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Original Article

Serosurvey of anti-*treponema pallidum* (syphilis), anti-hepatitis C virus and anti-HIV antibodies in homeless persons of São Paulo city, southeastern Brazil

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ABSTRACT

Homeless persons have been considered as one of the most susceptible populations to sexually transmitted infections. In Brazil, these population experienced an increase of 140% from 2012 to 2020. Accordingly, the present study aimed to assess the seroprevalence of anti-*Treponema pallidum*, anti-HCV, anti-HIV antibodies, and the risk factors associated with homeless persons in a daytime attendance shelter of São Paulo city during the syphilis epidemic in Brazil. Blood samples of 116 volunteers and epidemiological data were conveniently collected in the shelter from June through August 2018. Detection of syphilis, HCV, and HIV antibodies was performed by chemiluminescent microparticle immunoassay (CMIA). CMIA-reagent samples for anti-*T. pallidum* antibodies were confirmed by Venereal Disease Research Laboratory (VDRL) non-treponemal test. VDRL non-reagent samples were confirmed by treponemal rapid immunochromatographic test. A rapid immunoblot assay confirmed seropositivity to HIV. Overall, anti-*T. pallidum* antibodies were observed in 29/116 (25.0%), anti-HCV antibodies in 4/116 (3.4%), and anti-HIV antibodies in 2/116 (1.7%) individuals, both co-infected with anti-*T. pallidum* antibodies. Associated risk factors for syphilis in homeless persons were being born or previously living in another city ($p = 0.043$) and becoming homeless due to family conflicts ($p = 0.035$). Besides homeless vulnerability, worldwide shortage of benzathine penicillin supply and increasing of syphilis testing

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access through rapid testing in primary health care services may have also impacted disease spreading at the time. The prevalence of syphilis found herein is the highest worldwide to date in this population.

Introduction

Homeless persons have been considered one of the most susceptible populations to sexually transmitted infections (STI) such as those caused by *Treponema pallidum*, hepatitis C virus (HCV), and human immunodeficiency virus (HIV), mostly due to social vulnerability and limited access to preventive care and health services.[1,2]

Similarly, other infectious diseases frequently occur in this population.[3,4] A recent report estimates 221,869 homeless persons living in Brazil, of which 24,344 in São Paulo city. There was a 140% increase in the country's homeless population from 2012 to 2020, mostly associated with the economic crisis leading to unemployment and poverty.[5,6]

Homeless persons are at increased risk of acquiring infectious diseases due to the limited access to treatment and prevention programs, poor hygiene, increased use of alcohol and injectable drugs, and unprotected sex.[7,8] The present study aimed to investigate the presence of antibodies against syphilis, HCV, and HIV in homeless persons in São Paulo, Brazil, the largest city in Latin America.

Material and methods

This study was a descriptive cross-sectional seroepidemiological approach of a homeless population in a major daytime attendance shelter of western São Paulo city. The research was conducted along with the city's multi-professional team of the Secretary of Health, called "Street Outreach Office", part of the Brazilian Unified Health System (SUS).[9] This study was approved by the Ethics Committee in Human Research of the Federal University of Paraná (CAAE: 80099017.3.0000.0102, protocol number: 2.512.196), by the Ethics Committee in Human Health of the São Paulo City Secretary of Health (CAAE: 80099017.3.3004.0086, protocol number: 3.366.684) and by the Ethics Committee in Human Research of the Clinics Hospital at the Federal University of Paraná (CAAE: 80099017.3.3005.0096, protocol number: 3.623.845). The patients/participants provided their written informed consent to participate in this study.

Homeless persons were recruited by city health officials and invited to voluntarily participate in the research. Blood samples of 116 volunteers were conveniently collected from June to August 2018, which was the permitted timeframe. Epidemiological data collection was carried out using a questionnaire designed for homeless persons; refusal to fully or partially answer any question or incomplete answers was accepted and registered.

Detection of syphilis, HCV and HIV was performed by chemiluminescent microparticle immunoassay (CMIA) (Anility i Syphilis TP, Anti-HCV, HIVAg/Ab, Abbott Laboratories, Chicago, IL, USA). Cases of reactive serology for HIV were confirmed by a rapid immunoblot test (DPP HIV1/2®, Fiocruz, Rio de Janeiro, Brazil), as recommendations of the Brazilian Ministry of Health.[10] Although screening for viral hepatitis B by rapid test has been also incorporated by SUS, the São Paulo Secretary of Health had a shortage of these tests throughout 2018. CMIA-reagent samples for anti-*T. pallidum* antibodies were confirmed by Venereal Disease Research Laboratory (VDRL) non-treponemal test. VDRL non-reagent samples were confirmed by treponemal rapid immunochromatographic test (MedTeste Sífilis MedLevensohn®, São Paulo, Brazil), as recommended by SUS.[11] Reagent results may represent early syphilis in recent infection or untreated late syphilis, also in activity.[11]

Statistical analysis was performed using SPSS 20.0.[12] Frequencies of syphilis and HCV seropositivity (absolute and relative) were determined by the stratification of the observations according to variables. Chi-Square test was used to determine univariate association between studied variables, and odds ratios (OR) were used to assess the association between syphilis and HCV prevalence and potential risk factors. Association between factors was considered when $p < 0.05$.

Results

In short, profile of surveyed homeless volunteers included 103/116 (88.8%) males, 105/116 (90.5%) unmarried and 89/116 (76.7%) non-white individuals. A total of 77/116 (66.4%) persons had none to 8th grade as educational background and 97/116 (83.6%) declared receiving no income at the time. While 29/116 (25.0%) individuals were assisted by psychosocial service, 87/116 (75.0%) persons referred use of legal or illegal substances, mostly alcohol in 62/87 (71.3%), followed by tobacco in 32/87 (36.8%) and cocaine in 31/87 (35.6%) individuals. São Paulo was not the city of birth for 80/116 (69.0%) individuals, 64/116 (55.2%) slept in shelters at night, and family conflicts was the main reported reason to have become homeless in 47/116 (40.5%) answers (Table 1).

CMIA-reagent samples for anti-*T. pallidum* antibodies were confirmed by VDRL non-treponemal test. VDRL non-reagent samples were confirmed by treponemal rapid immunochromatographic test. A rapid immunoblot assay confirmed seropositivity to HIV. Overall, anti-*T. pallidum* antibodies were observed in 29/116 (25.0%), anti-HCV antibodies in 4/116 (3.4%), and anti-HIV antibodies in 2/116 (1.7%) individuals, both co-infected with *T. pallidum* (Table 2).

Associated risk factors for syphilis exposure in homeless were to be born or had previously lived in another city (p = 0.043) and to have become homeless due to family

Table 1 – Statistical results of univariate and multiple logistic regression models of associated risk factors for seropositivity of anti- *Treponema pallidum* and anti- HCV antibodies in homeless persons.

Variables		Syphilis				HCV			
		Positive/N	%	OR (CI 95%)	*p-value*	Positive/N	%	OR (CI 95%)	*p-value*
Sex	Male	24/103	23.3	0.49 (0.4–1.62)	0.234	3/103	2.9	0.38 (0.04–3.39)	0.383
	Female	5/13	38.5	(ref)		1/13	7.7	(ref)	
Pregnancy	Yes	1/2	50.0	3.07 (0.19–50.73)	0.410	0/2	0.0	*	0.932
	No	28/114	24.6	(ref)		4/114	3.5	(ref)	
Marital Status	Unmarried	25/105	23.8	0.55 (0.15–2.02)	0.360	4/105	3.8	*	0.668
	Accompanied	4/11	36.4	(ref)		0/11	0.0	(ref)	
Racial self-declaration	White	6/27	22.2	0.82 (0.29–2.28)	0.704	0/27	0.0	*	0.572
	Non-white	23/89	25.8	(ref)		4/89	4.5	(ref)	
Educational background	None to 8th grade	20/77	26.0	1.09 (0.44–2.71)	0.850	4/77	5.2	*	0.302
	High school and university	9/37	24.3	(ref)		0/37	0.0	(ref)	
Income	No income	26/97	26.8	2.93 (0.63–13.63)	0.154	2/97	2.1	0.19 (0.03-1.23)	0.115
	With income	2/18	11.1	(ref)		2/18	11.1	(ref)	
Assistance by Psycho-social Care Centers (CAPS)	Yes	4/29	13.8	0.39 (0.12–1,26)	0.108	2/29	6.9	3.00 (0.44-20.35)	0.260
	No	25/87	28.7	(ref)		2/87	2.3	(ref)	
Use of licit and/or illicit drugs	Yes	22/87	25.3	1.06 (0.40–2.83)	0.901	3/87	3.4	1.00 (0.11-9.24)	0.740
	No	7/29	24.1	(ref)		1/29	3.4	(ref)	
Alcohol consumption	Yes	16/62	25.8	1.09 (0.47–2.55)	0.830	2/62	3.2	0.87 (0.13–5.97)	0.637
Tobacco use	Yes	8/32	25.0	1.00 (0.39–2.56)	1.000	2/32	6.2	0.96 (0.10–8.84)	0.725
Cocaine use	Yes	4/31	12.9	0.36 (0.11–1.12)	0.069	1/30	3.3	2.62 (0.39–17.85)	0.305
Marijuana use	Yes	6/30	20.0	0.68 (0.25–1.89)	0.463	0/31	0.0	*	0.283
Crack use	Yes	3/14	21.4	0.79 (0.08–6.91)	0.742	0/14	0.0	*	0.593
Other drugs	Yes	1/5	20.0	0.74 (0.08–6.91)	0.792	0/5	0.0	*	0.836
City of origin	Others	25/80	31.3	3.52 (1.12–11.05)	0.043	4/80	5.0	*	0.394
	São Paulo	4/35	11.4	(ref)		0/35	0.0	(ref)	
Shelter type	Hostel	14/64	21.9	0.69 (0.29–1.60)	0.389	4/64	6.2	*	0.089
	Street	9/28	32.1	1.61 (0.63–4.11)	0.316	0/30	0.0	*	0.326
Causes for becoming homeless									
Family conflicts	Yes	6/47	12.8	(ref)	0.035	1/64	1.6	0.23 (0.02–2.31)	0.213
	No	19/64	29.7	2.88 (1.05–7.93)		3/47	6.4	(ref)	
Unemployment	Yes	8/33	24.2	1.15 (0.44–3.00)	0.778	1/33	3.0	0.79 (0.08–7.30)	0.657
	No	17/78	21.8	(ref)		3/78	3.8	(ref)	
Alcohol and drugs	Yes	5/26	19.2	0.77 (0.26–2.32)	0.646	2/26	7.7	3.27 (0.48–22.08)	0.233
	No	20/85	23.5	(ref)		2/85	2.4	(ref)	
Another motive	Yes	5/18	27.8	1.40 (0.45–4.41)	0.560	1/18	5.6	1.72 (0.19–15.64)	0.512
	No	20/93	21.5	(ref)		3/93	3.2	(ref)	
Housing loss	Yes	4/12	33.3	1.86 (0.51–6.77)	0.342	0/12	0.0	*	0.629
	No	21/99	21.2	(ref)		4/99	4.0	(ref)	

* The percentages can go higher than 100% because individuals could answer more than one option. N= number total; OR= odds ratio.

conflicts (p = 0.035). Other variables such as sex (p = 0.234), pregnancy (p = 0.410), marital status (p = 0.360), racial self-declaration (p = 0.704), educational background (p = 0.850), income (p = 0.154), assistance by psychosocial care centers (CAPS) (p = 0.108), use of licit and/or illicit drugs ($p > 0.05$), use of shelter (hostels, street, occupancy) ($p > 0.05$) were not statistically significant (Table 1). There were no risk factors significantly associated with the presence of anti-HCV antibodies ($p > 0.05$) (Table 1). Risk factors associated with HIV could not be analyzed due to the low HIV seropositive rate.

Discussion

To the authors' knowledge, the seroprevalence of anti-*T. pallidum* antibodies assessed herein (25.0%) is the highest in homeless persons worldwide, which ranges from 3/569 (0.5%) in Iran,[13] 5/175 (2.9%) in Kenya,[14] 22/554 (4.0%) in India,[15] to 18/132 (13.6%) in the USA.[16] Additionally, syphilis was detected in 19/330 (5.7%) homeless persons in 2002-2003,[17] and in 97/1,391 (7.0%) in 2006-2007,[2] both in São Paulo city, which is about 4-fold lower than the rate in present study. Five surveys have found a higher prevalence of syphilis, in other vulnerable populations, including 141/450 (31.3%) prisoners in Ethiopia,[18] 82/222 (36.9%) sex workers in Brazil,[19] 273/598 (45.6%) in Argentina [20] and 51.1% (1,010/1,978) in Rwanda,[21] and 269/529 (50.8%) refugees in Italy.[22]

In the present study, seropositivity of anti-*T. pallidum* antibodies among homeless persons was associated with city of birth or previously living in a city other than Sao Paulo (p = 0.043) and had become homeless due to family conflicts (p = 0.035). In São Paulo city, most homeless persons are migrants or refugees[17] and with broken or fragile family bonds,[9] corroborating the findings of a previous study with migrant workers in Eastern China, which reported higher seroprevalence of anti-*T. pallidum* antibodies among migrants with multiple sex partners and being divorced or widowed.[23] A retrospective case-control study in China with 17,585 inpatients screened for syphilis infection by serological tests, *T. pallidum* exposure was also associated to migration between

Table 2 – Results of anti-*Treponema pallidum*, anti-HCV and anti-HIV antibodies in homeless people of São Paulo city, Brazil.

Sample ID	Syphilis CMIA	Syphilis VDRL	Syphilis Treponemal Rapid Test	HCV CMIA	HIV CMIA	HIV imunoblot
SP01	seronegative			seronegative	seronegative	seronegative
SP02	seronegative			seronegative	seronegative	seronegative
SP03	17.86	1:2		seronegative	seronegative	seronegative
SP04	seronegative			seronegative	seronegative	seronegative
SP05	seronegative			seronegative	seronegative	seronegative
SP06	13.72	1:1		seronegative	seronegative	seronegative
SP08	seronegative			seronegative	seronegative	seronegative
SP11	seronegative			seronegative	seronegative	seronegative
SP13	15.23	1:4		seronegative	seronegative	seronegative
SP14	seronegative			seronegative	seronegative	seronegative
SP16	seronegative			seronegative	seronegative	seronegative
SP17	seronegative			seronegative	seronegative	seronegative
SP18	seronegative			seronegative	seronegative	seronegative
SP19	seronegative			seronegative	seronegative	seronegative
SP20	seronegative			seronegative	seronegative	seronegative
SP21	seronegative			seronegative	seronegative	seronegative
SP23	10.23	1:2		seronegative	seronegative	seronegative
SP24	seronegative			seronegative	seronegative	seronegative
SP25	seronegative			seronegative	seronegative	seronegative
SP26	5.19	1:1		seronegative	seronegative	seronegative
SP27	13.91	1:1		seronegative	692.63	HIV-1
SP28	seronegative			seronegative	seronegative	seronegative
SP29	15.88	1:1		seronegative	seronegative	seronegative
SP30	seronegative			seronegative	seronegative	seronegative
SP31	seronegative			seronegative	seronegative	seronegative
SP32	seronegative			seronegative	seronegative	seronegative
SP33	seronegative			seronegative	seronegative	seronegative
SP34	16.87	1:2		seronegative	seronegative	seronegative
SP35	seronegative			seronegative	seronegative	seronegative
SP36	seronegative			seronegative	seronegative	seronegative
SP37	1.57	NR	seropositive	seronegative	seronegative	seronegative
SP38	20.01	1:128		seronegative	seronegative	seronegative
SP39	seronegative			seronegative	seronegative	seronegative
SP40	seronegative			seronegative	seronegative	seronegative
SP41	20.09	1:128		seronegative	seronegative	seronegative
SP42	seronegative			seronegative	seronegative	seronegative
SP43	seronegative			seronegative	seronegative	seronegative
SP44	seronegative			seronegative	seronegative	seronegative
SP45	seronegative			seronegative	seronegative	seronegative
SP46	seronegative			seronegative	seronegative	seronegative
SP47	seronegative			seronegative	seronegative	seronegative
SP48	seronegative			seronegative	seronegative	seronegative
SP49	seronegative			seronegative	seronegative	seronegative
SP50	seronegative			seronegative	seronegative	seronegative
SP51	4.1	NR	seropositive	seronegative	seronegative	seronegative
SP52	seronegative			seronegative	seronegative	seronegative
SP53	seronegative			seronegative	seronegative	seronegative
SP54	seronegative			seronegative	0.91	seronegative
SP55	seronegative			seronegative	seronegative	seronegative
SP56	17.94			seronegative	seronegative	seronegative
SP57	seronegative			seronegative	seronegative	seronegative
SP58	seronegative			seronegative	seronegative	seronegative
SP59	seronegative			seronegative	seronegative	seronegative
SP60	seronegative			seronegative	seronegative	seronegative
SP61	seronegative			seronegative	seronegative	seronegative
SP62	seronegative			seronegative	seronegative	seronegative
SP63	seronegative			seronegative	seronegative	seronegative
SP64	seronegative			seronegative	seronegative	seronegative
SP65	seronegative			seronegative	seronegative	seronegative
SP66	seronegative			seronegative	seronegative	seronegative
SP67	13.76	1:1		seronegative	seronegative	seronegative
SP68	seronegative			seronegative	seronegative	seronegative
SP69	seronegative			seronegative	seronegative	seronegative

Table 2 (*continued*)

Sample ID	Syphilis CMIA	Syphilis VDRL	Syphilis Treponemal Rapid Test	HCV CMIA	HIV CMIA	HIV imunoblot
SP70	seronegative			seronegative	seronegative	seronegative
SP71	12.68	1:.1		seronegative	seronegative	seronegative
SP72	seronegative			seronegative	seronegative	seronegative
SP73	seronegative			seronegative	seronegative	seronegative
SP74	15.78	1:4		seronegative	seronegative	seronegative
SP75	seronegative			seronegative	seronegative	seronegative
SP76	seronegative			seronegative	seronegative	seronegative
SP77	seronegative			seronegative	seronegative	seronegative
SP78	seronegative			seronegative	seronegative	seronegative
SP79	2.33	NR	seropositive	1.25	seronegative	seronegative
SP80	seronegative			seronegative	seronegative	seronegative
SP81	seronegative			seronegative	seronegative	seronegative
SP82	seronegative			seronegative	seronegative	seronegative
SP83	seronegative			seronegative	seronegative	seronegative
SP84	seronegative			seronegative	seronegative	seronegative
SP85	2.52	NR	seronegative	seronegative	seronegative	seronegative
SP86	15.89	1:32		seronegative	seronegative	seronegative
SP87	seronegative			seronegative	seronegative	seronegative
SP88	seronegative			seronegative	seronegative	seronegative
SP89	19.89	1:256		seronegative	seronegative	seronegative
SP90	9.21	NR	seropositive	seronegative	seronegative	seronegative
SP91	20.01	1:8		seronegative	1.35	seronegative
SP92	seronegative			seronegative	seronegative	seronegative
SP93	seronegative			seronegative	seronegative	seronegative
SP94	seronegative			seronegative	seronegative	seronegative
SP95	seronegative			seronegative	seronegative	seronegative
SP96	seronegative			seronegative	seronegative	seronegative
SP97	seronegative			seronegative	seronegative	seronegative
SP98	14.15	1:4		seronegative	seronegative	seronegative
SP99	seronegative			seronegative	seronegative	seronegative
SP100	2.95	NR	seropositive	1.11	seronegative	seronegative
SP101	seronegative			15.97	seronegative	seronegative
SP102	seronegative			seronegative	seronegative	seronegative
SP103	seronegative			7.66	seronegative	seronegative
SP104	15.79	1:2		seronegative	261.79	HIV-1
SP105	seronegative			seronegative	seronegative	seronegative
SP106	seronegative			seronegative	seronegative	seronegative
SP107	18.77	1:8		seronegative	seronegative	seronegative
SP108	seronegative			seronegative	seronegative	seronegative
SP109	seronegative			seronegative	seronegative	seronegative
SP110	seronegative			seronegative	seronegative	seronegative
SP111	10.65	1:2		seronegative	seronegative	seronegative
SP112	seronegative			seronegative	seronegative	seronegative
SP113	seronegative			seronegative	seronegative	seronegative
SP114	seronegative			seronegative	seronegative	seronegative
SP115	seronegative			seronegative	seronegative	seronegative
SP 116	19.03	1:8		seronegative	seronegative	seronegative
SP118	seronegative			seronegative	seronegative	seronegative
SP119	seronegative			seronegative	seronegative	seronegative
SP120	15.96	1:16		seronegative	seronegative	seronegative
SP121	seronegative			seronegative	seronegative	seronegative
SP122	seronegative			seronegative	seronegative	seronegative
SP123	17.02	1:8		seronegative	seronegative	seronegative

cities.[24] No risk factors for HCV exposure among homeless persons were significantly associated in the present study. Future investigations should be conducted to fully ascertain risk factors for HCV and HIV coinfection in homeless persons.

Besides homeless vulnerability, worldwide shortage of benzathine penicillin supply, the drug of choice for active syphilis treatment, may have also impacted disease spreading at the time.[25] Not surprisingly, the Brazilian epidemic of syphilis contrasts with other Latin American countries, which have moved towards syphilis eradication.[26,27] Other aspects may have also contributed to increase syphilis rates, including greater access to syphilis testing through rapid testing in

primary health care services.[25] The detection rate of acquired syphilis in Brazil increased from 2.1/100,000 in 2010 to 34.1/100,000 in 2015[28] and to 75.8 100,000 inhabitants in 2018.[29]

According to the recent national guidelines for management of sexually transmitted infections, the diagnosis of syphilis should incorporate clinical history, history of previous treatment, symptoms, in addition to results of treponemal and non-treponemal tests.[11] As limitation, VDRL reagent results may represent early syphilis in recent infection or untreated late syphilis, also in activity. Therefore, the prevalence of syphilis in activity herein may be underestimated. Further studies should also consider molecular diagnosis, particularly due to higher sensitivity in primary syphilis, associated with clinical signs such as exanthema and ulcers.[30]

In the present study, 2/116 (1.7%) individuals have shown anti-HIV antibodies. Herein, a 4th generation test (CMIA) was used as screening test and confirmed by a 2nd generation test (immunoblot assay), according to a recognized algorithm of the Brazilian Ministry of Health.[10] Also, anti-HCV antibodies were detected in 4/116 individuals by CMIA, as recommended by Brazilian Ministry of Health.[11] Although the Ministry of Health has also recommended molecular testing for HIV and HCV clinical cases, the present study focused on the epidemiological approach of viral exposure rather than viral load, prognosis and treatment.

In summary, the highest worldwide syphilis prevalence to date found in the present study indicates multiple preventable causes, which may profoundly impact homeless persons' health and wellbeing. More critical, strongly associating syphilis to homeless vulnerability and lack of preventive measures and treatment. Further studies should be conducted to fully establish risk factors for sexually transmitted infections exposure in homeless people.

Conflicts of interest

The authors declare that they have no conflict of interest.

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