

HUMAN INFECTION WITH *Trypanosoma cruzi* IN NASCA, PERU: A SEROEPIDEMIOLOGICAL SURVEY (1)

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SUMMARY

We estimated the proportion of seropositivity for infection with *Trypanosoma cruzi* (Chagas' disease) in a sample of the rural population of the Province of Nasca, Department of Ica, southwestern Peru. Although *Triatoma infestans*, the only vector species identified in the Department of Ica, is often found in domestic environments, data of the extent of human infection with *T. cruzi* are scant. This study comprised 446 houses, known to be infested with triatomines, distributed in 19 rural localities. While visiting those houses we collected filter paper bloodspots from 864 occupants (of both sexes, aged one year or over). By means of the indirect fluorescent antibody test (IFAT), we detected anti-*T. cruzi* IgG antibodies in samples from 178 individuals (20.6%). Seropositivity was significantly more frequent in females (23.8%) than in males (17.5%). Among the 410 individuals in the 1- to 10-year-old age group (47.5% of the population sample), 85 (20.7%) were found seropositive, which is indicative of an early acquisition of the infection. Within this group no significant differences in seropositivity were associated with sex.

KEYWORDS: Chagas' disease; Seroepidemiology; Nasca, Peru.

INTRODUCTION

Chagas' disease is endemic in both the coastal desert and the forested eastern regions of Peru^{7, 9, 16, 17, 19, 20, 21}. ESCOMEL reported for the first time, in 1917, the occurrence of Chagas' disease in Peru, as quoted by CUBAS⁹, LUMBRERAS¹⁶ and VILLANUEVA²⁶. According to HERRER¹⁴, the vector species *Triatoma infestans* must have penetrated into the Peruvian territory around 1879, during the war between Peru and Chile. Since then, it has been found exclusively in domestic environments²⁶. The Province of Nasca, where this study was conducted, is part of the Department of Ica (southwestern Peru), which also comprises the provinces of Chincha, Pisco, Ica, and Palpa.

Triatomine bugs, locally known as *chirimachas*, were observed to occur in the Province of Nasca by AREBALO¹ in 1946. During the same year, an investigation by AYULO² in the Department of Ica failed to demonstrate infection of humans, other mammals and triatomines with *Trypanosoma cruzi*; but TEJADA²⁵ in 1962 and VILLANUEVA²⁶ in 1973 detected natural infection of *T. infestans* with *T. cruzi*, in the provinces of Nasca and Ica respectively. While conducting a seroepidemiological study in 14 localities of the Department of Ica, CORNEJO et al.⁸ (1986) found specimens of *T. infestans*

naturally infected with *T. cruzi* and, by the complement fixation (CFT) and the indirect hemagglutination (IHAT) tests, seropositivity in humans. More recently, CARRANZA & MARQUEZ (1992)⁵ used the indirect fluorescent antibody test (IFAT) and IHAT to identify anti-*T. cruzi* antibodies in serum samples from blood donors at two hospitals of Ica. PEREZ & TORRES (1994)²¹ also used IFAT and IHAT to detect anti-*T. cruzi* antibodies in prospective blood donors from the provinces of Palpa and Nasca.

Currently available seroepidemiological data are insufficient to convey an acceptable picture of the prevalence of American trypanosomiasis in Peru. We thus decided to design a first approach to a seroepidemiological investigation aimed at the human population of the Province of Nasca, where no comprehensive epidemiological study on Chagas' disease has been done, serological or otherwise. By means of a questionnaire, we collected information about housing conditions and the habit of keeping domestic animals in the immediate vicinity of, or inside the houses.

This study was aimed at gathering seroepidemiological data necessary to guide further investigation in Nasca, rather than es-

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timating the prevalence of Chagas' disease in each one of the localities where bloodspots were collected.

MATERIALS AND METHODS

The present study was done during 1994 and 1995. It comprised the examination of serum samples from residents of 19 rural localities of the Province of Nasca, viz: Belén Bajo, Buena Fe, Cantayo, Coyungo, Chauchilla, Copara, Hacienda Belén, Huachuca, Las Cañas, La Ayapana, Los Jardines, Orcona, Pajonal Bajo, Pongo Grande, Santa Fe, Saysongo, San Luís Pajonal, Taruga and Unión Victoria (Fig. 1).

According to data supplied by the Ministry of Health of Peru, there are 6,775 inhabitants and 1,544 houses (some of them vacant) in the region where this study was conducted²². No demographic information was available concerning Hacienda Belén. Nevertheless, we estimated that our sample included about 13% of the population of this region. The 446 dwellings surveyed had been previously found to be colonized by *T. infestans*, according to information supplied by personell in charge of field work on malaria.

We collected bloodspots, absorbed into Watman n° 3 filter paper, from 864 residents, of both sexes, aged one year or over. Estimated as recommended by FERREIRA & CARVALHO¹¹, each bloodspot contained approximately 15.18 µl of blood per cm² of filter paper.

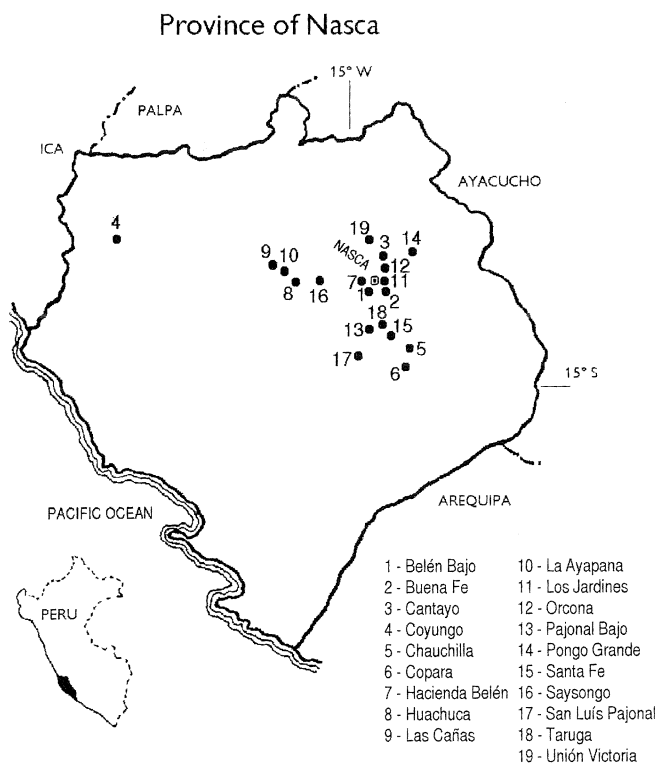


Fig. 1 - Spatial distribution of the 19 rural localities surveyed.

The Province of Nasca (4,828 km²) is part of the Department of Ica, to the southwest of the Peruvian coastal desert. It is bordered by the provinces of Ica and Palpa on the north, the Department of Arequipa on the south, the Department of Ayacucho on the east and the Pacific Ocean on the west (Fig. 1). The localities surveyed stand at an altitude of 588 meters.

Iron ore extraction, a wide range of handicrafts and tourism are among the most economically important activities in the Province of Nasca. Agriculture and cattle raising are sparse, largely due to local unfavorable climatic conditions¹⁵. A major tourist attraction in this province are the Lines of Nasca, huge figures traced on the dark surface of the desert soil, exposing the bright silica layer underneath. They represent human, animal, plant and geometrical shapes. Dated from the 3rd to the 6th century AD, the Lines of Nasca are, admittedly, astrological calendars. Such lines are best observed from the air¹⁵.

Most local houses have adobe walls in which crevices left by ill-fitting bricks are massively colonized by triatomine bugs. Although some of such walls are plastered, they often develop cracks for lack of maintenance. Earthen floors and roofs thatched with straw or cane are the rule. On the average, a house has 5 rooms. Traditionally, poultry and guinea-pigs are kept indoors. The guinea-pig (*cuy*, as it is called by locals), a notorious reservoir of *T. cruzi*⁶, was domesticated by the Incas in the pre-Columbian times.

The collection of blood samples in this region of the Province of Nasca was full of difficulties: no electricity, water in very short supply and most of the occupants of the houses out for work from dawn till dusk, only children remaining at home in the daytime.

We performed IFAT according to the technique described by CAMARGO³ with the modifications suggested by FERREIRA & CARVALHO¹⁰, using *T. cruzi* antigen fixed on microscope slides (Salck, São Paulo, Brazil), and fluorescein isothiocyanate conjugated caprine anti-human IgG (Salck, São Paulo, Brazil), previously standardized in terms of maximum reactivity (titer = 200). Positive (low and high titer) and negative control sera were used to check the intensity of fluorescence, as suggested by CAMARGO et al.⁴ and GUIMARÃES et al.¹³. In accordance with the recommendation by SADUN et al.²³, we chose a cut-off dilution of 1:16, not 1:32 (or its near equivalent 1:40) which is used frequently^{4,12}. The responsiveness of this population being unknown to us, we chose to improve sensitivity, even at the risk of some loss of specificity. Dilution series were conveniently expressed as powers of 2, as their logarithms (to the base 2) are integers²⁷. Doubtful results at the screening dilution were not considered, as we were not counting on the use of an alternative technique.

Statistical analysis of epidemiological data: G-test for independence²⁴. Level of significance 5% ($\alpha = 0.05$). Confidence limits (95%) for percentages were calculated by normal approximation to binomial²⁸.

RESULTS

IFAT seropositivity for anti-*T. cruzi* IgG antibodies was found in 178 (20.6%) of the 864 subjects under investigation, implying 95% confidence limits from 156 (18.0%) to 202 (23.4%). The num-

TABLE 1

Distribution by locality, and indirect fluorescent antibody test (IFAT) seropositivity for anti-*Trypanosoma cruzi* IgG antibodies, of 864 inhabitants of 19 localities of the Province of Nasca, Peru, 1994. Confidence limits for percentages calculated by normal approximation to binomial.

Locality	Population	Samples		Positive		Confidence limits (95%)	
		Examined	% Examined	Total	%	Lower %	Upper %
Buena Fe	289	177	61.2	26	14.7	10.2	20.7
Los Jardines	1,684	169	10.0	48	28.4	22.1	35.6
Copara	211	117	55.5	14	12.0	7.3	19.1
Pajonal Bajo	405	86	21.2	15	17.4	10.9	26.8
Cantalloc	232	46	19.8	13	28.3	17.3	42.5
Orcona	103	38	36.9	3	7.9	2.7	20.8
Taruga	187	34	18.2	8	23.5	12.4	40.0
Saysongo	155	34	21.9	9	26.5	14.6	43.1
Belén Bajo	45	33	73.3	16	48.5	32.5	64.8
Chauchilla	150	28	18.7	3	10.7	3.7	27.2
San Luís Pajonal	86	28	32.6	1	3.6	0.6	17.7
Las Cañas	21	26	123.8	7	26.9	13.7	46.1
La Ayapana	56	20	35.7	2	10.0	2.8	30.1
Huachuca	6	6	100.0	3	50.0	18.8	81.2
Pongo Grande	22	2	9.1	0	0.0	0.0	65.8
SUBTOTAL	3,652	844	23.1	168	19.9	17.4	22.7
Hacienda Belén	*	8	*	7	87.5	52.9	97.8
Coyungo	498	6	1.2	1	16.7	3.0	56.4
Santa Fe	2,367	5	0.2	1	2.0	3.0	62.4
Unión Victoria	258	1	0.4	1	100.0	20.7	100.0
TOTAL	6,775	864	12.8	178	20.6	18.0	23.4

* Population data lacking

bers of samples from Buena Fe, Los Jardines, Copara and Pajonal Bajo (Table 1) added up to 549 (63.5% of the total). This group included 103 (18.8%) seropositives. The remaining 315 samples (36.5% of the total) included 75 seropositives (23.8%). The hypothesis was accepted that seropositivity was not significantly different in both groups ($G = 3.074$ $p = 0.08$ [$X^2_{0.05(1)} = 3.841$]).

Table 2 shows the distribution of the 864 subjects according to seropositivity, sex and age group. The proportion of seropositives was significantly higher in females (23.8%) than in males. (17.5%). ($G = 5.29$ $p = 0.022$ [$X^2_{0.05(1)} = 3.841$]).

The 1-to 10-year-old age group was composed of 244 individuals (28.2% of the total), 244 males and 166 females. Se-

TABLE 2

Distribution, by age group, sex and indirect fluorescent antibody test (IFAT) seropositivity for anti-*Trypanosoma cruzi* IgG antibodies, of 864 inhabitants of 19 localities of the Province of Nasca, Peru, 1994.

Age group Females (years)	Males			Females			Males + Females		
	Total	Posit.	% Posit.	Total	Posit.	% Posit.	Total	Posit.	%Posit.
0 → 5	25	7	28.0	22	3	13.6	47	10	21.3
5 → 10	219	49	22.4	144	26	18.1	363	75	20.7
10→ 15	119	10	8.4	104	24	23.1	223	34	15.2
15→ 20	24	3	12.5	35	15	42.9	59	18	30.5
20→ 25	9	2	22.2	26	8	30.8	35	10	28.6
25→ 30	11	3	27.3	25	6	24.0	36	9	25.0
30→ 35	11	2	18.2	14	5	35.7	25	7	28.0
35→ 40	7	0	0.0	11	6	54.5	18	6	33.3
40→ 45	3	0	0.0	10	2	20.0	13	2	15.4
45→ 50	2	0	0.0	12	4	33.3	14	4	28.6
> 50	10	1	10.0	21	2	9.5	31	3	9.7
TOTAL	440	77	17.5	424	101	23.8	864	178	20.6

TABLE 3

Physical characteristics of the 446 domiciles investigated. Province of Nasca, Peru, 1994.

Walls	Number of houses	Percentage
Adobe	432	96.8
Mud-stick	6	1.3
Reed	6	1.3
Straw matting	1	0.2
Brick	1	0.2
Roof		
Reed	436	97.7
Thatch (straw)	10	2.2
Floor		
Earthen	232	52.0
Cement	214	48.0
Plaster		
Yes	345	77.4
No	101	22.6
Cracks		
Yes	371	83.2
No	75	16.8
TOTAL	446	100.0

TABLE 4

Domestic animals in 446 houses. Province of Nasca, Peru, 1994.

Animals	Number of houses	Percentage
Mammals and birds	319	71.5
Mammals only	54	12.1
Birds only	37	8.3
Subtotal	410	91.9
No animals	36	8.1
TOTAL	446	100

ropositivity was demonstrated in 56 males (23.0%) and 29 females (17.5%). This difference was not significant. ($G = 1.83$ p = 0.18 [$X^2_{0.05(1)} = 3.841$]).

Fig. 2 shows variation in seropositivity by age group.

Table 3 summarizes epidemiologically relevant peculiarities of local dwellings. Data about keeping domestic animals in or near the houses are shown in Table 4.

Serological results and the use of keeping animals indoors (Table 5) were demonstrated to be statistically independent ($G = 0.32$ p = 0.57 [$X^2_{0.05(1)} = 3.841$]).

DISCUSSION

A rather high prevalence of Chagas' disease in the Province of Nasca would be expected in view of the ubiquitous presence of *T. infestans* in domestic environments. Indeed, the proportion of

TABLE 5

Seropositivity or seronegativity by the indirect fluorescent antibody test (IFAT) of the population sample investigated and the presence or absence of animals in the houses. Nasca, Peru, 1994.

Inhabitants	Number of cases	Percentage
Positives: animals inside the house	165	92.7
Positives: no animals inside the house	13	7.3
Subtotal	178	100.0
Negatives: animals inside the house	627	91.4
Negatives: no animals inside the house	59	8.6
Subtotal	686	100.0
TOTAL	864	100.0

seropositivity we found in Nasca (20.6%) is closer to that recorded by NAQUIRA et al.¹⁸ elsewhere in southern Peru (32%) than to overall results for Peru (between 6% and 10%)¹⁷. In spite of the fact that Chagas' disease has for a long time been recognized as a serious public health problem in Peru, comprehensive epidemiological data concerning the human population are still lacking.

The results of this cross-sectional sampling study are indeed a cause for concern. The epidemiological factors involved in the natural transmission of Chagas' disease demonstrably occur throughout the region under investigation. *T. infestans* is omnipresent and local human habitations provide the necessary conditions for its breeding: most houses have adobe walls, but loosely laid bricks leave spaces which are profusely colonized by triatomines. Such spaces are also abundant in mud-stick and wooden walls (Table 5). Earthen floors and roofs thatched with reeds or straw are the rule.

Domestic animals, chiefly guinea-pigs and poultry are often kept indoors. The latter have been, since pre-Columbian times, used by locals as a source of proteins. Thus, an ecological relationship has been established between these animals and humans (HERRER¹⁴, 1955; VILLANUEVA²⁶, 1973). Under such circumstances, natural transmission of Chagas' disease is a hazard, particularly to very young children who remain indoors for long periods and to women, who handle the carcasses of possibly infected guinea-pigs. To what extent the latter circumstance is responsible for a higher proportion of infected women is a subject for investigation. As shown in Table 2, among 410 individuals in the 1- to 10-year old age group (47.5% of the population sample), 85 (20.7%) were found seropositive, which is indicative of an early acquisition of the infection.

A program intended to control the transmission of Chagas' disease has been recently started in Peru (CARRANZA & MARQUEZ⁵, 1992), but evaluation of its results imply a nationwide epidemiological investigation. Since the first observation by ESCOMEL, in 1917, only scattered data have been collected about the distribution of tritomines in Peru. The same applies to data about human infection, which has been serologically investigated by CORNEJO & LUMBRERAS (1951)⁷; CUBAS (1965)⁹; NAQUIRA (1967)¹⁷; LUMBRERAS (1972)¹⁶; HERRER

(1955)¹⁴; VILLANUEVA (1973)²⁶; CORNEJO et al. (1986)⁸; CARRANZA & MARQUEZ (1992)⁵; PEREZ & TORRES (1994)²¹.

In Peru, it is surmised that a considerable proportion of cases of cardiopathy and megaviscera, ascribed to other causes for lack of etiologic diagnosis, were indeed complications of Chagas' disease. As pointed out by CORNEJO et al.⁸, investigation is not often directed towards tracing sudden death cases back to Chagas' disease.

There is no question about the importance of Chagas' disease as a cause of disability among those who would otherwise compose the work force of Peru. As a chronic disease which can cause irreversible tissue damage and for which no satisfactory specific treatment is available, it involves heavy economical losses and human suffering.

A thorough understanding of the epidemiological conditions prevailing in the Province of Nasca, obviously invaluable in the control of natural transmission of Chagas' disease, has not been attained until now; but the experience acquired in other regions of South America, where natural transmission of American trypanosomiasis has eventually been controlled, can provide investigators and the community with relevant information and a great deal of stimulus to achieve this goal.

RESUMO

Infecção humana por *Trypanosoma cruzi* em Nasca, Peru: um inquérito soroepidemiológico

Estimamos a proporção de positividade sorológica para infecção chagásica em amostra da população rural da Província de Nasca, Departamento de Ica, sudoeste do Peru. Embora *Triatoma infestans*, o único vetor identificado no Departamento de Ica, seja encontrado frequentemente em ambientes domésticos, são escassos os dados sobre a infecção de indivíduos humanos por *Trypanosoma cruzi*. Este estudo compreendeu 446 unidades domiciliares sabidamente infestadas por triatomíneos, distribuídas em 19 localidades. Ao visitá-las colhemos, em papel-filtro, amostras de sangue de 864 pessoas de um ano ou mais de idade, sem seleção por sexo. Por meio da reação de imunofluorescência indireta (RIFI), detectamos anticorpos IgG anti-*T. cruzi* em amostras de 178 (20,6%) pessoas. A proporção de positivos foi significativamente maior nos indivíduos do sexo feminino (23,8%) do que nos do masculino (17,5%). Dentre os 410 indivíduos pertencentes ao grupo de idades entre 1 e 10 anos (47% da amostra da população), 85 (20,7%) resultaram sorologicamente positivos, o que indica aquisição precoce da infecção. Neste grupo, a positividade sorológica se mostrou independente de sexo.

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