

High Prevalence of Common Sexually Transmitted Diseases in Patients with Urological Symptoms Attending the Public Hospital Network from the City of Bogotá, Colombia

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Abstract

Sexually transmitted infections (STI) are a group of bacterial, viral, parasite and fungal diseases of high public health concern worldwide, affecting both men and women. The aim of the present study was to establish the prevalence of a set of STI causative microorganisms in urine samples from a sexually active population presenting urological symptoms and attending a public hospital network in Bogotá, Colombia. The presence of *Chlamydia trachomatis*, *Neisseria gonorrhoeae*, *Mycoplasma genitalium*, *Ureaplasma urealyticum*, *Ureaplasma parvum*, *Trichomonas vaginalis*, *Mycoplasma hominis*, HPV 6/11 and Herpes simplex virus types 1 and 2, was determined using a molecular approach. Overall, positivity for any of the studied STI was 60 and 10% for women and men, respectively. The presence of urinary symptoms was associated with the highest percentage (61.1 and 75% in women and men, respectively). These results provide insights into the movement and behavior of microorganisms causing STI in the sexually active population and their possible association with urinary problems.

Keywords: Sexually transmitted infections; Prevalence; Molecular approach

Introduction

Sexually transmitted infections (STI) are considered a public health problem that may cause disease symptoms in sexually active men and women [1]. STI involve several bacteria (*Chlamydia trachomatis*, *Neisseria gonorrhoeae*, *Ureaplasma spp.*, *Mycoplasma spp.*, *Treponema pallidum* and *Haemophilus ducreyi*), viruses (human Papillomavirus types 6 and 11 [HPV 6/11], Herpes simplex virus types 1 and 2 [HSV-1/2], human immunodeficiency virus [HIV] and hepatitis B virus [HBV], parasites (*Trichomonas vaginalis*) and yeasts (*Candida albicans*), promoting globally distributed diseases with high morbidity and mortality [2].

According to the World Health Organization (WHO), 357 million new cases of Chlamydia, *Neisseria gonorrhoeae*, *Treponema pallidum* and *Trichomonas vaginalis* were reported in the past year, only in the US continent the region of the Americas [3]. This situation should urge national and international organizations to exert every effort to control STI-caused diseases by designing promotion and prevention programs for sexual and reproductive health improvement. However, the diagnostic systems used in district and national screening programs do not cover all the microorganisms associated with STI, and the efficacy of these identification systems relies not only on their sensitivity and specificity, on sample type and positive and negative predictive values, but also on the prevalence of these pathogens in the general population [4]. In this sense, molecular techniques provide higher sensitivity (90% to 95%) and specificity (95%) through non-invasive alternatives such as urine samples, allowing an easy estimation of the prevalence of pathogens in sexually active men and women [5]. Also, the analysis of urine with molecular approaches enables STI identification in certain pathologies where other microorganisms with similar clinical symptoms are responsible for disease development and are not properly treated, such as urinary tract infections (UTI), urethritis, cystitis and kidney stones [6]. The aim of the present study was to establish the

prevalence of *Chlamydia trachomatis*, *Neisseria gonorrhoeae* and other microorganisms causing STI, such as *Mycoplasma hominis*, *Mycoplasma genitalium*, *Ureaplasma urealyticum*, *Ureaplasma parvum*, *Trichomonas vaginalis*, HPV 6/11 and HSV 1/2, in urine samples from a sexually active population attending a public hospital network in Bogotá, Colombia.

Materials and Methods

One hundred urine samples obtained from 60 women and 40 men between 18 and 65 years of age were collected in the period January-June 2015. All patients signed a voluntary informed consent and responded a brief questionnaire. Sexually inactive subjects, minor patients or patients under antibiotic treatment were excluded. The study protocol was approved by the Ethics Committee from the Secretary of Public Health, Bogota, Colombia.

Urine sample collection

The Cobas® PCR Urine Sample Kit (Roche Molecular Diagnostics, USA) was used to collect 20 to 50 ml urine for *Chlamydia trachomatis* and *Neisseria gonorrhoeae* DNA detection. Another 15 ml of urine were centrifuged at 2500 g for 10 min, and the pellet was processed for

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detection of *Mycoplasma hominis*, *Mycoplasma genitalium*, *Ureaplasma urealyticum*, *Ureaplasma parvum*, *Trichomonas vaginalis*, HPV 6/11/11 and HSV 1/2 DNA. Samples were stored at 8°C and -20°C, as appropriate, in the facilities of the Public Health Laboratory, Bogotá, Colombia, until processing for DNA analysis.

Molecular detection

The molecular identification of *Chlamydia trachomatis* and *Neisseria gonorrhoeae* was performed using the Cobas® 4800 CT/NG Test (Roche), according to the manufacturer's instructions [7]. Molecular testing of *Mycoplasma hominis*, *Mycoplasma genitalium*, *Ureaplasma urealyticum*, *Ureaplasma parvum*, *Trichomonas vaginalis*, HPV 6/11 and HSV-1/2 was performed using the commercial Geno Flow STD Array Test Kit (Diagcor, Hong Kong, China). This technique is based on PCR amplification and subsequent "continuous flow" hybridization, which allows the identification of nine pathogens in a single PCR reaction and one hybridization process [8].

Statistical analysis

Sample size was calculated with the number of positive urine cultures obtained at the Hospital "El Tunal ESE Bogota Level III" during 2014 and STI prevalence in the district of Bogotá [9]. Epi Info version 3.5.1 was used for data analysis, indicating that 100 samples were enough to reach 95% confidence and 5% total error. STI prevalence as well as all additional relationships of sociodemographic and clinical data were analyzed in a database developed in Microsoft Excel.

Results

Sixty percent (36/60) of women had at least one STI causative

organism. Among them, the prevalence of *Ureaplasma spp.* was 94.0%, followed by *Mycoplasma hominis* (22.2%), *Chlamydia trachomatis* (14%) and *Trichomonas vaginalis* (2.8%). All female patients were negative for *Neisseria gonorrhoeae*, *Mycoplasma genitalium*, HPV 6/11 and HSV-1/2 (Figure 1). However, 30.6% of samples were positive for multiple infections, namely, *Ureaplasma spp.* and *Mycoplasma hominis* showed the highest prevalence (18.0%), followed by *Mycoplasma hominis* and *Chlamydia trachomatis* (13.0%). In men 10% (4/40) of the analyzed samples had at least one STI causative organism. Similarly, the prevalence of *Ureaplasma spp.* was 60.0%, followed by *Chlamydia trachomatis* and HPV 6/11 (20.0%).

Results of the relationship between STI prevalence in women and UTI episodes showed that 22/39 (61.1%) of women had the highest STI prevalence, being *Ureaplasma spp.* responsible for the highest number of STI cases (n=21) (Table 1). Considering the microscopic findings in the 60 female patients, 40 (66.7%) underwent urinalysis and 20 (33.3%) underwent urine culture. More than half (51.7%) of patients showing a cellularity ≥ 2 cells/field presented an STI, while patients with a cellularity ≥ 3 cells/field (27.5%) and >4 leukocytes/field (25.0%) were always STI-positive. Further, 75.0% of patients with ≥ 3 bacteria/field and patients with a positive urine culture were always positive for an STI.

Among men reporting urinary tract symptoms, 66% presented *Ureaplasma spp.* infection and 33% displayed *Chlamydia trachomatis* infection. Microscopic findings showed that 27/40 male patients underwent urinalysis and 13/40 underwent urine culture. Almost 7.4% of patients referred to urinalysis (≥ 2 cells/field) had an STI. Patients with ≥ 4 leukocytes/field were always positive for an STI, and 4% of

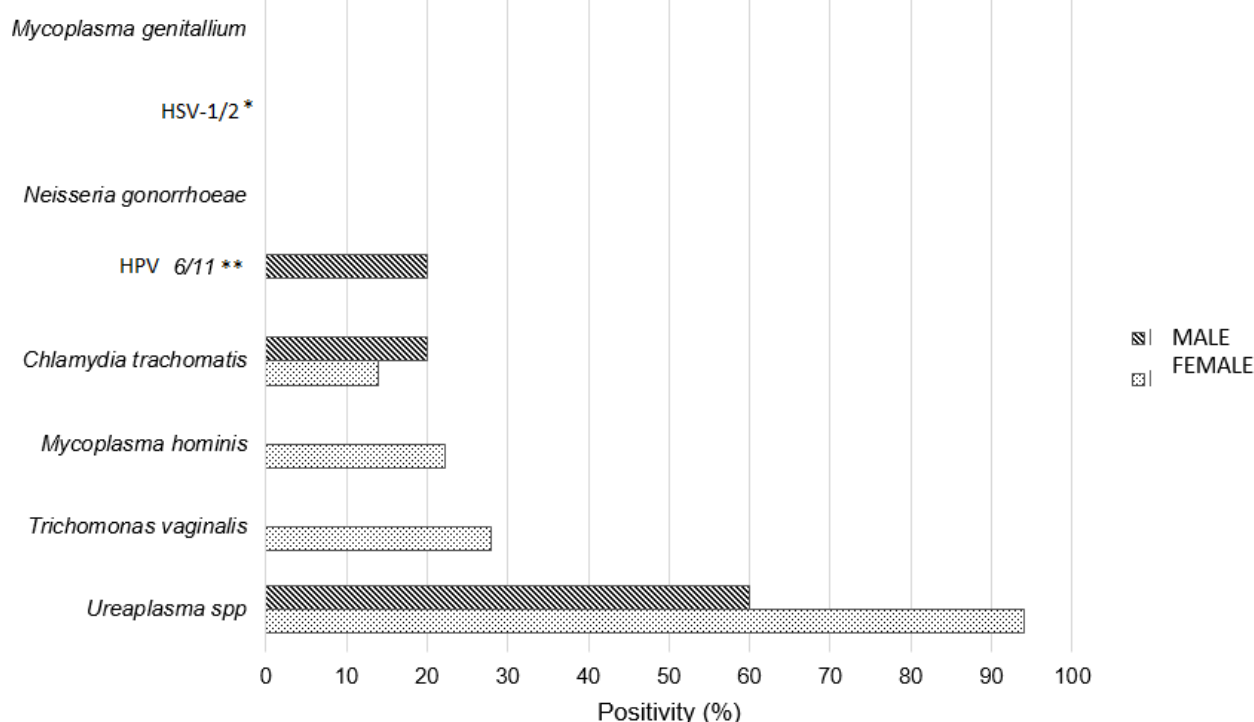


Figure 1: Positivity of STI-causative micro-organisms according to sex. * HSV-1/2: Herpes simplex virus type 1 (HSV-1) and Herpes simplex virus type 2 (HSV-2). ** HPV: Low-risk human papillomavirus types 6 and 11.

Urinary symptom	Positive STI cases			
	WOMEN		MEN	
	n=36	%	n=4	%
Yes	22	61.1	3	75.0
No	12	33.3	1	25.0
Unknown	2	5.6	0	0

Table 1: Positive STI according to urinary symptoms.

URINALYSIS	WOMEN			MEN		
	(n=60)	Positive STI (n=36)	%	(n=40)	Positive STI (n=4)	%
Cellularity ≤ 2 XC	29	15	51.7	27	2	7.4
Cellularity ≥ 3 XC	11	11	100	0	0	0
Total	40	26	--	27	2	--
Leukocytes ≤ 3 XC	30	16	53.3	26	1	3.8
Leukocytes ≥ 4 XC	10	10	100	1	1	100
Total	40	26	--	27	2	--
Bacteria ≤ 2+	36	23	63.9	26	1	3.8
Bacteria ≥ 3+	4	3	75	1	1	100
Total	40	26	--	27	2	--
Urine Culture						
Positive	5	5	100	3	0	0
Negative	15	5	33.3	10	2	20
Total	20	10	--	13	2	--

XC: Observations per microscopic field. 2+, 3+: Bacteria crossover according to the microscopic field observed.

Table 2: Positive STI associated with urinalysis and urine culture.

patients showing ≥ 2 bacteria/field showed an STI. Finally, 100% of men with ≥ 3 bacteria/field were positive for an STI. Among patients with a negative urine culture (10/13), 20% showed an STI (Table 2).

Discussion

Our results showed that 94% of women were positive for *Ureaplasma spp.*, 22.2% were positive for *Mycoplasma hominis* and 14.0% for *Chlamydia trachomatis*. According to Sánchez and coworkers, *Ureaplasma spp.* is the most common organism in infections associated with urogenital mycoplasma species, and it may associate with other biological agents [9]. Thus, this group of pathogens is quite important in sexual and reproductive health and should be evaluated in routine microbiology laboratories. In the case of *Chlamydia trachomatis* infections, our results agree with those recently published reporting 9.7% prevalence in patients with cervicovaginal symptoms, 12.2% prevalence in sexually active women from Brazil and, more recently, 15.79% prevalence using an immunofluorescence technique [10-12]. In general, chlamydial genital infections present non-specific signs and symptoms, although most infections are asymptomatic [11]. This is crucial due to the significant role played by *C. trachomatis* in the etiology of urethritis in adults and children and because of its association with urinary tract infections, causing important clinical pictures such as cervicitis, pelvic pain syndrome and vaginal discharge syndrome in sexually active women [12,13].

In this study, the prevalence of multiple infections in women was 30.6%, being *Mycoplasma hominis* and *Trichomonas vaginalis* always associated with *Ureaplasma spp.* A report published by Sanchez and coworkers showed that 45.5% of positive cases were due to multiple infections, always associated with *Ureaplasma spp.* and *Mycoplasma hominis* [14]. Therefore, the fact that mycoplasma infections appear associated with other pathogens could be probably explained by the microflora present in vaginal mucosa, which can be exchanged with other microorganisms from mucous membranes and skin during

sexual activity [15,16]. Among men, 60% of samples were positive for *Ureaplasma spp.*, 20% for *Chlamydia trachomatis* and 20% for HPV 6/11. According to Laspina and colleagues *Ureaplasma spp* and *Chlamydia trachomatis* are the most common pathogens responsible for clinical situations in men, along with gonococcal urethritis and other genitourinary infections that, in some way, can be a source of infection for women [17-19].

Around sixty percent of STI positivity was associated with urinary symptoms. Such prevalence would be justified by the high number of microorganisms affecting the genitourinary system of women. Further, it could be explained because the female urethra is shorter and therefore promotes not only the interaction of both the normal flora of mucous and the surrounding skin, but also the spread of STI causative microorganisms. Similarly, the rate of UTI in women may increase with age and postmenopausal sexual activity due to pelvic prolapse, lack of estrogen and peri urethral normal flora colonization, allowing the concomitant growth of other microorganisms [20]. In another study, Sánchez and coworkers showed that urinary symptoms could present similarities with specific STI symptoms, such as cystitis, burning sensation when urinating, increased number of urination events and discomfort due to friction with the urethra [14]. These authors also related the presence of different UTI with organisms causing STI.

Ureaplasma spp. had the highest positivity in women who reported urinary symptoms (58.3%), followed by *Mycoplasma hominis* (19.4%). These results are comparable with those reported by Acosta and coworkers, who mentioned that 48% of cultures from patients with chronic urinary symptoms were *Ureaplasma spp.* positive [21]. Also, 7.9% of urine samples from patients showing chronic pyelonephritis obtained by suprapubic puncture were positive for *Ureaplasma spp.* and *Mycoplasma hominis*. These microorganisms have also been isolated from patients with stones, because ureaplasma induces struvite phosphate and calcium crystallization in urine [22]. Thus, they should be differentiated as STI episodes in UTI-associated pathologies.

In the group of women who underwent urinalysis, 11 presented ≥ 3 STI-positive cells per field. This result is supported by Martinez et al., who described several mechanisms of response to infections produced by the host when reporting the pathophysiology of UTI, such as adhesion of bacteria to specific molecules on the cell surface of urothelial epithelium, or the drag of infected cells by urine flow, which could be responsible for the association of STI with cell observation [23]. This process would also be assisted by the production of secretory IgA, which magnifies the inflammation and cell desquamation process.

In men, STI positivity related to urinary symptoms was 75.0%, showing that symptoms such as dysuria and burning during urination are associated not only with UTI but also with a silent STI not clearly diagnosed [24]. The same as in women, *Ureaplasma spp.* had the highest percentage of positivity (50.0%). These data are directly related to the diagnosis of nongonococcal urethritis, which produces itching in the meatus, urethral discharge and dysuria, deviating from the appropriate clinical management for disease treatment [25]. However, another study considered that infections by *Ureaplasma spp.* depend directly on the bacterial load and the microbial genotype involved in episodes of asymptomatic or symptomatic nongonococcal urethritis [26]. In this sense, genotyping of ureaplasma could help differentiate commensal from pathogenic microflora in the male urethra.

Among male patients who underwent urinalysis, 2/27 showing ≤ 2 cells/field were STI-positive (7.4%). This drop-in positivity, compared with women, could be explained because the male urethra is five times larger than the female urethra, decreasing cell desquamation and justifying the low recovery of normal or pathological microorganisms in male urine [27]. Therefore, since male urine is hypocellular and poor in pathogens, another kind of samples should be used, such as balanoprepucial swabs or urethral exudates for a better recovery of intracellular microorganisms of public health importance.

Conclusion

This is the first study performed in the city of Bogotá in symptomatic UTI patients determining positivity profiles of STI causative microorganisms that may trigger important public health clinical pictures, which are disregarded when designing promotion and prevention sexual and reproductive health programs.

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