Seroprevalence and sociocultural conditionants of Chagas disease in school aged children of marginal zones of Asunción

Seroprevalencia y condicionantes socio-culturales de la enfermedad de Chagas en niños escolares de zonas marginales de Asunción

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Abstract Chagas disease is becoming a public health problem in Latin America due to the wide distribution, the high prevalence, the magnitude of the damage caused and the difficulties to control it. In Paraguay, the disease is mainly distributed in the departments of Paraguari, Cordillera and Central. Prevalence in marginal zones, where migrations from rural populations and endemic areas make possible the urbanization of the disease, has no been studied yet. This is a descriptive study with a cross-sectional sampling and a probabilistic system recruitment carried out in school aged children from marginal zones of Asuncion to determine the prevalence of Chagas' disease. Serological methods, parasite isolation and questionnaires were used to achieve the goals. Nine hundred and fifty three children were studied to determine the prevalence of Chagas' disease in marginal zones which was 1.4%.

Key-words: Trypanosoma cruzi. School children. Risk factors. Chagas' disease.

Resumen La enfermedad de Chagas es un problema de salud pública en América Latina debido a su amplia distribución, elevada prevalencia, por la magnitud del daño que produce y las dificultades para lograr su control. En Paraguay la enfermedad se encuentra distribuída principalmente en los departamentos de Paraguarí, Cordillera y Central. La prevalencia en zonas marginales de Asunción, donde las migraciones de poblaciones rurales y de áreas endémicas hacen posible la urbanización de la enfermedad, no ha sido estudiada aún. Este es un estudio descriptivo con muestreo de corte transversal y reclutamiento por sistemas probabilísticos, llevado a cabo en niños en edad escolar de zonas marginales de Asunción para determinar la prevalencia de la enfermedad de Chagas en este grupo. Métodos serológicos, aislamiento de parásitos y cuestionarios fueron utilizados para alcanzar los objetivos. Novecientos cincuenta y tres niños fueron estudiados para determinar la prevalencia de la enfermedad de Chagas en zonas marginales dando como resultado el 1,4%.

Palabras-claves: Trypanosoma cruzi. Niños escolares. Factores de riesgo. Enfermedad de Chagas.

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Chagas disease is endemic in most Latinoamerican countries¹⁷ and it is considered the third most important disease after malaria and schistosomiasis in Latin America¹⁸.

In Paraguay, Chagas' disease is a public health problem (Ministerio de Salud Pública y Bienestar Social, SENEPA, Paraguay. Plan Nacional para el Control de la enfermedad de Chagas. Documento mimeografiado, 1992) and transmission by vector occurred in wide zones while blood bank control is not mandatory¹⁰.

The most important migratory current orientated to Asunción, capital of Paraguay, is the originated in the meager landed property central zone. About 40% of the immigrants to Asunción, came from the Departments of Paraguari, Cordillera and Central⁷ which are highly endemic for Chagas' disease (Rosner J: Final Report: A new approach for a seroepidemiological clinical and educational program on Chagas' disease in Paraguay. IDRC Grant No. PY-089 unpublish. Document, 1986).

The vector can be passively transported with the migrant domestic household goods and the migrant himself could become an insect and parasite disperser outside of the endemic zone also contributing to the increment of transfusion transmission, the main via of disease transmission in urban zones. Eventually, infected migrant women, even though far from the vector source, could infect their children by transplacentary via.

The early detection of the chagasic infection is an useful tool to prevent the disease chronicity in children in whom recent infection are assumed. An efficient treatment has a great individual impact by eliminating the parasite presence and a community impact by reducing the parasite offer and thus difficulting the transmission chain¹³. However, there is little information about the prevalence of Chagas' disease in children living in marginal zones of urban areas and the risk factors that could be associated to the transmission in these zones where the presence of the vector is infrequent.

The specific objectives of this study were to estimate the prevalence of Chagas' disease in school aged children from marginal zones of Asunción by determining the presence of anti- \mathcal{T} . Cruzi antibodies and the parasite in serologically positive children to confirm the infection criteria, and to analyze biological and socio-cultural conditionants related to the transmission of the disease in the study population.

MATERIAL AND METHODS

Geographic location and study population. The study was carried out in 18 marginal neighborhoods of Asunción which is a non endemic area for Chagas' disease. One thousand and twenty nine children under 13 years of age and their mothers or tutors were surveyed in schools aleatory and systematically selected from a list provided by the Department of Elementary Schools of the Ministry of Education.

Aleatory sampling of schools and sampling of the school population. The schools located in the peripheric area of Asunción were located with the support of the staff of the Municipality of Asunción and they were then subjected to a random and systematic sampling selecting 18 schools.

Stadigraph Z was used for the calculation of the sample size for descriptive studies and dichotomous variables⁸. Using a 95% confidence level, p < 0.05 with 2 tails and an expected proportion of 5%, 456 subjects were the initial minimum sample size. During the pilot study, a low prevalence was detected causing the duplication of the sample size (approximately 1,000 children).

Design. A descriptive study was carried out to determine the prevalence of infection in the sample. In this study, the cases were defined as those individuals that had 1:20 or higher antibody titers determined by Enzymelinked Inmunoabsorvent (ELISA) and indirect immunofluorescence (IIF) test.

1. Seroprevalence: To determine the prevalence of anti-T. cruzi antibodies 1,029 samples were analyzed. Blood samples were obtained by finger puncture with a sterile lancet and collected on filter paper filling circles of 6mm diameter (equivalent to 5µl of sample). Blood spots were eluted with 250µl of incubation buffer for 24h. Samples were then processed by an ELISA test (Chagas Test ELISA, IICS, Asunción) to detect anti-T. cruzi antibodies9. Positive results were confirmed by another ELISA test as well as 10% of the negative samples,in order to confirm the result venous blood samples were obtained from seropositive children and processed by ELISA, IIF and immunoblotting (IB)15. Anti-T. cruzi IgM antibodies were also analyzed by IIF, ELISA and immunoblotting in order to establish if seropositive children were in the acute phase of the disease. Parasitologic tests were carried

out (direct parasitemia and hemoculture) to determine the presence of the parasite in blood. Seropositive children were referred to a pediatrician for a clinical evaluation. The mothers and other close relatives of the seropositive children were also studied by the same methods.

Study of biologic and social factors: A questionnaire was used to interview the father, mother or child tutor in order to evaluate the following variables:

Mother and child migration: Mother and child's birth-place, previous residence locations, trips to the countryside. Also, it was considered if birth-place or previous residence place was located in an endemic area for Chagas' disease or not.

Dwelling: Zone or neighborhood where child lived, dwelling condition (good or bad) determined by the materials used in the house construction.

Disease knowledge: The mother or tutor, attitude towards the disease was evaluated in by the use of insecticides.

Occupation of the family head (father, mother or tutor), monthly income of the family head, education level of the family head.

Family environment: Number of people living in the house, number of children of the mother, the person with whom the child was living at the time of the study.

Biological variables: Mother and child serology, reception of blood transfusion by the mother or the child, vector bites suffered by the mother or child, presence of triatomines or vestiges in the house.

3. Data analysis: The data of this study were divided into groups according to the frequency and proportion (or percentage) of each category (seropositive or seronegative to T. cruzi infection). The relation between the predictor variables and infection was individually examined by the chi-square test and relative risks with confidential limits. A value of p < 0.05 was considered as statistically significant. The analysis was carried out by using the Statistical Package for Social Sciences (SPSS-PC, 1987) and EPI INFO 5.01 (OMS, 1990) softwares.</p>

RESULTS

One thousand and twenty nine children were serologically studied for *T. cruzi* obtaining a prevalence of 1.3%. However, we will refer to 953 children for the rest the variables analyzed because 76 questionnaires were eliminated due to incomplete data. Of the 953 children, 1.4% (13/953) presented anti-*T. cruzi* IgG antibodies

by three serologic methods (Table 1). Fifteen percent (2/13) of the seropositives presented anti-*T. cruzi* IgM antibodies by IIF and IB. Parasitological methods were negative and clinical manifestations compatible with Chagas' disease were not detected. Nevertheless, both children were treated.

Table 1 - Seroprevalence of Trypanosoma cruzi infection in school children of Asunción-Paraguay by sex and agea.

Age(years)	Male		Female	Totalb	
	pos/nc	%	pos/n %	pos/n	%
4-6	0/61	0 d	0/68 0	0/129	0
7-9	5/195	1.3	2/204 0.5	7/399	1.8
10-13	2/186	0.5	4/226 1	6/142	1.5
Total	7/442	1.6e	6/498 1.2e	13/940	1.4

a. Based on the Indirect Inmmunofluorescence, ELISA and Immunoblotting; **b**. x² for trend by sex = 0.684; p = 0.41; **c**. pos/n = number of positive samples/total samples; **d**. Figures in parenthesis are percentages; **e**. x² = 0.24; p = 0.62.

Seropositivity of children did not present any significant association with age, sex and school level (Table 1). For each group, boys presented a high prevalence in the group of 7-9 years old while girls presented it in the group of 10-13 (Table 1). However, there were significant differences concerning child seropositivity when it was

compared with the following variables: child's birth-place in endemic areas (p = 0.02, RR = 4.57), their residence (p = 0.032) and temporary mobility to endemic areas (p = 0.03, RR = 3.43) (Table 2). None of the seropositive children had received blood transfusions and neither triatomines nor vestiges were found in the dwellings.

Also, there was a significant association between the child seropositivity and the mother's or tutor's birth-place in endemic area (p = 0.035), knowledge of the vector of the interviewed people (mother, father or tutor) (p = 0.04), statement of parents or tutors that they had been bitten by the vector (p = 0.018) as well as with crowding in dwellings (p = 0.04). It is worthwhile to point out that there were evidences of a marginal association when the seropositivity of the mothers (of seropositive children) and their birth in endemic areas were analyzed (Table 2).

Concerning migration, data about the mothers and children's birth-places and previous residence location were collected. Then, these locations were classified as endemic or not and it was found that 82% of the mothers of the seropositive children was born in endemic areas

of the country while only 18% was born in Asunción (p < 0.05) (Table 2). A higher percentage of children seropositives (61.5%) was born in Asunción. Concerning migration of seropositive family members living in the same dwelling, 67% was born in endemic areas and 33% was born in Asunción (data not shown).

It is important to mention here that the main migration reason in the sample was the possibility of getting a job (72.5% of the interviewed population). It should be noticed that 90% of the mothers of the seropositive children was also seropositive.

Only 18% of the seropositive mothers had received a blood transfusion after having the studied children. About 21% (9/42) of the cases had close relatives with *T. cruzi* infection. The

Table 2 - Prevalence of variables associated to the presence of anti-T. cruzi antibodies and analysis of the statistic significance in the children of marginal zonas of Asunción, 1994.

	Characteristic	Serology of chidren and interviewed people				Statistics
		Positive n = 13 (1.4%)		Negative n = 940 (98.6%)		relative risk
Variables						(confidential limits
		nº	%	nº	%	at 95%)
Child's birth-place	Endemic zone	5	4.2	113	95.8	RR = 4.4(1.5-13.3)
	Non-endemic zone	8	0.96	827	99.04	p = 0.02
Child always lived in the same place	Yes	8	1.02	777	98.98	RR = 0.28(0.1-0.8)
	No	5	3.65	132	96.35	p = 0.03
Trips of the child to the countryside	Yes	6	3.1	188	96.9	RR = 3.4(1.1-9.9)
	No	7	0.92	752	99.08	p = 0.03
Interviewed person's birth-place	Endemic zone	11	2.1	520	97.9	RR = 4.32(0.9-19.3)
	Non-endemic	2	0.5	418	99.5	p = 0.06*
Knowledge of the vector by the	Yes	11	2.1	523	97.9	RR = 4.32(0.9-19.3
interviewed person	No	2	0.48	417	99.52	p = 0.07*
Mother or tutor referred having been	Yes	4	9.8	37	90.2	RR = 4.9(1.5-16.3)
bitten by the vector	No	7	1.96	350	98.04	p = 0.02
Crowding of the dwelling	Yes	10	2.4	408	97.6	RR = 4.1(1.1-14.9)
	No	3	0.6	515	99.4	p = 0.04

^{*} not significant.

four seropositive children's relatives were cases' brothers being their mothers also seropositive. None of the case children had received any blood transfusions.

Table 3 shows the odds ratios for antibodies against $\it T.~cruzi$ and controlling by sex in the

group of 10-13 years old with those with 4-9. Although no significant differences were observed in the risk of infection in these groups, the girls of 10-13 and boys of 4-9 showed a tendency of higher infection when compared with the opposite sex of their same groups of age.

Table 3 - Odds ratio for T. cruzi antibodies in school children according to sex and age.

Group of age		Odds ratio	
(years)	male	female	Totala
10-13	1.5 (0.11-20.3)b	2.4 (0.34-26.7)	1.1 (0.3-3.85)
4-9	2.7 (0.43-28.08)	0.4 (0.04-2.33)	0.9 (0.26-3.31)

a. Odds ratio for 10-13 years old compared with 4-9 years ajusted by sex; b. Figures in parenthesis are confidence limits at 95%.

DISCUSSION

The national survey of seroprevalence of Chagas' disease in Paraguay carried out in endemic zones, showed a prevalence of 22% of positive *T. cruzi* serology in populations who mainly live of in the nearest departments of the capital city¹¹.

Although Asunción is, age group considered, a non-endemic area for Chagas' disease, this study showed a seroprevalence of 1.4% in the children living in marginal zones of the capital city.

A tendency of increasing seroprevalence with age found in this study it was also described in other surveys1.

However it is important to mention that no transmission was observed in the group of age under 6 years old. In Paraguay congenital transmission in maternities from non-endemic areas oscillates betwen 2 to 3%6 11 12.

Even though the main transmission via of the disease is the vectorial in rural areas, there are enough evidences to assume a transplacentary transmission in the seropositive children of this study, who were born in Asunción. However, some variables such us mobility to endemic areas and stability of residence, were difficult to be measured in this sample.

Since 82% of the seropositive mothers in this study came from endemic areas, migration should be considered as an important factor of the urban expansion of Chagas' disease, as previously suggested³ ¹⁴. Therefore, the analysis of the migration impact and migratory dynamics should be included as major factors for the definition of public health policies that will be applied in the various levels of the health system⁴.

According to our results, the main cause of migration (72.5% of the studied population) was the possibility of getting a job. This is probably the result of the low possibility of improvement in education and job opportunities in rural areas. The attraction pole for the migrant rural women in Paraguay is the marginal zone of urban areas where the life condition of the migrants becomes

worse because they have to perform activities linked to sub-employment or domestic tasks of low labor significance and reduced profit (Arias de Martínez SE: "Situación de la mujer en el Paraguay. Area de los Minifundios". Fondo de las Naciones Unidas para la Infancia (UNICEF). Sria. Técnica de Planificación. Depto de Desarrollo Social. Seminario de Alto Nivel. Asunción-Paraguay. Documento mimeografiado 58p, 1992).

The prevalence of chagasic infection in pregnant women is usually associated with migratory records from endemic areas and their socio-economic level, estimated by occupation, instruction and housing, is low14. In some areas, migratory flows from other endemic regions of rural areas induce and increase the congenital transmission frequency of Chagas' disease in the cities.

The urban persistence of Chagas' disease is attributable to a congenital transmission cycle of second generation³. When the family nuclei of the child was studied, only some brothers were seropositive. The seropositivity is explained by the congenital transmission and the seronegativity confirms that the chagasic mother does not always transmit the infection to all her children⁵. In this study, three seropositive mothers had 2 seropositive children each, having one seronegative child more.

Neither triatomines nor vestiges were found in the dwellings of the cases and only two cases referred having been bitten by the vector. The presence of the vector in urban areas is not frequent, but it is very important in endemic rural areas where a significant association between the presence of triatomines and the seropositivity of the children is found².

The mothers and tutors in our study stated that they knew the vector of Chagas' disease, especially for having seen it in their origin zones, but not having been bitten by it. The lack of awareness of the disease, the poor information and their declaration of the absence of the vector showed an indifferent attitude towards the

insect and the disease. The main occupation of the seropositive children mothers was housework. Although information about the causes of the lack of occupation outside of the house was not collected, it should be pointed out that it corresponded to a higher lack of financial resources of the family nuclei of the chagasic child.

The health system of Latin America mainly focuses his fight against Chagas' disease basically in the vector control in rural areas¹⁶ and in transfusion control in urban zones, but practically no effort has been put in congenital transmission control.

Surveillance by serological *T. cruzi* screening in school children could also be useful for mapping morbidity patterns and for detecting any changes in the time trend, thereby permitting comparisons between regions1.

Based in our results, it can be said that the chagasic children of marginal zones of Asunción belong to families who have migrated from endemic areas of Chagas' disease. They mostly migrate looking for jobs, live in crowding conditions and in the poorest neighborhoods of these zones.

They receive almost null assistance from the health system which is orientated to disease symptomatologies and not to their prevention and control. In this way, a silent widening of the chagasic frontier is being carried out because of the simultaneous confluence of all the biological and socio-economic characteristics of these migrant population groups.

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REFERENCES

- Andrade A, Zicker F, Luquetti A, Oliveira R, Silva S, Souza J, Martelli C. Surveillance of *T. cruzi* transmission by serological screening of schoolchildren. Bulletin of the World Health Organization 70:625-629, 1992.
- 2. Andrade A, Zicker F, Luquetti AO, Lonizete S, Souza JMP and Martelli CMT. Risk factors for *Trypanosoma*

- *cruzi* infection among children in central Brazil: A case-control study in vector control settings. The American Journal of Tropical Medicine and Hygiene 52:183-187, 1995.
- Azogue CE. La mujer y la enfermedad de Chagas congénito en Santa Cruz, Bolivia: Aspectos epidemiológicos y socio-culturales. Women and Tropical Diseases. International Development Research Centre(IDRC) MR3/4e, p. 62-70, 1992.
- Benencia R, Mercer H. Migraciones y Enfermedad de Chagas en la Argentina. Medicina. Buenos Aires 53 (supl I):38-39, 1993.
- Bittencourt ACL. Doenca de Chagas congénita na Bahia. Tesis. Revista. Bahiana Saude Pública 11:165-209, 1984.
- Browner WS, Black D, Newman TB, Hulley SB. Estimating sample size and power. *In:* Hulley S, Cummings S (eds) Designing Clinical Research. Williams & Wilkins, Baltimore, p.139-150, 1988.
- Canese J. Diagnóstico de la enfermedad de Chagas congénito. Revista Paraguaya de Microbiología 14:3-18. 1979.
- Dirección General de Estadísticas y Censos Divulgación de Resultados de Análisis del Censo de 1982, Asunción, Paraguay, p. 8-9, 1984.
- Kaspar P, Velázquez G, Monzón I, Meza T, Vera ME, Pozzoli L, Guillén I, Merlo R, Samudio M, Cabral M, Ferro E, Rodríguez A. Un nuevo kit para la detección de anticuerpos IgG anti *Trypanosoma cruzi*. Enfermedad de Chagas en el Paraguay, Memorias del Instituto de Investigaciones en Ciências de la Salud (I.I.C.S.), nº 15. EFACIM-JICA-Asunción, Paraguay, 1990.
- Rojas de Arias A, Funk de Isaac L. Situación de la Transfusión de sangre inocua en el Paraguay. Período (1990-1994). Annual Report (in press) nº 80, p. 46, 1997.
- Rojas de Arias A. Chagas' disease in Paraguay. Panamerican Health Organization. HCP/CHT/76/96, 1996.
- 12. Russomando G, Guillén I, Fleitas N, Carpinelli MT, Vera de Bilbao N, Ferreira ME, Samudio M, Velázquez G, Kasamatsu E, Rojas de Arias A. Prevalencia de la enfermedad de Chagas en embarazadas de la Maternidad Nacional de Asunción y Transmisión Transplacentaria. In: Libro de Resúmenes Congreso de Pediatría, Asunción, Paraguay, p. 28, 1991.
- 13. Sosa Estani S. Tratamiento de la infección parasitaria. Actualizaciones en la enfermedad de Chagas. In: Resúmenes del Simposio Satélite. Noviembre. Organismo Oficial del Congreso Nacional de

- Medicina. Córdoba, Argentina, p. 283, 1992.
- Streiger M, Fabbro D, Del Barco M, Beltramino R, Bovero N. Chagas congénito en la ciudad de Santa Fé. Medicina (Buenos Aires) 55:125-133, 1995.
- Towbin H, Staehelin T, Gordon J. Electrophoretic transfer of proteins from polyacrylamide gels to nitrocellulose: Procedure and some applications. Proceeding of Natural Academy of Sciences 76:4350-4354, 1979.
- World Health Organization (WHO). Report of Chagas' disease. Report of a WHO Expert Committee. Serie

- 811. Geneva, p. 47-51, 1991.
- World Health Organization. Advances in research on Chagas' disease: twenty years of progress. Special Programme for Research and Training in Tropical Disease. nº 20. Geneva, Switzerland: UNDP/WORLD/BANK/WHO p. 168, 1993.
- World Bank. World Development Report. Washington DC p. 216-218, 1993.