

tivitis during July–September 2007. Reported treatment included home remedies and a nonprescription, locally available, cream containing a sulfa drug. Review of municipal hospital records during June 1–September 30, 2007, identified no additional cases of conjunctivitis. After the last case-patient died, 17 other persons were identified with purulent conjunctivitis: 4 at the municipal hospital and 13 during active case-finding in schools and the community. All were treated with oral amoxicillin and chloramphenicol optic solution, and 76 contacts were treated prophylactically with oral rifampin. No further suspected BPF cases were detected. Test results for acute arbovirus infection and PCR were negative for all patients (Table).

This outbreak of highly fatal illness is clinically compatible with BPF. Compatible features included young age, antecedent purulent conjunctivitis, signs and symptoms (i.e., antecedent conjunctivitis, fever 39.5°C–41.0°C, abdominal pain, nausea, vomiting, petechiae or ecchymoses), and high case-fatality rate. Epstein-Barr infection has reportedly produced similar symptoms (4) but with an illness lasting >7 days in contrast to the <24 hours for our case-patients.

We did not detect *H. aegyptius* in peripheral blood by culture or in serum or CSF by PCR in the 2 surviving children and in contacts of case-patients. One reason could be the remoteness of the investigation site, which resulted in improper sample collection, storage, and processing in the field before samples reached reference laboratories. Hemagglutination tests for arboviruses have low specificity. Therefore, another known or novel pathogen could have caused these cases.

Timely treatment with antimicrobial drugs for patients with suspected disease, prophylaxis of contacts, and treatment of children with conjunctivitis resulted in no additional cases. Intensive surveillance for febrile illness

preceded by conjunctivitis, immediate treatment, contact prophylaxis, and appropriate prompt laboratory testing are essential for continued control of BPF in this region.

**Eucilene A. Santana-Porto,
Adriana A. Oliveira,
Marcos R.M. da Costa,
Amiraldo Pinheiro,
Consuelo Oliveira,
Maria L. Lopes, Luiz E. Pereira,
Claudio Sacchi,
Wildo N. Araújo,
and Jeremy Sobel**

Author affiliations: Ministry of Health, Brasília, Brazil (E.A. Santana-Porto, A.A. Oliveira, W.N. Araújo, J. Sobel); Municipal Secretariat of Health, Anajás, Brazil (M.R.M. da Costa); Secretariat of Health of Pará State, Belém, Brazil (A. Pinheiro); Ministry of Health, Belém (C. Oliveira, M.L. Lopes); Instituto Adolfo Lutz, Secretariat of Health of São Paulo State, São Paulo, Brazil (L.E. Pereira, C. Sacchi); and Centers for Disease Control and Prevention, Atlanta, GA, USA (J. Sobel)

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References

1. Tondella ML, Paganelli CH, Bortolotto IM, Takano OA, Irino K, Brandileone MC, et al. Isolation of *Haemophilus aegyptius* associated with Brazilian purpuric fever, of Chloropidae (Diptera) of the genera *Hippelates* and *Liohippelates* [in Portuguese]. Rev Inst Med Trop Sao Paulo. 1994;36:105–9. DOI: 10.1590/S0036-46651994000200002
2. Kerr-Pontes LR, Ruffino-Neto A. Epidemiological study of Brazilian purpuric fever. Epidemic in a locality of São Paulo state (Brazil), 1986 [in Portuguese]. Rev Saude Publica. 1991;25:375–80. DOI: 10.1590/S0034-89101991000500009
3. Harrison LH, Simonsen V, Waldman EA. Emergence and disappearance of a virulent clone of *Haemophilus influenzae* biogroup *aegyptius*, cause of Brazilian purpuric fever. Clin Microbiol Rev. 2008;21:594–605. DOI: 10.1128/CMR.00020-08
4. Virata M, Rosenstein NE, Hadler JL, Barrett NL, Tondella ML, Mayer LW, et al. Suspected Brazilian purpuric fever in a toddler with overwhelming Epstein-Barr virus infection. Clin Infect Dis. 1998;27:1238–40. DOI: 10.1086/514988

Address for correspondence: Eucilene A. Santana-Porto, Ministry of Health—Secretariat Surveillance of Health, SCS quadra 04 Bloco A Edifício Central 6º andar, Brasília, Distrito Federal 70304000, Brazil; e-mail: eucilene.porto@saude.gov.br

Hepatitis C Virus in Blood Donors, Brazil

To the Editor: The Fundação de Hematologia e Hemoterapia do Amazonas is a public health service in Manaus, Brazil, that is responsible for serologic screening the serum of all blood donations in the region. In the state of Amazon, 9.0% of donated blood is discarded on the basis of serologic findings; discarding because of hepatitis C virus (HCV) antibodies declined from 1.25% in 1995 to 0.32% in 2007. The aim of this study was to characterize the serologic and molecular profile of HCV-antibody-positive blood donors from the Fundação de Hematologia e Hemoterapia do Amazonas.

For the study, 154 donors were selected from a routine database of voluntary blood donors who had donated from September 2005 through April 2007 (82,851 donations). Fresh plasma samples were sent to the laboratory in Manaus through the usual transportation systems for regular donations; i.e., samples from 27 cities are transported by air for ≈2 hours, and samples from 21 localities are transported by boat or road, all under refrigerated conditions.

An in-house standardized nested-PCR was used to detect HCV RNA (*I*). Genotype assignment was based on type-specific motifs on the sequenced amplicons delimited by primers HC11/HC18 from the 5' untranslated region

(2). Viral load was determined by commercial assay (HCV Monitor, Roche Molecular Systems, Inc., Branchburg, NJ, USA).

An association was observed between HCV RNA and donor age; the same trend was seen in the first-time blood donor group. Associations between HCV-RNA detection and gender ($p = 0.875$) and place of donation ($p = 0.989$) were not significant. Using 18–25 years of age as the reference group, we found that odds ratios (ORs) for having HCV viremia were higher for those 45–55 years of age (OR 8.19, $p < 0.001$) and 35–45 years of age (OR 3.49, $p = 0.003$).

We observed increasing rates of RNA detection according to the signal-to-cutoff (S/CO) ratio. However, some donors had a weak S/CO ratio (between 1 and 2) with positive nested-PCR tests (Figure). Although adopting an S/CO ratio as a criterion for referring for further testing by recombinant immunoblot assay (RIBA) has been advocated by some groups (3), our data show that this criterion may be misleading and would deny a confirmatory diagnosis by giving false-negative results for many persons.

A total of 113 samples were analyzed by RIBA; among 48 RIBA-reactive samples, 9 (18.8%) were negative for HCV RNA in plasma. However, because PCR results may sometimes be negative for persons who are actually infected, a single negative PCR result should not be relied on as evidence that virus has cleared from plasma. Such patients must be observed for years before they may be declared cured (4).

Among 97 RIBA-positive or -indeterminate samples, viral load was detectable in only 33 samples: 27 (81.8%) RIBA-positive samples and 6 (18.2%) RIBA-indeterminate samples. Only HCV genotypes 1 (87.1%) and 3 (12.9%) were found. Geographic distribution shows genotypes 1 and 3 in Manaus and only genotype 1 in other Amazon cities. This genotype

geographic distribution is similar to that found for many Brazilian cities and Eastern countries and may reflect the route of HCV introduction into the Amazon; the virus was probably brought to the Amazon region by European immigrants and blood-derived medicines imported to Brazil. This hypothesis is corroborated by the finding of genotype 3 exclusively in Manaus, suggesting that this city is the point of arrival of HCV and that new strains were disseminated from Manaus to inner localities. Historical reconstruction of HCV in Amazon could be attempted by using these isolates as well as others from hepatitis patients in the region, including genotype 2 (5).

We found a higher-than-expected rate of 50% for indeterminate immunoblot results among samples that were HCV-RNA positive by nested PCR. The presence of HCV RNA in plasma samples from 70%–75% of blood donors with indeterminate immunoblot results has also been reported by other groups in Brazil (6,7); however, in contrast, other investigators have reported RNA prevalence in such samples to be $\approx 2.5\%$ (1,8). Indeterminate RIBA test results can indicate seroconversion or seroreversion or, occasionally, a chronic infection when RNA HCV is detected in plasma

(9,10). To provide better understanding of the meaning of these indeterminate results, ongoing follow-up studies are examining the immune status of these persons.

Our data offer insights for counseling of donors who have repeatedly HCV-reactive results. We suggest that Amazon region blood banks screen by enzyme immunoassay and use molecular testing as the first supplemental test and that immunoblot be applied to the remaining HCV-RNA nonreactive samples to distinguish between true and false anti-HCV carriers. This new algorithm would save considerable resources currently spent on immunoblot-indeterminate persons in addition to HCV-RNA reactive persons who do not require further testing for confirmation. Moreover, according to current policy, those with false-positive results may be reinstated as donors if they have negative retesting results after 6 months.

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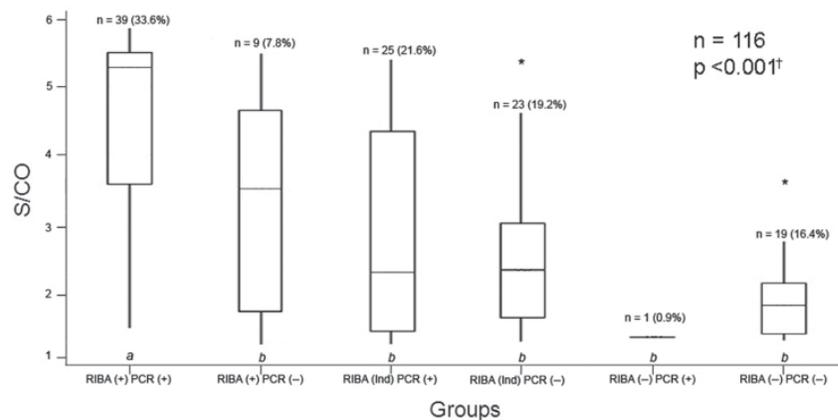


Figure. Distribution of hepatitis C virus (HCV) enzyme immunoassay signal-to-cutoff (S/CO) values by recombinant immunoblot assay (RIBA) interpretations among HCV-RNA-positive [PCR (+)] and HCV-RNA-negative [PCR (-)] donated blood samples. Group a differs statistically from all groups b with 95% confidence intervals. The Mann-Whitney test was used to compare the 2 groups. (+), positive; (-), negative; (Ind), indeterminate. *S/CO values outside interquartile intervals; †Kruskal-Wallis test.

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**Kátia Luz Torres,
Adriana Malheiro,
Adriana Tateno,
Tatiane Amabile de Lima,
Laura Patricia Viana Maia,
João Paulo Diniz Pimentel,
Márcia Poinho Encarnação de
Morais, Christiane
Santana de Melo Usui,
Flavia de Oliveira Braga,
Igor Araújo Ferreira Silva,
Felicien Vasquez,
and José Eduardo Levi**

Author affiliations: Fundação de Hematologia e Hemoterapia do Amazonas, Manaus, Amazon, Brazil (K.L. Torres, A. Malheiro, T.A. de Lima, L.P.V. Maia, J.P.D. Pimentel, M.P.E. de Moraes, C.S. de Melo Usui, F. Vasquez); Universidade Federal do Amazonas, Manaus (A. Malheiro, F. de Oliveira Braga, I.A.F. Silva); and Instituto de Medicina Tropical de São Paulo, São Paulo, Brazil (J.E. Levi)

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References

- Wendel S, Levi JE, Takaoka DT, Silva IC, Castro JP, Torezan-Filho MA, et al. Primary screening of blood donors by NAT testing for HCV-RNA: development of an "in-house" method and results. *Rev Inst Med Trop Sao Paulo.* 2007;49:177–85. DOI: 10.1590/S0036-46652007000300008
- Smith DB, Mellor J, Jarvis LM, Davidson F, Kolberg J, Urdea M, et al. Variation of the hepatic C virus 5' non-coding region: implications for secondary structure, virus detection and typing. *J Gen Virol.* 1995;76:1749–61. DOI: 10.1099/0022-1317-76-7-1749
- Barreto AMEC, Takei K, Sabino EC, Bellesa MAO, Salles NA, Barreto CC. Cost-effective analysis of different algorithms for the diagnosis of hepatitis C virus infection. *Braz J Med Biol Res.* 2008;41:126–34.
- Kleinman SH, Stramer SL, Brodsky JP, Caglioti S, Busch MP. Integration of nucleic acid amplification test result into hepatitis C virus supplemental serologic testing algorithms: implications for donor counseling and revision of existing algorithms. *Transfusion.* 2006;46:695–702. DOI: 10.1111/j.1537-2995.2006.00787.x
- Campiotto S, Pinho JRR, Carrilho FJ, Da Silva LC, Souto FJD, Spinelli V, et al. Geographic distribution of hepatitis C virus genotypes in Brazil. *Braz J Med Biol Res.* 2005;38:41–9. DOI: 10.1590/S0100-879X2005000100007
- Gonçales NSL, Costa FF, Vassalo J, Gonçalves-JR FL. Diagnosis of hepatitis C virus in Brazilian blood donors using a reverse transcriptase nested polymerase chain reaction: comparison with enzyme immunoassay and recombinant protein immunoblot assay. *Rev Inst Med Trop Sao Paulo.* 2000;42:263–7. DOI: 10.1590/S0036-46652000000500005
- Amorim RMS, Oliveira CP, Wyant PS, Cerqueira DM, Câmara GNL, Flores LS, et al. Hepatitis C virus genotype in blood donors from the Federal District, central Brazil. *Mem Inst Oswaldo Cruz.* 2004;99:895–7. DOI: 10.1590/S0074-02762004000800019
- Andrade AFB, Oliveira-Silva M, Silva SG, Motta II, Bonvicino CR. Seroprevalence of hepatitis B and C virus markers among blood donors in Rio de Janeiro, Brazil, 1998–2005. *Mem Inst Oswaldo Cruz.* 2006;101:673–6.
- Lefrère JJ, Girot R, Lefrère F, Guillaume N, Lerable J, Le Marrec N, et al. Complete or partial seroreversion in immunocompetent individuals after self-limited HCV infection: consequences for transfusion. *Transfusion.* 2004;44:343–8. DOI: 10.1111/j.1537-2995.2004.00656.x
- Bernardin F, Tobler L, Walsh I, Williams JD, Busch M, Delwart E. Clearance of hepatitis C virus RNA from the peripheral blood mononuclear cells of blood donors who spontaneously or therapeutically control their plasma viremia. *Hepatology.* 2008;47:1446–52. DOI: 10.1002/hep.22184

Address for correspondence: Kátia Luz Torres, Research Division, Fundação de Hematologia e Hemoterapia do Amazonas, Av Constantino Nery, 4397-Manaus, AM, Brazil; email: katia.torres@hemoam.am.gov.br

Leishmaniasis in Chaparé, Bolivia

To the Editor: In Bolivia, most cases of leishmaniasis are caused by *Leishmania (Viannia) braziliensis* (I). The parasite is transmitted zoonotically by several sandfly species and, when transmitted to humans, may cause cutaneous leishmaniasis (CL), and potentially, mucosal leishmaniasis (ML) (2).

Data on the prevalence and effects of CL in Bolivia have been scarce, even though anecdotal and official reports indicate a dramatic increase in the number of human CL cases in Bolivia in the past decade (1,3). Also, although CL was originally a sylvatic disease in Bolivia, some evidence indicates that the transmission cycle has adapted to the peridomestic habitat. However, this evidence is largely based on individual case reports. No information is available on parasite species, vectors, and reservoirs in such a peridomestic transmission cycle.

A preliminary study to guide future research focus and assist in immediate leishmaniasis prevention and control policy decision making is underway in Isiboro-Secure National Park, Chaparé, Bolivia. Our objectives were to collect data on the prevalence of leishmaniasis in that area and evidence for peridomestic *Leishmania* transmission.

A survey was carried out during April–July 2007 in 2 communities in Isiboro-Secure National Park, San Gabriel (16°40'31"S and 65°37'38"W) and San Julian (16°41'59"S and 65°38'10"W). These 2 communities were selected because of local knowledge of disease in the community, their moderate degree of urbanization (i.e., ≈50% of the communities' houses are clustered around the main access road), and the accessibility of the sites to the field team. In this area, CL is transmitted from April through October.