Hepatitis B virus, syphilis, and HIV seroprevalence in pregnant women and their male partners from six indigenous populations of the Peruvian Amazon Basin, 2007–2008

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SUMMARY

Objective: To assess the seroprevalence of hepatitis B virus (HBV), syphilis, and HIV and associated risk factors in pregnant women and their male partners from six indigenous populations of the Peruvian Amazon Basin.

Methods: A cross-sectional study was performed in six indigenous populations from the Peruvian Amazon Basin. Blood samples were obtained and tested for HBV (antibodies to the hepatitis B core antigen (anti-HBc) and hepatitis B surface antigen (HBsAg)), for syphilis (rapid plasma reagin and microhemagglutination assay for Treponema pallidum antibodies), and for HIV (ELISA and indirect immunofluorescence test). A survey was also performed to identify associated risk factors.

Results: One thousand two hundred and fifty-one pregnant women and 778 male partners were enrolled in the study. The seroprevalence of anti-HBc in pregnant women was 42.06% (95% confidence interval (CI) 39.28–44.85%) and in their male partners was 54.09% (95% CI 50.32–57.86%). The seroprevalence of HBsAg in pregnant women was 2.11% (95% CI 0.78–3.44%) and in their male partners was 3.98% (95% CI 1.87–6.08%). The seroprevalence of syphilis in pregnant women was 1.60% (95% CI 0.86–2.33%) and in their male partners was 2.44% (95% CI 1.22–3.66%). HIV seroprevalence in pregnant women was 0.16% (95% CI 0.02–0.58%) and in their male partners was 0.29% (95% CI 0.04–1.03%). Sexual risk factors were strongly related to blood markers of syphilis and HBV.

Conclusions: Hepatitis B was found to be hyperendemic and strongly related to sexual factors, suggesting an important sexual component in the transmission of the disease in the populations studied. Syphilis was found to have an endemicity in pregnant women above the national level and this may be indicative of high mother-to-child transmission. HIV has started to show its presence in indigenous populations of the Amazon Basin and the results suggest the epidemic is concentrated.

1. Introduction

HIV, *Treponema pallidum* (the etiologic agent of syphilis), and hepatitis B virus (HBV) produce three infectious diseases, and their main routes of transmission are sexual intercourse, parenteral transmission, and vertical (mother-to-child) transmission. Horizontal transmission of HBV is also very important, especially in areas of high prevalence.[1–3]

HBV is the major cause of chronic infection in the world, with more than 350 million people chronically infected. The World Health Organization (WHO) estimates that approximately 600 000 people die annually as a result of HBV infections, including hepatocellular carcinoma and cirrhosis.[3] In Latin America, the endemicity of HBV varies between countries with a seroprevalence of antibodies to the hepatitis B core antigen (anti-HBc) in urban populations ranging from 0.6% in Chile to 21.4% in the Dominican Republic.[4] In South America, 100 000 people are infected each year, and estimations show that it is responsible for 25–67% of all chronic hepatitis B cases and 10–70% of all primary hepatocellular carcinoma cases.[5] Even though estimations show that the level of HBV endemicity in our region is low, these same estimations point to the Amazon Basin, shared by Bolivia, Brazil, Colombia, Ecuador, French Guyana, Guyana, Peru, Suriname, and Venezuela, as an area of high endemicity.[4,5]

Estimations made for year 2009 show that 2.6 (2.3–2.8) million people became infected with HIV, with 92 000 of the infections occurring in Central and South America, accounting for a

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prevalence of 0.5%; 1.4 million people were living with HIV in these areas. Over the last 20 years of the epidemic, the seroprevalence of HIV has remained stable in the countries of the region, with a tendency towards a rise in the number of people living with HIV due to increased access to highly active antiretroviral therapy (HAART).[2] In Peru, the HIV epidemic is considered to be concentrated. Evidence shows the seroprevalence of HIV to have remained consistently under 0.5% in the general population but over 5% in high-risk populations.[1,6] HIV cases are mostly located in urban areas of the coast and jungle (Amazon) geographic regions,[1,7] and are particularly concentrated in high-risk groups such as men who have sex with men (MSM).[1,8,9]

The prevalence of syphilis in Peru is considered to be 0.5% in men and 1.0% in women,[10] although a large study in Lima City found a seroprevalence of 1.6% in pregnant women.[6] Nevertheless, available data principally reflect the situation of HIV and syphilis in urban areas, where most cases are detected. In small populations, even a small number of cases might pose a serious public health problem, especially if cases occur in highly vulnerable populations.

Forty-two indigenous populations live in the Peruvian Amazon jungle, each with its own cultural characteristics and with its own social and economic structures. Within these populations, health is considered in a holistic way, because it expresses dynamic relationships between the individual and the collective, the natural and social environments. These populations are highly vulnerable to health problems, due to a lack of access to health services, mainly because of an inadequate health offer, the disarticulation of their knowledge of traditional/natural medicine, and to the small size and dispersion of their populations.[11–15] Scientific investigations carried out in indigenous populations of the Amazon have found a high seroprevalence of HBV in Peru.[16–18] Nevertheless, none of these studies have identified risk factors associated with HBV or with the seroprevalence in specific population groups. Also, reports of AIDS cases in Amazon indigenous populations began to appear in 2004. In 2007, a high seroprevalence was reported in an indigenous population located in a peri-urban area of the district of Yurimaguas in the province of Alto Amazonas, in Loreto Region.[19] Nevertheless the study comprised a small indigenous population located in a peri-urban area, and specific population groups were not addressed.

This situation is occurring within an international context in which the Pan American Health Organization and the WHO are promoting an initiative to eliminate the vertical transmission of HIV and syphilis in the Americas and the world.[20] Studying indigenous pregnant women and their male partners would allow sexually active population groups to be examined, which is key to understanding both the sexual and vertical transmission of HIV, syphilis, and HBV. Therefore the objective of this study was to identify the seroprevalence and factors associated with HBV, syphilis, and HIV in pregnant women and their male partners from six indigenous populations of the Peruvian Amazon Basin.

2. Methods

A cross-sectional study was performed between July 16, 2007 and June 16, 2008. Six of the 42 indigenous populations that reside in the Peruvian Amazon were selected by means of convenience sampling regarding their ease of access, population size, and area of residence: Kandozi, Shapra, Shiwilo (Chayahuita), Matsigenka, Ashaninka, and Shipibo Konibo. They are located in the political regions of Loreto, Ucayali, Cusco, Junín, and Pasco. The study population constituted pregnant women and their current sexual partners from the six indigenous populations selected.

In Loreto Region, all the native communities of the Kandozi, Shapra, and Shiwilo populations were included in the study. In Ucayali, all the communities of the Shipibo Konibo population

were included (although there are some communities in Loreto, the main numbers are located in Ucayali). In Cusco, all Matsigenka communities were included. In Junín and Pasco, all Ashaninka communities were included. The Ashaninka make up the largest indigenous population in the Amazon jungle, and as well as living in Junín and Pasco, they also reside in Huánuco and Ucayali, and a small number reside in Loreto. A communal census was performed in each indigenous community in order to identify all pregnant women and their male sexual partners, and all were invited to participate in the study. All pregnant women or their male partners who did not give their informed consent were excluded from the study, as well as those without the capacity to provide this.

HIV infection was detected using a fourth-generation ELISA as the screening test and an indirect immunofluorescence test (IIFT) as the confirmatory test. Two tests were performed for the detection of syphilis, the rapid plasma reagin (RPR) test as the screening test and a microhemagglutination assay for *Treponema pallidum* antibodies (MHA-TP) as the confirmatory test. Finally, ELISA tests for anti-HBc and hepatitis B surface antigen (HBsAg) were performed. Blood samples were obtained by venous puncture in the forearm, drawing a quantity of 7 cm^3. Serum samples were obtained directly in the field and were maintained in a cold chain (2–8 °C) until arrival at the regional laboratories at which the ELISA tests for HIV and anti-HBc, RPR, and MHA-TP tests were performed. Samples positive for HIV were sent to the National Institute of Health for confirmation by IIFT. Also, samples positive for anti-HBc were sent to the Regional Laboratory of the South Lima Health Department where samples were also tested using an ELISA for HBsAg. Even though kits from different producers were used at each laboratory, all complied with the standardized technical specifications provided by the National Institute of Health (http://www.ins.gob.pe/fichasins/default.asp).

A survey was also administered to identify risk behaviors, following the Joint United Nations Programme on HIV/AIDS (UNAIDS) recommendations. The survey was reviewed by theme experts and by indigenous leaders during a workshop with the participation of the Asociación Interétnica de Desarrollo de la Selva Peruana (AIDESEP), which is the national organization of the indigenous populations residing in the Peruvian Amazon.

Hepatitis B endemicity was considered to be low when chronic infection was less than 2% or anti-HBc less than 15%; intermediate when chronic infection was 2–7% or anti-HBc prevalence ranged from 15% to 40%; and high when chronic infection was over 7% or anti-HBc prevalence was over 40%.

Syphilis seroprevalence was considered to be high in pregnant women and their male partners if it was above the national mean estimations. The Action Plan for Congenital Syphilis Elimination states that to achieve the objective of eliminating congenital syphilis, more than 95% of pregnant women with syphilis should be identified and treated, and that syphilis seroprevalence during pregnancy should be reduced to below 1%.[21]

HIV seroprevalence was evaluated using the World Bank/ UNAIDS/WHO criteria for a low level epidemic, concentrated epidemic, and generalized epidemic.

Study teams were formed in all political regions. These teams consisted of health professionals from the departments of epidemiology and health services and the laboratories, who worked in conjunction with the organizations representing the indigenous populations participating in the study. Workshops were conducted with these teams and the organizations representing the indigenous populations to plan field activities. Local teams were strengthened by infirmary professionals and intercultural facilitators of both sexes.

Local teams visited the study area three times. The first visit was to present the study to the local authorities in all communities of the selected populations, and to obtain their authorization to begin the study. Also a census was conducted to identify all pregnant

women and their partners. All pregnant women and their partners identified in this census were invited to join the study. The second visit was done to obtain informed consent from the participants, to collect the serum samples, and to carry out the survey. Prior to obtaining serum samples, pretest counseling was done following the norms of the National Strategy for HIV Control and Prevention. The third visit was done to deliver the test results individually and in confidentiality. As part of this process, personal post-test counseling was delivered.

The statistical analysis was performed using SPSS v. 13 (IBM Corp., NY, USA) and Epidat v. 3.1 (Dirección Xeral de Saúde Pública, Consellería de Sanidade – Xunta de Galicia, España; Organización Panamericana de la Salud, Washington DC, USA). Confidence intervals (95% CI) were calculated for seroprevalence data. Odds ratios (OR) and the Chi-square test were used to test statistical associations for qualitative variables; the Student's t-test and the Mann–Whitney U-test were used for quantitative variables. For the multivariate analysis, a binary logistic regression test was used, applying a manual entry method, in order to identify the risk factors associated with anti-HBc positivity. A total of 32 variables exploring socioeconomic status, sexual behavior and parity, and past history of sexually transmitted infections (STIs) and knowledge on prevention were first included in the analysis. Only those showing significance at the 5% level were kept.

The study project was reviewed and approved by an ethics committee. For this study, authorization was solicited and obtained from the indigenous population authorities at the national, regional, and local levels. Also, individual written consent was obtained from all participants before applying the survey and obtaining the blood samples. Pre-test and post-test counseling was delivered to all participants, and the results of all their tests were delivered to them in privacy, according to the State Law CONTRASIDA 26626 of our country. Action was taken to ensure that all patients who tested positive had access to treatment provided by the government health services. All processes were carried out with the indigenous authorities present in order to conduct the study in a proper cultural way. Also, male and female translators accompanied the survey teams.

3. Results

A total of 1633 pregnant women were identified, of whom 1251 agreed to participate in the study. Seven hundred and seventy-eight of their male partners also agreed to participate. All serum samples from the pregnant women were tested for HIV and syphilis, and 1241 for HBV. For male partners, 697 serum samples were tested for HIV, syphilis, and HBV.

Table 1 shows the seroprevalence results. Two HIV cases were detected in pregnant women and two in male partners. One positive male was the sexual partner of an HIV-positive pregnant

woman. The other positive male was the partner of an HIV-negative pregnant woman. The male partner of the other HIV-positive pregnant women did not agree to participate in the study.

HBV anti-HBc seroprevalence results for each indigenous population included in the study are presented in Table 2.

The mean age of the pregnant women was 25.1 years (standard deviation (SD) 7.1 years) and of their male partners was 29.8 years (SD 8.6 years). The percentage of pregnant women without a formal education was 10.6% (129/1215) and for their male partners this was 5.5% (40/730; $p < 0.001$). Pregnant women achieved a lower level of education compared to males ($p < 0.001$), with a higher percentage attaining basic/elementary education (61.2% vs. 46.6%) and a lower percentage attaining a high school education (27.2% vs. 42.2%) or college education (1.0% vs. 5.7%).

The main economic activities of the indigenous pregnant women were housework (55.7%; 693/1244) and subsistence agriculture (41.6%; 517/1244). The main male economic activity was agriculture (87.7%; 682/778). Also, male partners had greater access to other productive activities like working in the education sector (3.0% vs. 0.2%) and in local government (1.1% vs. 0.1%).

The mean number of pregnancies was 3.9 (SD 2.7) and the mean number of living children was 2.7 (SD 2.3). Pregnant women were younger at sexual initiation when compared to their male partners (14.9 years, SD 2.4 vs. 16.1 years, SD 2.7; $p < 0.001$). The mean number of sexual partners during their lifetime was lower for pregnant women compared to their male partners (2.3 vs. 8.6, $p < 0.001$), as well as the mean number of sexual partners other than their official sexual partner in the 12 months prior to the survey (0.16 vs. 1.72, $p < 0.001$) and in the 3 months prior to the survey (0.10 vs. 1.85, $p < 0.001$). Fewer pregnant women than their male partners reported having had sexual contact with people other than their partner during the 12-month period (10.6% vs. 27.6%, $p < 0.001$) and 3-month period (6.5% vs. 14.5%, $p < 0.001$) before the survey. More than a fifth (21.4%; 250/1168) of the pregnant women believed their partners to have been unfaithful to them: with other indigenous women (87.2%), female sex workers (7.6%), or MSM (0.8%); 4.4% were not able to identify with whom their partner had been unfaithful. Of the pregnant women, 12.6% (158/1251) reported having received treatment for STIs; 9.4% (118/592) of their male partners reported treatment for STIs. Even though 41.1% (502/1221) of the pregnant women and 70.6% (480/680) of their male partners recognized that using condoms is an effective method to prevent the transmission of STIs, only 13.8% (170/1228) and 28.7% (197/687), respectively, reported having used one at some time. Further, only 12.0% (76/634) of the male partners reported using one with the pregnant women. Only 2.6% (32/1228) of the pregnant women and 5.5% (38/687) of male partners reported always using a condom.

The assessment of STI transmission knowledge among 687 pregnant women and 568 male partners showed that 76.4% and 79.0%, respectively, identified sexual contact as an important

Table 1
Seroprevalence of serum markers for HIV, syphilis, and hepatitis B in pregnant women and their male partners from six indigenous Amazon populations in Peru

Variable	Pregnant women			Male partners			p-Value[a]
	n/N	Prevalence (%)	95% CI	*n/N*	Prevalence (%)	95% CI	
HIV							
Elisa/IFFT	2/1251	0.16	0.02–0.58	2/697	0.29	0.04–1.03	0.554[b]
Syphilis							
RPR/MHA-TP	20/1251	1.60	0.86–2.33	17/697	2.44	1.22–3.66	0.202
Hepatitis B							
Anti-HBc	522/1241	42.06	39.28–44.85	377/697	54.09	50.32–57.86	0.002
HBsAg	11/522	2.11	0.78–3.44	15/377	3.98	1.87–6.08	0.109

95% CI, 95% confidence interval; IFFT, indirect immunofluorescence test; RPR, rapid plasma reagin test; MHA-TP, microhemagglutination assay for *Treponema pallidum* antibodies; anti-HBc, antibodies to the hepatitis B core antigen; HBsAg, hepatitis B surface antigen.
[a] Chi-square test.
[b] Fisher's exact test.

Table 2
Anti-HBc seroprevalence by indigenous population

Variable	Pregnant women		Male partners	
	n	Prevalence (%)	n	Prevalence (%)
Populations	n = 1237		n = 663	
Ashaninka	212/728	29.1	147/351	41.9
Kandozi	18/18	100	12/12	100
Shapra	36/36	100	21/21	100
Shiwilo	35/94	37.2	38/69	55.1
Matsigenka	84/180	46.7	47/87	54.0
Shipibo Konibo	130/176	73.9	88/109	80.7
Mestiza[*]	4/5	80	11/14	78.6

Anti-HBc, antibodies to the hepatitis B core antigen.
[*] Mestiza: non indigenous people married to indigenous men or women.

transmission route. However, only 15.9% of pregnant women and 17.3% of male partners identified having multiple sexual partners as a risk factor for STIs. Even less, 6.4% and 7.2% of pregnant women and male partners, respectively, recognized sexual contact with strangers as a risk factor for STIs. Having one sexual partner was the behavior most frequently recognized as a preventive measure against STIs: 64.0% (424/663) of pregnant women and 69.0% (387/561) of their male partners identified this correctly. Condom use was the second most frequently identified preventive measure, reported by 22.8% (151/662) of pregnant women and 23.9% (134/561) of their male partners.

Due to the limited number of HIV cases it was not possible to identify factors associated with this infection. A similar situation occurred with cases positive for HBsAg. Bivariate and multivariate analysis was performed for syphilis and anti-HBc markers for both pregnant women and their male partners.

3.1. Factors associated with syphilis

Logistic regression analysis showed that syphilis positivity in pregnant women was associated only with syphilis in their male partners (Table 3). Bivariate analysis showed that the risk factors of sexual contact with strangers (OR 5.23, 95% CI 2.10–13.02, $p = 0.001$) and a vaginal secretion episode during their lifetime (OR 2.90, 95% CI 1.11–7.60, $p = 0.030$) were also associated with syphilis positivity in pregnant women; knowledge of any preventive measure (OR 0.09, 95% CI 0.01–0.79, $p = 0.027$) played a protective role. Male partner risk factors associated with syphilis

in pregnant women were sexual contact with other women (OR 2.50, 95% CI 1.01–6.17, $p = 0.053$), genital discomfort episode during lifetime (OR 4.80, 95% CI 1.19–19.40, $p = 0.024$), and genital itching episode during lifetime (OR 8.53, 95% CI 1.63–44.69, $p = 0.038$).

Logistic regression analysis showed that syphilis in male partners was associated with syphilis positivity in pregnant women, and with sexual contact with MSM, having ≥11 sexual partners in their lifetime, and being >39 years of age (Table 4). Bivariate analysis also showed other variables to be associated with syphilis in male partners: pregnant women factors were age over 25 years (OR 3.05, 95% CI 1.06–8.76, $p = 0.030$), working as an artisan (OR 5.88, 95% CI 1.23–28.01, $p = 0.062$), sexual contact with strangers (OR 6.22, 95% CI 2.29–163.87, $p = 0.001$), and vaginal secretion (OR 3.33, 95% CI 1.19–9.35, $p = 0.031$); male partner factors were sexual initiation with a woman (OR 0.09, 95% CI 0.02–0.46, $p = 0.022$), sexual initiation with an MSM (OR 14.04, 95% CI 1.38–142.43, $p = 0.095$), sexual contact with an MSM in the 12 months prior to the survey (OR 13.66, 95% CI 3.40–54.81, $p = 0.003$), genital discomfort (OR 6.44, 95% CI 1.99–20.86, $p = 0.001$), and having received treatment for an STI (OR 5.41, 95% CI 2.04–14.36, $p = 0.001$).

3.2. Factors associated with HBV

Logistic regression analysis showed that anti-HBc positivity in pregnant women was associated with age at pregnancy and age at sexual initiation <16 years in the women, and with anti-HBc positivity, sexual contact with MSM during lifetime, and condom use in their male partners (Table 5).

For the male partners, logistic regression analysis showed anti-HBc positivity was associated with anti-HBc positivity in the pregnant women and sexual initiation of the women before 16 years of age, and to male partner's age and sexual contact with MSM during their lifetime (Table 6).

4. Discussion

This is the largest study in the indigenous populations of the Amazon Basin in Peru to be published and the first to address pregnant women, their male partners, and their sexual habits.

HIV seroprevalence was low in both the pregnant women and their male partners, although it was higher in male partners than in

Table 3
Factors associated with syphilis in indigenous pregnant women

Model	Coefficient			Odds ratio		
	Point estimation	Standard error	p-Value	Point estimation	95% CI	
					Lower limit	Upper limit
Syphilis in male partner	2.32	0.83	0.005	10.19	2.01	51.55

95% CI, 95% confidence interval.

Table 4
Factors associated with syphilis in male partners of indigenous pregnant women

Model	Coefficient			Odds ratio		
	Point estimation	Standard error	p-Value	Point estimation	95% CI	
					Lower limit	Upper limit
Syphilis in pregnant woman	2.47	0.90	0.006	11.85	2.03	69.28
Male sexual contact with MSM in last 12 months	2.56	0.79	0.001	12.93	2.77	60.43
Male with ≥11 sexual partners during lifetime	1.26	0.59	0.033	3.52	1.11	11.18
Male aged ≥39 years	1.29	0.61	0.035	3.62	1.10	11.92

95% CI, 95% confidence interval; MSM, men who have sex with men.

Table 5
Factors associated with anti-HBc in indigenous pregnant women

Model	Coefficient			Odds ratio		
	Point estimation	Standard error	*p*-Value	Point estimation	95% CI	
					Lower limit	Upper limit
Woman's age at pregnancy	0.06	0.02	<0.001	1.06	1.03	1.09
Woman's age at sexual initiation <16 years	0.62	0.26	0.018	1.86	1.11	3.11
Male anti-HBc-positive	1.92	0.20	<0.001	6.85	4.60	10.18
Male sexual contact with MSM	1.18	0.47	0.012	3.24	1.29	8.12
Male condom use	−0.55	0.22	0.014	0.58	0.38	0.90

Anti-HBc, antibodies to the hepatitis B core antigen; 95% CI, 95% confidence interval; MSM, men who have sex with men.

Table 6
Factors associated with anti-HBc in male partners of indigenous pregnant women

Model	Coefficient			Odds ratio		
	Point estimation	Standard error	*p*-Value	Point estimation	95% CI	
					Lower limit	Upper limit
Anti-HBc in pregnant woman	1.80	0.22	0.000	6.07	3.97	9.28
Woman's age at sexual initiation <16 years	0.99	0.28	0.000	2.68	1.55	4.64
Male sexual contact with MSM	0.95	0.46	0.036	2.60	1.06	6.35
Male age	0.04	0.01	0.002	1.04	1.02	1.07

Anti-HBc, antibodies to the hepatitis B core antigen; 95% CI, 95% confidence interval; MSM, men who have sex with men.

pregnant women (0.29% vs. 0.16%); nevertheless the difference was not statistically significant. Also, the small number of cases limited the possibility of determining associations. HIV seroprevalence reports in indigenous populations are uncommon. Zavaleta et al. found an HIV seroprevalence of 7.5% in 80 adults of a native community of the Chayahuita (Shiwilo) indigenous population living in a peri-urban area in Loreto Region.[19] In 2006 Bartlett et al. found two HIV cases in 282 people from four indigenous communities of the same Chayahuita population, but located at a greater distance from the urban areas.[22] Our study, with a larger sample, from communities located 4–48 h by speedboat from urban areas, found a lower seroprevalence. This lower seroprevalence is closer to that reported from Brazil by Machado et al., who found an HIV seroprevalence of 0.6% in the Tiriyo tribe of the Brazilian Amazon Region.[23]

Some other studies involving non-indigenous Peruvian pregnant women have been published. In 1996–1997 Alarcon et al. found an HIV seroprevalence of 0.5% in pregnant women from Lima City.[6] In 2002 and 2005 the Dirección General de Epidemiología (DGE) found an HIV seroprevalence of 0.21% (95% CI 0.13–0.30%) and 0.23% (95% CI 0.10–0.50%) in pregnant women from selected urban areas of the country (DGE, unpublished data). Also, in 2003–2004 Clark et al. found an HIV seroprevalence of 0.8% (95% CI 0.5–1.4%) in male partners in four cities of Peru.[24] Similar to those studies, the HIV seroprevalence found in our study was under 1% and was higher in males than in pregnant women. These results suggest that the HIV epidemic in the studied populations may be at the low or concentrated level;[1] nevertheless it is still necessary to identify the HIV prevalence in high-risk groups to determine the epidemic level. In the study of Zavaleta et al. up to 44.7% of men admitted sexual contact with other men. Also three of the four self-identified MSM in that community were infected with HIV.[19] Bartlett et al. found two HIV cases in 135 men; of the 126 men who answered the question, 39.7% admitted having homosexual contact at least once. Both positive cases were in this latter group.[22] In 2007, in another study in the Chayahuita population of the same area, the DGE found three HIV cases in 44 men (6.4%) who admitted having at least one homosexual contact during their lifetime (DGE, unpublished data). In the present study, 47 of 756 men (6.2%) admitted having homosexual contact at least once, but

none were HIV-positive. It is worth mentioning that while conducting the study, HIV testing was offered to 15 indigenous MSM who were not in the study population; five of them (33.3%) were found to be HIV-positive. Even though a larger study with a better structured sample is needed to reach a conclusion on HIV prevalence in indigenous MSM, the available evidence suggests that the HIV epidemic may be at a concentrated level in the Amazon indigenous population.

The seroprevalence of syphilis was 1.60% in pregnant women and 2.44% in their male partners, but the difference was not statistically significant. The seroprevalence of syphilis in pregnant women of 1.60% found in our study is above 1% and may be indicative of a high risk of mother-to-child transmission.[21] Seroprevalences were higher than national estimates,[10,21] but lower than other reports in indigenous populations. Zavaleta et al. found a syphilis prevalence of 6.3% in a small sample of adult individuals of the Chayahuita (Shiwilo) indigenous population, whose males reported having a high frequency of homosexual contact (44.1%).[19] Bartlett et al. found a seroprevalence of syphilis in males of 3.7% for whom self-reported MSM activity was also high (39.7%).[22] The difference with our study results may be explained by the frequency of MSM activity and the ease of access to urban areas. In the present study, when analyzing the factors associated with syphilis in males, MSM contact in the last 12 months was strongly related, but the frequency of MSM contact was significantly lower than in the studies of Zavaleta et al. and Bartlett et al.: 6.2% during lifetime and 2.1% in the last 12 months. Also the frequency of MSM contact varied between the indigenous groups included in the study. MSM activity was highest in Shipibo Konibo males with 29.8% during lifetime and 11.7% in the last 12 months; also 14 of 17 confirmed cases of syphilis found in the study belonged to this indigenous group, with a seroprevalence of 12.4%. The other three cases were Matsigenka males, with a seroprevalence of 3.3%. The association between syphilis and MSM activity has also been observed in urban areas of the Peruvian coast by Snowden et al.[10] These results suggest that sexual behavior and health risks associated with it may vary importantly between different indigenous populations and even between communities from the same groups. The distance to urban areas may be another factor related to differences in syphilis prevalence. Bartlett et al.

and Zavaleta et al. found similar MSM behavior in their studies, but the seroprevalence of syphilis was almost halved in the study of Bartlett et al. whose sample was obtained from indigenous communities located at a greater distance from urban areas than that included in the Zavaleta et al. study. This is also consistent with our study in which the communities of the six indigenous populations were located 4–48 h by small speedboat from urban areas. The distance to urban areas may be working as an indicator of access to these areas.

Syphilis seroprevalence in both groups was also higher when compared to the results of other studies of pregnant women and their partners in urban areas. A study performed by the DGE in 2002 found syphilis seroprevalences in pregnant women of 0.6% in urban areas of the coast natural region and 0.9% in urban areas of the jungle (Amazon) natural region (DGE, unpublished data). In male partners of pregnant women, Clark et al. found a syphilis seroprevalence of 1.6% (95% CI 1.0–2.2%) in urban areas of the coast region,[24] which is lower than the seroprevalence found in this study. The higher prevalence in indigenous pregnant women and male partners may also be the result of poor access to health services and treatment when compared to those living in urban areas.

Other risk factors associated with syphilis in males were ≥11 sexual partners during their lifetime and male age ≥39 years. In agreement with this Clark et al. found that syphilis infection was associated with ≥10 sexual partners during their lifetime in urban areas of the Peruvian coast.[24] For pregnant women the only variable associated with syphilis in the logistic regression analysis was the presence of syphilis in their male partners. Probably the small number of positive cases may have limited the power of the study to find other associated factors. All these results may indicate that the syphilis infection is being introduced into the studied indigenous populations by indigenous males who make contact with high-risk groups in urban areas.

The study results were also indicative of a high prevalence of HBV in the indigenous groups studied. Even though HBsAg seroprevalences in pregnant women and their male partners were under 8%, the anti-HBc seroprevalence was over 40% (42.06% and 54.09%, respectively), a situation that confirms the results of other studies in similar populations.[4,5,16–18,25–29]

Although it is well known that the main route of transmission of HBV in areas of high prevalence is perinatal, from mother to child, or from person to person during early childhood,[3] our study identified sexual characteristics associated with anti-HBc in pregnant women and in their male partners, suggesting that sexual transmission could play an important role even in populations where endemicity is high. Of the five factors associated with anti-HBc in pregnant women, four were related to sexual habits, like sexual initiation before 16 years of age for the women, male partner positive for anti-HBc, male partner with homosexual contact, and male partner condom use. Age was the other variable associated with anti-HBc in pregnant women, showing an increment of 6% in the probability of being positive for each year of age. For the male partners, anti-HBc positivity was associated with the presence of anti-HBc in the pregnant women and women's sexual initiation before 16 years of age, and to male sexual contact with MSM. Age of the male partners was also related to anti-HBc positivity, with an increment of 4% for each year of age added.

Other studies in Amazon indigenous populations from other countries have also identified low or intermediate levels of HBsAg, suggesting that apart from vertical transmission, sexual transmission and household cultural habits –like feeding small children with food previously chewed by the mothers – may play an important role in the transmission of HBV in indigenous populations.[25–29] El Khouri et al. found similar results in non-indigenous populations from urban and rural areas of the Amazon, with anti-HBc and HBsAg seroprevalences of 40.74% and 2.88%,

respectively.[30] de Paula et al., also in non-indigenous populations from urban and rural areas of the Amazon in Brazil, found an anti-HBc seroprevalence of 66.2%, identifying age and sexual activity as related factors.[31] All these results suggest that in populations of high endemicity and low HBsAg seroprevalence, sexual transmission may be the transmission route responsible for the majority of the cases in the community.

The anti-HBc seroprevalence found in our study confirms the wide dissemination of HBV in the indigenous populations living in the Peruvian Amazon Basin, which represents a high risk for hepatitis B chronic disease in an area characterized by a weak health system and extreme poverty. Previous studies in Peruvian Amazon indigenous populations have identified hepatitis B as a main cause of mortality and potential years of life lost (PYLL).[11–15] Therefore, careful assessment of the hepatitis B situation by the health authorities is needed in these populations to implement appropriate control measures.

Our study has limitations. Different ELISA tests were used; however standardized criteria for sensitivity and specificity recommended by the National Institute of Health were used for their acquisition. Also the tests were performed at each regional laboratory, which may have introduced a bias in the measurement of the main variables that is difficult to estimate. Nevertheless, personnel in these laboratories have been trained in performing the tests used in this study. The sampling method used limited the ability to represent the study population, especially since only some of the Ashaninka and Shipibo Konibo pregnant women and male partners were included. The number of male partners who agreed to participate in the study was smaller than expected, which may have introduced a selection bias and may have affected the power of the study to identify associations and to make proper estimations for this group. Regrettably no data were obtained from the missing male partners making it very difficult to estimate how their absence may have biased the study results. Face to face interviewing may also have introduced an information bias both in women and their male partners, even though they were interviewed separately and by people of the same sex; this may have produced an underreporting of culturally reproved behaviors. The number of pregnant women who answered the questions related to knowledge of transmission and prevention of the diseases was approximately half of the total number of surveyed women, which may have introduced a selection bias in these areas. When we analyzed the population that chose not to answer these questions, we found that 62.9% (338/537) had an incomplete elementary/primary education level or had attained no formal education level at all, while 60.2% (408/678) of the group that answered the questions had a complete elementary school or higher level of education, indicating that knowledge on these areas may be even lower than observed.

The results of our study express an elevated vulnerability of the indigenous populations to these diseases. In the case of the HIV epidemic this may lead to a faster progression than has been observed in the urban areas of the country where the epidemic has reached a stabilization point and is focused on groups with high-risk sexual habits.[1,2] The seroprevalence of syphilis in the indigenous pregnant women is indicative of a high congenital syphilis prevalence.[21,32] HBV has a wide distribution in the indigenous populations studied and may represent a serious risk for their survival, especially in the smallest groups, with a very important sexual-related transmission. To overcome this situation, prevention and control strategies should be developed and adapted to an intercultural framework; these should be based on successful experiences in similar populations.

Recently a new strategy to prevent and control HBV has been developed, providing treatment for HBV to the Kandozi and Shapra indigenous populations. This effort should be expanded to take in

HIV and syphilis prevention and control, and should be complemented with an intercultural prevention framework. These initiatives should be extended promptly to reach all the other indigenous populations in the Peruvian Amazon.

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