region located between longitude 64°30' and 58°50' W of the Greenwich meridian and latitude 17°58' and 22°20' S. The majority of the inhabitants live in rural communities, where houses are poor dwellings with walls constructed of wattle-and-daub or unplastered adobe bricks and thatched roofs.

Between September 2017 and September 2019, three cross-sectional parasitological surveys were carried out within a program of soil-transmitted helminths monitoring, 12 months apart, in nine rural communities of the Bolivian Chaco, recruiting



**Figure 1.** Study cohort selection from the populations of school-age children living in rural communities of the Bolivian Chaco, surveyed in September 2017, September 2018 and September 2019 (N=120) and longitudinal evaluation of the prevalence for *T. cruzi* antibodies. RDT: rapid diagnostic test.

581, 553 and 535 school-age children, respectively. In each community, a minimum of 50 children was enrolled among those attending the third-year class, usually 8–9 years old. At enrolment, a blood sample was collected from all participants through a finger prick and stored as dried blood spots (DBSs) on filter paper. Approximately 25  $\mu$ L of blood were dropped on a circle 1.2-cm in diameter on filter paper. Serum samples were extracted from DBSs following protocols previ-

ously described<sup>2</sup> and stored at  $-20^{\circ}\text{C}$  thereafter. All samples from the three surveys were then tested at the same time in 2020. *T. cruzi* infection status was established based on the consensus results of two serological assays for immunoglobulin G (IgG) anti-*T. cruzi*, an enzyme-linked immunosorbent assay (ELISA)-based test (Bioelisa Chagas, Biokit SA, Barcelona, Spain) and a rapid diagnostic test (Chagas Quick Test, Cypress Diagnostics, Hulshout, Belgium). Discrepant samples were tested

with a third ELISA (Novalisa Chagas *Trypanosoma cruzi* IgG, NovaTec Immundiagnostica, Dietzenbach, Germany).

## Results

Careful review of the database revealed 165 children were enrolled in all the studies, of whom 120 had an available blood sample collected on a dry blood spot at the three time points. Among this longitudinal cohort, 69 (57.5%) were male and 51 (42.5%) were female (male:female ratio 1:0.7). The mean age at enrolment was 8.6 y (standard deviation [SD] 1; range 7–12). *T. cruzi* infection was detected in 7/120 (5.8%) at the first assessment (2017). After 12 and 24 months, one and three seroconversions, defined as a positive result in a previously negative child, were observed, generating a prevalence of 8/120 (6.7%) and 11/120 (9.2%), respectively (Figure 1). Overall, an average incidence of new *T. cruzi* infection of 1.76 per 100 person-years was observed.

A comprehensive review of studies investigating *T. cruzi* seroprevalence in the general population of the Bolivian Chaco showed a partial reduction in CD prevalence over the past 40 y, with an age-dependent trend.<sup>3</sup> In 2011–2012, Samuels et al.<sup>4</sup> found an overall 20% *T. cruzi* seroprevalence among children living in a rural community of the Bolivian Chaco, with a steep increase in prevalence between age 2 and 15 y, which suggested that vector-borne transmission was responsible for most infections in this age group. Based on catalytic models, the authors estimated a force of infection (FOI), defined as the proportion of the susceptible population that becomes infected over a given period of time, >2.5% per year. In 2017, Hopkins et al.<sup>5</sup> found a seroprevalence of 22% among children (11 y [SD 4.4], range 4–21) living in a village near Camiri in the Bolivian Chaco. Again, the prevalence increased with age and the annual incidence estimated from the FOI was 2%.

In our population, 6% of school-age children were already seropositive for *T. cruzi* at the first assessment, being difficult to distinguish whether they had infection acquired via a vectoral or other route, such as congenital transmission, which continues to be concerning in the Bolivian Chaco. At the national level, the infection rate among pregnant women was 16.4%, and maternal transmission in Bolivia was estimated at 1.3–2.3% in 2016.<sup>1</sup> In the Bolivian Chaco, the situation is more challenging, especially in rural areas, where maternal seroprevalence stands at 60–75% and the congenital transmission rate is 4–6%.<sup>1,3</sup>

Our findings should be interpreted with some caution, due to the uncertain diagnostic performance of these serological tests on dry blood spot samples. Nevertheless, we can assume that four new infections with *T. cruzi* likely due to vector-borne transmission were identified through our longitudinal assessment. The oral route is less probable, although not negligible. This finding supports the failure to control domestic *T. infestans* in the Chaco region. Campaigns for indoor residual spraying of insecticides were sporadic and/or intermittent in this area.<sup>3,4</sup> Low indoor residual spraying effectiveness has been attributed to vulnerable house construction, suboptimal spray practices, short insecticide residuality and reduced vector susceptibility to pyrethroid insecticides. Moreover, the presence of multiple animal reservoirs, including the canine population, was confirmed as a further challenge for disease control and prevention strategies in

this area.<sup>6</sup> Sustained multidisciplinary efforts that include vector control, clinical care and community-based approaches tailored to the local cultural context are urgently needed.<sup>7</sup>

**Authors' contributions:** MSp, FM, AB and SG were responsible for conceptualization. FM, ALV and SG were responsible for the methodology. MSp, FM, VP, MSt, AB and SG were responsible for the investigation. HG, ALV, MSt, MR, RV, AB and SG were responsible for resources. MS and SG were responsible for data curation and original draft preparation. All authors were responsible for reviewing and editing the manuscript. All authors have read and agreed to the published version of the manuscript.

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**Competing interests:** None declared.

**Ethical approval:** The study was approved by a local ethics committee (Colegio Médico de Santa Cruz, TDEM CITE No. 005/2016) and written informed consent was obtained from a parent or legal guardian.

**Data availability:** The data presented in this study are available upon request from the corresponding author.

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