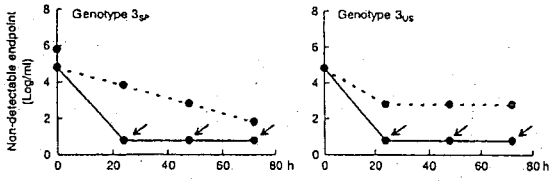

Fig. 2 Inactivation kinetics of the two HEV isolates during dry-heating. Solid lines: at 80°C. Broken lines: at 60°C. Arrow: infectious virus not detected.

Table 2 Viral removal by nanofiltration using filtres of various pore sizes

	HEV[a]				
BMM-filtre	3_{JPN} (swJB-N2)	3_{US} (swJB-MS)	3_{SP} (swJB-E10)	3_{SP} (cultured HEV[d])	4_{JP} (swJB-H1)
BMM-35N (35 ± 2 nm)	(6·1/4·8)[b] 1·3[c]	(6·9/< 3·3) ≥ 3·6	(6·4/3·8) 2·6	(6·0/< 3·2) ≥ 2·8	(5·6/4·5) 1·1
BMM-20N (19 ± 2 nm)	(6·1/< 2·3) ≥ 3·8	(6·9/< 3·3) ≥ 3·6	(6·4/< 3·2) ≥ 3·2	(6·0/< 3·2) ≥ 2·8	(5·6/< 3·0) ≥ 2·6
BMM-15N (15 ± 2 nm)	(6·1/< 2·3) ≥ 3·8	(6·9/< 3·3) ≥ 3·6	(6·4/< 3·2) ≥ 3·2	(6·0/< 3·2) ≥ 2·8	(5·6/< 3·0) ≥ 2·6

[a]HEV is in PBS.
[b]Genome amount is indicated as total log copies. Left: before filtration; right: after filtration.
[c]Log reduction factor. Log reduction factor was calculated from the genome amount in the samples before and after filtration.
[d]Derived from cultured media of HEV-infected A549 cells.

≥ 4·0 after treatment at 80°C for 24 h in any samples. However, although the infectivity of HEV was reduced at an LRF of 2·0 and 3·0, respectively, residual infectivity was detected in all samples that were treated at 60°C for 72 h (Fig. 2). These results indicated that the heat sensitivity is different not by genotype or cluster, but by the composition of the sample.

Filtration of HEV

The putative particle size was also evaluated using Planova filtres. All purified HEV isolates were removed to below the detection limit using Planova-15N and -20N, whereas significant amounts of HEV were detected after filtration using Planova-35N. In particular, the removability by Planova-35N was variable for the HEV isolates (Table 2). The result also showed a similar log reduction of viral removal between viruses derived from faeces and cell cultures of genotype 3_{SP}, and suggested that the diameter of viral particles in the purified sample derived from faeces was not affected by contaminants derived from faeces. These results may suggest that the particle size of HEV is around 35 nm, as previously reported [1].

Discussion

Several reports suggested that some industrial swine farms and commercial swine livers in industrial as well as developing countries could be contaminated by HEV [4,9]. Yazaki *et al.* detected HEV genomes in commercial swine livers that had been eaten by a hepatitis E-infected patient, as shown by the identical sequences of HEV in the liver and patient's sample by genome analysis. They reported that the patient became infected by eating uncooked liver [4]. Our infection studies using piglets demonstrated that HEV was mainly detected in liver, intestines, serum and faeces, but not detected in muscles [17]. Current epidemiological studies revealed that the prevalence of HEV RNA or anti-HEV IgG-positive blood donors in Hokkaido and Tokyo was 0·01% (56/432,167) of RNA and 3·9% of IgG, and 0·01% (3/44,322) of RNA and 8·6% of IgG, respectively. In addition, the prevalence of anti-HEV IgG in Japan varies according to locality, 1·0–8·6% [11]. These results also suggest that although the possibility of transmission is not considered to be high at the moment, some patients who have HEV in their blood may donate blood and this could lead to a transfusion-transmitted infection. Consequently, a monitoring study for donated blood has been initiated in Hokkaido, Japan.

Huang *et al.*, Emerson *et al.*, and Takahasi *et al.* reported on the heat sensitivity of HEV [13–15]. Several strains heated at 56°C for 1 h were sensitive. Some strains were inactivated to below the detection limit whereas in others, ~< 1% of the virus was still infectious. Unfortunately, these results were not shown with log reduction, time kinetics and effect by stabilizer at 60°C. Furthermore, there has been no report of heat inactivation of freeze-dried samples containing HEV. In

this study, we investigated the heat sensitivity in liquid and dry conditions over longer periods of time using several HEV isolates belonging to genotypes 3 and 4. The results suggest that the inactivation could be greatly influenced by the conditions. In addition, HEV was inactivated gradually at 60°C during dry-heating, whereas it was inactivated to below the detection limit within 24 h at 80°C. This result suggests dry-heating at 80°C to be effective for the inactivation of HEV [18]. The inactivation patterns of HEV at 60°C with albumin and fibrinogen were similar to those of canine parvovirus, which is used as a model of heat-resistant viruses (data not shown). This result suggests that HEV is a heat-resistant virus.

We also evaluated particle size using nanofilters that have a nominal pore size of 15, 19 and 35 nm using isolates from infected swine faeces and from medium cultured with the infected cells. The viral particle size is consistent with a diameter of around 35 nm as reported previously in an electronic microscopic analysis [1].

We reported that the heat sensitivity of parvovirus B19 is also influenced and subsequently varied its inactivation patterns, using different compositions of the inactivation matrix [19]. In addition, although the mechanism of viral particle removal by nanofiltration is size-exclusion, the removal capabilities of these virus-removal filters are also influenced by viral load and the condition/composition of the filtre [20–23]. Therefore, a safety evaluation for HEV contaminants, especially inactivation by heating and removal using, for example, nanofilters, should be performed using validated manufacturing conditions.

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No. 9

医薬品 研究報告 調査報告書

識別番号・報告回数			報告日	第一報入手日	新医薬品等の区分	総合機構処理欄
				2008. 10. 17	該当なし	
一般的名称	人全血液		研究報告の公表状況	Piron M, Vergés M, Muñoz J, Casamitjana N, Sanz S, Maymó RM, Hernández JM, Puig L, Portà s-M, Gascón J, Sauleda S. Transfusion. 2008 Sep; 48(9): 1862–8.	公表国	使用上の注意記載状況・その他参考事項等
販売名(企業名)	人全血液-LR「日赤」(日本赤十字社) 照射人全血液-LR「日赤」(日本赤十字社)				スペイン	人全血液-LR「日赤」 照射人全血液-LR「日赤」

研究報告の概要

○カタルーニャ(スペイン)の高リスク供血者における*Trypanosoma cruzi*感染の抗体陽性率

背景:ラテンアメリカ人のヨーロッパ(特にスペイン)移入が増加するにつれ、シャーガス病(中南米農村部に流行する人畜共通感染症)などの新たな病原体が報められるようになった。媒介生物サシガメがいない場合、非流行地域のシャーガス病伝播の主な形態のひとつは、輸血を介するものである。

研究デザインおよび方法:カタルーニャ血液銀行は、高リスク供血者におけるシャーガス病スクリーニング計画を実行し、供血者集団で*Trypanosoma cruzi*(*T. cruzi*)感染の血清学的検査陽性率を調査した。全検体にシャーガス抗体試験、ID-PaGIA (DiaMed社)とbioelisaシャーガス検査(Biokit社)の2種類の市販検査を使用した。

結果:全体の血清学的検査陽性率は0.62%であり、検査を実施した高リスク供血者1,770名のうち11名の陽性が確定した。最も陽性率が高かったのはボリビア出身の供血者であった(10.2%)。陽性供血者11名のうち1名は、シャーガス病流行地域に数年間滞在したことのあるスペイン人であった。さらに、陽性供血者の1名は、検出可能な寄生虫血症を呈した。

結論:本試験の結果は、非流行国の高リスク供血者に*T. cruzi*スクリーニング検査を実施する必要性を強調する。流行地域に居住経験のある(必ずしも当該地域で誕生することを意味するわけではない)高リスクに分類される人々を、検査対象に含めることが重要な知見として挙げられる。*T. cruzi*スクリーニングが全供血者に対してルーチンに実施されない場合には、供血前の問診における高リスク供血者の特定が重要である。

血液を介するウイルス、細菌、原虫等の感染
vCJD等の伝播のリスク

報告企業の意見	今後の対応
スペイン、カタルーニャの高リスク供血者集団における*T. cruzi*感染の抗体陽性率は0.62パーセントであったとの報告である。	日本赤十字社は、輸血感染症対策として献血時に海外渡航歴の有無を確認し、帰国(入国)後4週間は献血不適としている。また、シャーガス病の既往がある場合には献血不適としている。日本在住の中南米出身献血者については、厚生労働科学研究「献血血液の安全性確保と安定供給のための新興感染症等に対する検査スクリーニング法等の開発と献血制限に関する研究」班と共同して検討する予定である。今後も引き続き情報の収集に努める。

Seroprevalence of *Trypanosoma cruzi* infection in at-risk blood donors in Catalonia (Spain)

Maria Piron, Mireia Vergés, José Muñoz, Natàlia Casamitjana, Sergi Sanz, Rosa María Maymó, José Manuel Hernández, Lluís Puig, Montserrat Portús, Joaquim Gascón, and Sílvia Sauleda

BACKGROUND: The increasing arrival of Latin Americans to Europe and, particularly, to Spain has led to the appearance of new pathologies, such as Chagas disease, a zoonotic infection endemic to rural areas of Central and South America. In the absence of the triatomid vector, one of the main modes of transmission of Chagas disease in nonendemic regions is through blood transfusion.

STUDY DESIGN AND METHODS: The Catalonian Blood Bank has implemented a screening program for Chagas disease in at-risk blood donors and has performed a study to determine the seroprevalence of *Trypanosoma cruzi* infection in the donor population. The two commercial tests used in all samples were the ID-PaGIA Chagas antibody test (DiaMed) and the bioelisa Chagas assay (Biokit).

RESULTS: Overall seroprevalence was 0.62 percent, with 11 donors confirmed positive among the 1770 at-risk donors studied; the highest rate (10.2%) was in Bolivian donors. Interestingly, 1 of the 11 positive donors was a Spaniard who had resided various years in a Chagas disease endemic area. Furthermore, 1 of the positive donors presented detectable parasitemia.

CONCLUSION: The results of this study emphasize the need for *T. cruzi* screening in at-risk blood donors in nonendemic countries. An important finding is the relevance of including in the at-risk category persons who have resided in, but were not necessarily born in, an endemic region. If *T. cruzi* screening is not routinely performed in all donations, it remains highly dependent on proper identification of at-risk donors during the predonation interview.

American trypanosomiasis or Chagas disease is a zoonotic infection endemic to Latin America. In endemic countries, approximately 8 million people are carriers of the disease, approximately 50,000 new cases are diagnosed every year, and fatal cases are estimated at 14,000 per year.[1]

Trypanosoma cruzi, the causal agent of Chagas disease, can be detected in blood during the initial acute phase, which lasts from 6 to 8 weeks. Most patients are asymptomatic or oligosymptomatic, but when symptoms manifest, the acute stage of the illness may be characterized by fever, lymphadenopathy, mild splenomegaly, and edema, sometimes involving the myocardial tissue and producing acute myocarditis or encephalomyelitis. If they remain untreated, 5 to 10 percent of these patients die.[2] After this phase, the infection usually progresses to the chronic stage, in which the parasite is rarely detected in blood. When it is clinically silent, the chronic phase is called the indeterminate form of the disease. Many patients remain in this clinical situation for the rest of their lives, but 15 to 30 percent will progressively develop symptomatic disease.[2,3] Cardiologic manifestations are

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the hallmark of the chronic stage. The most threatening complications are heart failure and excitability and conductivity disorders leading to cardiac arrhythmia and sudden death. These conditions often require recurrent hospitalization, surgery, or more expensive cardiologic procedures such as pacemakers, implantable automatic defibrillators, and even heart transplants.[2,4] Less frequently, Chagas disease involves the digestive tract.[2,3]

In endemic areas, Chagas disease is commonly transmitted by a triatomid vector that releases parasite-infected excreta into lacerated skin or mucosa. Congenital and transfusion-related transmission are the other principal modes of acquiring *T. cruzi* infection.[2,5] Transmission of Chagas disease via blood transfusion has been recognized since 1952,[6] but it was only with the advent of the HIV pandemic in the 1980s that blood control programs began to be implemented in most Latin American countries. Legislation requiring blood transfusion screening has decreased the incidence of transfusion-related Chagas disease. There are varying degrees of success, however, in implementing these control measures in some endemic regions.[7]

In countries where it is not endemic, such as Spain, Chagas disease is considered an emerging infection because of the increasing number of immigrants coming from Latin America. Spain houses approximately 4 million immigrants, and 1.5 million of them were born in a country endemic for Chagas disease.[8]

Transmission of *T. cruzi* in countries where the vector does not exist occurs mainly through maternal-fetal transmission, organ transplantation, and blood transfusion.[9] Despite this knowledge and confirmed reports of *T. cruzi* infection through congenital transmission[10,11] and blood transfusion in nonendemic countries,[12] little attention has been paid to assuring optimal screening and control measures.

Since September 2005, Spanish regulatory law requires that all at-risk donors be screened for Chagas disease or otherwise be excluded from donation.[13] Donors considered at risk by the Spanish Ministry of Health include persons born in an endemic area, those born of a mother native to an endemic area, and those who have undergone transfusion in an endemic area. The main objective of this article is to estimate the prevalence of *T. cruzi* infection in blood donors in Catalonia through implementation of a *T. cruzi* antibody screening test in donors considered at risk by the Spanish Ministry of Health, as well as all residents for more than 1 month in an endemic area.

MATERIALS AND METHODS

Donor selection and study design

Individuals included in the study belonged to one of the following risk groups: Group 1, donors born or transfused in an endemic area; Group 2, donors born of a mother native to an endemic area; and Group 3, residents in an endemic area for more than 1 month. For the first group, which was expected to contain the largest number of individuals, we calculated a sample size of 1500 subjects for an estimated prevalence of 0.6 percent of *T. cruzi* infection (95% CI, 0.2%-1%). Blood donation was accepted if there was no other reason for rejection (e.g., malaria). In patients who had grounds for rejection, a blood sample was requested only for *T. cruzi* determination.

Each donor answered an epidemiologic questionnaire to obtain information on age, sex, birth place, date of arrival in Spain, visits to endemic regions in Latin America, and living conditions in the endemic area (rural environment, adobe house). The donors signed an informed consent form and the study design was approved by the Ethics Committee for Research of our center. Clinical assessment and follow-up was offered to all positive donors.

Detection methods

Serum samples from at-risk donors were processed for the presence of *T. cruzi* antibodies by two EC-approved tests, according to the manufacturer's instructions. Each of these tests claimed 100 percent sensitivity based on various performance evaluation studies presented in the insert. Screening was performed with a commercially available Chagas antibody test (ID-PaGIA, DiaMed, Cressier sur Morat, Switzerland), a particle gel immunoassay that contains two recombinant antigens: Ag2 and TcE. All blood donations with an initially reactive result in the screening test were rejected. It should be noted that independently of the result of Chagas determination, platelet concentrates were not made from at-risk donors.

The second test used in all samples was the Chagas bioelisa assay (Biokit, Lliçá d'Amunt, Spain), which also contains a recombinant antigen, TcF antigen (*T. cruzi* fusion protein), and consists of a linear assembly of four serologically active peptides PEP-II, TcD, TcE, and TcLoE1.2. When a positive result was obtained in at least one of these tests, a conventional in-house enzyme-linked immunosorbent assay (ELISA) test utilizing whole *T. cruzi* antigens from Maracay strain epimastigotes was also performed. Samples were confirmed positive when at least two tests gave a positive result (Fig. 1).

All initially positive samples by ID-PaGIA Chagas antibody test and/or Chagas bioelisa assay were retrospectively tested with the *T. cruzi* ELISA test system (Ortho-Clinical Diagnostics, Raritan, NJ), which was FDA- and EC-approved after the beginning of this study. This last test uses epimastigote lysate antigens.

Furthermore, all initially positive samples were assessed for the presence of parasite DNA in blood, using in-house real-time polymerase chain reaction (PCR).[11]

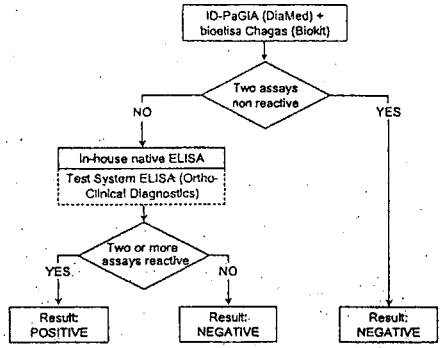

Fig. 1. Algorithm for *T. cruzi* serology interpretation.

The PCR technique is designed to amplify a highly represented fragment of 166 bp in the satellite DNA of *T. cruzi*, it contains an internal control for DNA extraction and amplification (human RNase P gene), and has an estimated sensitivity of 2 parasites per mL (95% positive hit rate).

RESULTS

Epidemiologic data

Between September 2005 and September 2006, a total of 1770 donors were enrolled in the prevalence study and were screened for *T. cruzi* antibodies. These individuals accounted for 1.1 percent of all blood donors in the first 3 months of the study (Table 1).

Sex distribution (51% men) was similar to that of the general Catalonian donor population (53% men), whereas the mean age was lower than that of the general donor population (35 ± 11 years vs. 42 ± 12 years). Approximately half the donors included in the study arrived to Spain after 2000, 5 years before the beginning of recruitment for the study.

According to risk groups, 1524 (86.1%) individuals were born in an endemic area (Group 1), 37 (2.1%) were born of a mother from an endemic area (Group 2), and 209 (11.8%) were temporary residents in an endemic country (Group 3; Table 1). Twenty-one donors (1.2%) stated that they had undergone transfusion in a country endemic for Chagas disease. Only 20.7 percent of donors born in an endemic area stated that they had lived in a rural environment and only 9 percent declared to have lived in an adobe house. For temporary residents, the proportions were 66.5 and 22 percent, respectively (Table 2).

The most highly represented country of origin was Colombia, accounting for 22.3 percent of at-risk donors included in the study, followed by Argentina and Ecuador, accounting for 19.5 and 14.6 percent, respectively (Table 3). The majority of mothers of the 37 donors in Group 2 came from Argentina (10), followed by Colombia (7), Chile (7), and Peru (3). Most donors from Group 3 (n = 209) had visited various endemic countries during one or several trips.

Prevalence of *T. cruzi* infection in blood donors in Catalonia

In the serologic screening, 21 donors presented an initially reactive result by ID-PaGIA Chagas and 25 by bioelisa Chagas. Samples showing faint agglutination with the use of ID-PaGIA or an inconclusive result with bioelisa (ratio absorbance:cutoff between 0.9 and 1) were considered initially reactive. Only 11 donors were reactive in both tests. The third test (in-house ELISA) was only positive in the 11 serum samples that resulted positive by the two commercial tests used in the screening (Table 4). The results obtained with the *T. cruzi* ELISA test system (Ortho-Clinical Diagnostics) agreed with those obtained with the in-house ELISA (35/35), also based on whole parasite lysate antigens. In addition, 1 of the 11 donors had detectable parasitemia by PCR analysis.

Overall prevalence was 0.62 percent in the at-risk population. Ten of the eleven positive donors were from Group 1 (0.66%), and one was from Group 3 (0.48%) (Table 5). The countries of origin of positive donors were Bolivia (6 cases), Argentina (2), Ecuador (1), and Paraguay (1), and there was one Spaniard who had been living in Venezuela for 27 years. We should emphasize that the number of positive subjects among Bolivians (6 out of 59 Bolivian donors) represents a prevalence of 10.2 percent for this country. None of the 37 donors born of a mother native to an endemic area and none of the donors transfused in an endemic area (n = 21) were positive for *T. cruzi* antibodies. Only 3 of the 11 positive donors declared that they had been living in a rural area or an adobe house (Table 5).

DISCUSSION

In endemic countries, blood transfusion is the second most important way to acquire Chagas disease. Screening coverage in blood banks has reached 100 percent in many countries, and this has reduced the risk of transmitting the infection by transfusion.[15] Nevertheless, cases of *T. cruzi* transmission by blood transfusion have been recently described in Mexico where screening coverage, which is not mandatory at this time, is one of the lowest of all Chagas disease endemic countries.[15,16]

In nonendemic countries, blood transfusion is one of the main modes of acquiring the infection, and cases of transmission before screening for *T. cruzi* infection became mandatory in blood donors have been reported in Spain.[17,18] European legislation requires permanent rejection of persons with a history of Chagas disease for blood donation.[19] Nevertheless, most people do not present any health problem until many years after acquiring the infection. Because of the increasing number of people from Latin America residing in Europe, and European people who reside for a time in an endemic area, implementation of screening programs for this disease in at-risk donors may be advisable in all European blood banks.

The Catalonian Blood Bank implemented a screening program for Chagas disease in all at-risk donors and simultaneously initiated a study to determine the seroprevalence of *T. cruzi* infection in its blood donor population. The countries of origin of the largest percentages of at-risk donors in the present study were Colombia,

TABLE 1. Epidemiologic data of donors included in the study

Donors included by group of risk		Transfused in endemic area*	Sex		Deferred before donation*	Age (years)†
Group	Number (%)		Male*	Female*		
1. Born in an endemic area	1524 (86.1)	21 (1.4)	758 (49.7)	766 (50.3)	95 (6.2)	35 (10.7)
2. Born of a mother native to an endemic area	37 (2.1)	0	18 (48.6)	19 (51.4)	1 (2.7)	28 (10.0)
3. Temporary resident in an endemic area	209 (11.8)	0	119 (56.9)	90 (43.1)	19 (9.0)	38 (10.7)
Total	1770	21 (1.2)	895 (50.6)	875 (49.4)	115 (6.5)	35 (10.8)

* Data are reported as number (%).
† Data are reported as mean (SD).

TABLE 2. Living conditions in endemic area

Group 1: donors born in endemic region		Group 3: resident in endemic region	
Has lived in rural area	Has lived in adobe house	Has lived in rural area	Has lived in adobe house
315/1524 (20.7%)	137/1524 (9.0%)	139/209 (66.5%)	46/209 (22.0%)

TABLE 3. Distribution of donors born in an endemic region and of positive donors by country of origin

Country	Tested for anti-*T. cruzi**	Percentage of official immigrant population in Catalonia	Number	Anti-*T. cruzi*–positive donors Rate by country (%)
Colombia	340 (22.3)	13.8		
Argentina	298 (19.5)	11.7	2	2/298 (0.67)
Ecuador	223 (14.6)	28.2	1	1/223 (0.45)
Uruguay	127 (8.3)	4.4		
Peru	123 (8.1)	8.9		
Brazil	113 (7.4)	3.9		
Venezuela	86 (5.6)	2.4		
Chile	77 (5.0)	4.2		
Bolivia	59 (3.9)	8	6	6/59 (10.2)
Mexico	40 (2.6)	2.6		
Paraguay	15 (1.0)	1.1	1	1/15 (6.7)
Honduras	10 (0.7)	1.3		
El Salvador	6 (0.4)	0.4		
Nicaragua	3 (0.2)	0.1		
Costa Rica	2 (0.1)	0.1		
Guatemala	1 (<0.1)	0.1		
Panama	1 (<0.1)	0.1		
Total	1524		10	

* Data are reported as number (%).

TABLE 4. Distribution of results obtained with the two commercial kits ID-PaGIA (DiaMed) and bioelisa Chagas (Biokit)*

	Initial result with the bioelisa Chagas	
Initial result with ID-PaGIA	Positive	Negative
Positive	11†	10‡
Negative	14‡	1735

* All initially reactive results were confirmed as positive or negative by in-house native ELISA. Cohen's kappa index, 0.471.[31]
† In-house native ELISA result positive.
‡ In-house native ELISA result negative.

TABLE 5. Epidemiologic data of the 11 positive donors

Positive donor number	Sex (male/female)	Age at donation (years)	Country	Town, State	Did you live in a rural area?	Did you live in an adobe house?	Born in Spain	Date of arrival in Spain	Have you returned recently to your country?	Transfusion in an endemic country
1	F	34	Ecuador	Machala, El Oro	No	No	No	2000	Yes	No
2	F	34	Bolivia	Cochabamba, San Benito	Yes	Yes	No	2002	No	No
3	M	42	Argentina	Guaymallen, Mendoza	Yes	Yes	No	2002	No	No
4	F	36	Bolivia	Santa Cruz, Santa Cruz	No	No	No	2005	Yes	No
5	M	38	Bolivia	Santa Cruz, Santa Cruz	No	No	No	2004	No	No
6	M	45	Bolivia	Santa Cruz, Santa Cruz	No	No	No	2003	No	No
7	F	31	Venezuela	Caracas	Yes		Yes	2003		No
8	F	36	Bolivia	Cochabamba, Cochabamba	No	No	No	2003	No	No
9	F	40	Bolivia	Santa Cruz	No	No	No	2003	No	No
10	M	49	Argentina	San Juan	No	Yes	No	1988	No	No
11	F	51	Paraguay	San Estanislas, San Pedro	No	No	No	1978	Yes	No

Argentina, and Ecuador, and these were also the countries of origin of the largest percentages of immigrants in Catalonia in 2005 (Table 3).[8]

Overall seroprevalence was 0.62 percent in the 1770 at-risk donors included, and positive donors were mainly from Bolivia, with a 10.2 percent prevalence among donors from this country. The seroprevalence of *T. cruzi* infection in Bolivian donors is very high and is in keeping with the 9.9 percent reported in 2001 in that country (86.1% screening coverage at the time of the study), which is the most highly affected by Chagas disease.[15] The remaining positive donors born in endemic areas were from Argentina, Paraguay, and Ecuador. The seroprevalence of *T. cruzi* infection in blood donors reported in 2001 or 2002 for these countries was 4.5 percent (second most highly affected country), 2.8 percent (third most highly affected country), and 0.4 percent, respectively.[15]

One important finding of this study is the relevance of including persons who have resided in, but were not necessarily born in, an endemic area as an at-risk donor group for *T. cruzi* infection. This population is not considered at risk in the current Spanish regulations.[13] One of the 11 positive donors described herein was born in Spain and had resided for many years in Venezuela.

Various studies have reported seroprevalence data in the immigrant population and in blood donors in countries that are not endemic for Chagas disease. In Canada and Germany, for example, seroprevalences of 1 and 2 percent have been described, respectively, in cohorts of asymptomatic immigrants coming from Latin America.[20,21]

As to blood donors, two recent surveys in the United States reported a seroprevalence of 0.02 to 0.03 percent among all donors in blood centers in California, Arizona,[22] and Texas.[23] A previous study carried out in Los Angeles and Miami blood centers identified 7.3 and 14.3 percent of donors as at risk for Chagas disease, with a 0.2 and 0.1 percent seroprevalence of *T. cruzi* infection, respectively, in these at-risk populations.[24]

In Spain, some blood banks have implemented Chagas' disease screening in at-risk donors and seroprevalence data have been described, although some of the results are preliminary. *T. cruzi* infection seroprevalence varies from 0.05 to 1.38 percent in the available studies.[17,25-27] A mean seroprevalence of 0.65 percent can be calculated from data proceeding from all Spanish blood centers that have performed (or initiated) a survey, including, as a whole, 10,388 blood donors at risk for *T. cruzi* infection. The results obtained in Catalonia are consistent with these data.

The epidemiologic questionnaire provided some interesting information. First, the mean age of the at-risk donors proceeding from an endemic area (Group 1 donors) is lower than the general no-risk population (35 years vs. 42 years), as would be expected in immigrants who generally come to Spain to work and improve their

living conditions. Half the population included arrived in Spain after 2000, a fact that illustrates the increasing immigration rates from Latin America observed over the past years. Another interesting result from the questionnaire was that the information obtained about living conditions in the Chagas disease endemic area (rural area, adobe house) did not correlate with the presence or absence of antibodies to *T. cruzi*. People born in endemic regions (7 of 11 positive donors) generally declared that they had never lived in a rural environment or an adobe house (Table 2), as is commonly assumed. Hence, this question is not useful for differentiation purposes. Interestingly, the same conclusion was drawn from the Berlin study, in which 95 of 100 immigrants declared that they came from an urban area, including the 5 cases of confirmed Chagas disease.[21]

The two serologic assays used in this study were chosen because at the beginning of the study they were commercially available and EC-marketed. Both are based on recombinant antigens, whereas the third conventional in-house ELISA is based on whole parasite lysate. All samples confirmed as positive had been initially reactive with both recombinant antigens assays, and all samples initially reactive with only one assay presented a nonreactive result in the in-house ELISA and were considered false-positive samples. It is worth noting that many discrepant results observed between both assays corresponded to low 0.9 to 1 signal-to-cutoff rates for bioelisa Chagas (Biokit) or doubtful reactions with ID-PaGIA (DiaMed), which were all considered as initially reactive in this study. Additionally, it should be mentioned that the *T. cruzi* ELISA test system performed on all initially reactive samples (with one or two tests) confirmed the results obtained with the conventional in-house ELISA. The high rate of inconclusive or false-positive results obtained when one diagnostic test is used underscores the need to confirm all initially positive results with a second serologic technique. In any case, there is still a need for a real confirmatory test to overcome the issues of discrepancies and false results (positive or negative). The ID-PaGIA assay allows testing of a small number of samples at a time. Although this system has the drawback of rather subjective reading, it could be useful in blood centers with a small volume of donations and is now even more reliable since a third antigen has been recently added to increase the sensitivity of the test. The ELISA format, which allows for automation and objective reading, should be indicated in other blood centers. An even more appropriate strategy would be the use of two screening tests, one based on recombinant antigens and the other on crude antigens.[28]

In summary, this study reports a seroprevalence of *T. cruzi* infection of 0.62 percent among at-risk donors in Catalonia and emphasizes the need to include individuals who have resided in, but were not necessarily born in endemic areas as at-risk donors. The difficulty of this type of selective screening is proper identification of the risk population, which essentially depends on the predonation interview. Latin Americans accounted for more than 1 percent of the total of donors in our study, and this substantial contribution underscores the need to accept them as donors.

In the future, techniques to inactivate or reduce the parasite load, which are currently under development or evaluation,[29,30] might be applicable to blood components. At this time, however, detection of *T. cruzi* infection is the only preventive measure available to accept at-risk blood donors.

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No. 33

医薬品 研究報告 調査報告書

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研究報告の概要

○アレナウイルスと特定された未知の疾患

南アフリカ、ヨハネスブルグで3名の死者を出したウイルスは、暫定的に西アフリカのラッサウイルスに近い、齧歯類媒介性アレナウイルスであると特定された。ウイルスは感染マウスの排泄物を介し、人間の食物やハウスダストを汚染する可能性がある。

南アフリカ国立感染症研究所(NICD)と保健省は共同で、このウイルスが体液を介してヒトからヒトに感染するため、「患者の看護に特別な予防的措置が必要である」との声明を発表した。3名の死因を確定するには更なる検査が必要である。新たなアレナウイルスが南アフリカの齧歯類に存在することはまだ示されていないとNICDは述べた。

ウイルスであるかどうか、ならびに当該ウイルスの分布について検討を行う必要がある。ヒトに疾患を引き起こすアレナウイルスが南

1人目の女性患者は、9月中旬に重篤な容態でザンビアから搬送され、Morningside Medi-Clinicに入院し、2日後に死亡した。約2週間後、1人目の患者の搬送に同行した救急救命士が死亡し、間もなく看護師が死亡した。

1人目の患者と接触した他の3名の患者は退院したことが確認されているが、依然として2名が厳重な監視下に置かれている。1名は、発熱およびインフルエンザ様症状を発症した救急救命士であり、もう1名は2人目の患者をケアした女性看護師である。彼女は、隔離され抗ウイルス剤ribavirinの投与を受けており、現在は安定している。

今後の対応	
報告企業の意見	
南アフリカ、ヨハネスブルグで3名の死者を出したウイルスは、暫定的に西アフリカのラッサウイルスに近い、齧歯類媒介性アレナウイルスであると特定されたとの報告である。	日本赤十字社では、輸血感染症対策として問診時に海外渡航歴の有無を確認し、帰国(入国)後4週間は献血不適としている。今後も引き続き、新興・再興感染症の発生状況等に関する情報の収集に努める。

使用上の注意記載状況・その他参考事項等

人全血液-LR「日赤」
照射人全血液-LR「日赤」

血液を介するウイルス、細菌、原虫等の感染
vCJD等の伝播のリスク

(1)

BuaNews ONLINE
www.buanews.gov.za

Compiled by the Government Communication and Information System
Date: 13 Oct 2008
Title: Unknown illness identified as Arenavirus

By Luyanda Makapela

Johannesburg - The virus which has caused the death of three people has been provisionally identified as the rodent-borne Arenavirus.

The Arenavirus, related to the Lassa Fever Virus of West Africa, causes chronic infections in multimammate mice. Infected mice's excretion contains the virus which can contaminate human food or house dust.

A joint statement by the National Institute for Communicable Disease (NICD) and the Department of Health explained that the Arenavirus is a disease spread from human to human through the contact of body fluids.

"Special precautions are required in nursing patients," a statement said.

The finding follows blood samples being sent to Atlanta, in the United States to determine the cause of the deaths of three people who had been suspected of contracting Viral Haemorrhagic Fever.

The virus is similar to Lassa Fever, the department said. It has previously been found in rodents elsewhere in Africa, but has not been found to cause disease in humans other than in West Africa.

Further tests are needed to confirm the diagnosis by growing the virus in culture.

"It needs to be determined whether it is a previously unrecognised member of the Areaviruses, and what its distribution is. There is no indication as yet that Arenaviruses which cause disease in humans are present in South African rodents," the NICD said.

The first victim, who had to be flown in from Zambia in a critical condition, was admitted to the Morningside Medi-Clinic in mid September. She died two days later.

About two weeks later, the paramedic who had flown in with the first victim, was admitted at the same clinic presenting the same symptoms.

A nurse, Gladys Mthembu died shortly afterwards. According to certain reports Ms Mthembu's family has been given a go-ahead to continue with the funeral arrangements as her bedroom had been cordoned off by health officials

Maria Mokubung, a cleaner at the Morningside Medi-Clinic, who also died last weekend has since been ruled out as a possible victim of the virus

Meanwhile the Gauteng Health Department has confirmed that the three other patients, including nurse's female supervisor, who had been under observation for showing symptoms of the virus have been discharged.

They had been in contact with the nurse who died.

However, departmental spokesperson Phumelele Kaunda said there were two contacts that were still under active surveillance after being admitted for observation.

The one patient is a paramedic who had contact with the first patient and developed fever and flu-like symptoms. He was admitted initially in Flora Clinic and then transferred to Morningside Medi-Clinic with a diagnosis of kidney stones.

The other patient is a nurse who attended to the second patient and developed signs and symptoms similar to the first three patients. She is being treated in isolation and received the anti-viral medication, ribavirin. The patient is presently stable.

Gauteng Health MEC Brian Hlongwa meanwhile has sent condolences to the families of those that were killed by the viral infection, particularly families of health professionals who died in the line of duty.

"This illustrates the dedication of our health professionals and the need to society to respect and honour the work that they do," said MEC Hlongwa.

He also thanked the NICD, the National Health Laboratory Service, Centre for Disease Control in Atlanta and the World Health Organisation for ensuring that the results were made available soon. - BuaNews