Urethritis in women attending an STD clinic

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SUMMARY Of an unselected group of 159 women attending a sexually transmitted diseases (STD) clinic 20% (32) had symptoms of urethritis. A positive correlation existed between the finding of more than 10 polymorphonuclear leucocytes (PMNL) per high-power field in the Gram-stained urethral smear and the presence of *Neisseria gonorrhoeae*, *Chlamydia trachomatis*, and *Trichomonas vaginalis*. Conversely, these organisms were rarely isolated if no PMNL were present. Fewer cultures gave positive results for these organisms if micturition had occurred less than four hours before examination. *C trachomatis* was recovered from the urethra or endocervix in 29/150 (19.3%) and from the urethra alone in six women. In contrast, *N gonorrhoeae* was never recovered from the urethra in the absence of endocervical infection. Of the 159 women 10% had bacteriuria due to non-sexually transmissible agents; 50% had asymptomatic bacteriuria. All, however, had other urethral pathogens isolated as well. Thus, sexually transmitted disease agents are highly prevalent in women attending an STD clinic who have signs and symptoms of urethritis. As in non-gonococcal urethritis in men, *C trachomatis* may be an important cause of urethritis in women.

Introduction

The diagnosis of urethritis in men is relatively simple since it is established by using two objective criteria: the presence of urethral discharge and the finding of polymorphonuclear leucocytes (PMNL) in the urethral smear.¹ Thus, it is possible to speak of symptomatic urethritis in men—a common condition in sexually transmitted disease (STD) clinics—in which there is no dysuria, urgency, or frequency, yet the objective criteria of discharge only after urethral stripping and of PMNL in the exudate are still met.

On the other hand, it is difficult to establish this diagnosis with certainty in women because frequency and dysuria are common complaints and cystitis and bacteriuria are frequent diagnoses.² No objective criteria have been established to help distinguish urethritis from infections occurring elsewhere in the female urinary tract.

The aims of this study were to define the occurrence of sexually transmissible agents in the urethras of an unselected population of women attending an STD clinic and to correlate these with the number of

PMNL in the urethral smear—that is, to determine if these agents were colonisers or were causing an inflammatory process.

Patients and methods

STUDY POPULATION

All women over 18-years of age attending a suburban STD clinic in Atlanta, Georgia, USA, for the first time between November 1977 and April 1978 were potential candidates for the study. Those who had taken antibiotics during the two weeks before their first visit or who were pregnant or did not agree to participate were excluded. Women were purposely given no specific instructions about voiding before examination; thus none was excluded because she had micturated just before examination.

One physician (JW) obtained informed written consent and then interviewed and examined all the women.

CLINICAL PROCEDURES

A speculum pelvic examination was performed and material obtained for smears and cultures from three sites: the urethra (calcium alginate swab); the cervical canal, after cleansing externally with sterile saline

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(cotton swab); and the posterior vaginal pool (cotton swab). Gram-stained urethral smears were scanned under low power to locate the area of greatest leucocyte density where the average number of PMNL in five high-power fields ($\times 970$) was calculated. The smears were then examined for Gram-negative intracellular diplococci.

CULTURE

Specimens were cultured for *Neisseria gonorrhoeae*, *Chlamydia trachomatis*, *Mycoplasma hominis*, *Ureaplasma urealyticum*, *Gardnerella* (*Corynebacterium*) *vaginalis*, *Trichomonas vaginalis*, and *Candida* species. The posterior forniceal specimens were cultured for only the last three agents, since they are more commonly found in this location than in either the urethra or endocervix. The order in which urethral and endocervical specimens were collected was randomly changed every second week to avoid selection bias for any particular organism.

Patients were instructed how to clean the periurethral area and to catch a midstream urine sample in a sterile container. Clean-voided urine was cultured quantitatively to identify the pathogens commonly associated with non-sexually transmitted urinary tract infections, the *Enterobacteriaceae* and aerobic cocci.

MICROBIOLOGY

Urethral specimens

Swabs for the isolation of *C trachomatis* were placed in a buffered sucrose phosphate transport medium, immediately snap-frozen in an acetone-dry ice slush, and stored at -70°C until thawed for culture in the IUDR-treated McCoy cell system.³ Another swab was rinsed thoroughly in 0.6 ml of trypticase soy broth; one drop (0.1 ml) was examined immediately for trichomonads and others were plated on to peptone-starch-dextrose agar,⁴ 'V' agar⁵ for the isolation of *G vaginalis*, Saboraud's chloramphenicol agar for the isolation of yeasts, and Thayer-Martin medium for the isolation of *N gonorrhoeae*. The remainder was placed into two tubes of Diamond's medium⁶ for the isolation of *T vaginalis*. The plates for isolation of *N gonorrhoeae* and *G vaginalis* were placed into a candle-extinction jar and incubated at 35°C before transport to the laboratory. The last urethral swab was placed into mycoplasma transport medium⁷ and handled exactly like the specimens for chlamydia until thawed for culture of *U urealyticum* and *M hominis*.

Endocervical specimens

The procedure outlined for urethral swabs was followed, except that an individual cotton swab was used for each culture.

Posterior forniceal specimens

Cotton swabs were cultured for *G vaginalis*, *T vaginalis*, and yeasts in most patients.

Urine specimens

A calibrated loop was used to plate clean-voided urine on to eosin-methylene blue agar and 5% sheep blood agar for the quantitative estimation of organisms.

Smears and wet mounts were examined at the clinic; none of the laboratory personnel knew the clinical diagnosis.

STATISTICAL ANALYSIS

We used multivariate regression analysis to test for the statistical significance of the association between the type of microbial flora isolated from the urethra, endocervix, or vaginal pool and the presence or absence of urethral inflammation (presence of PMNL). We also used χ^2 analysis with Yates's correction to analyse the contingency tables and calculated predictive values. The predictive value of a positive test result—a measure of the probability that a positive test result correctly indicates infection—was calculated according to the following formula:

$$\frac{(\text{Prevalence of disease}) \times (\text{Sensitivity of test})}{(\text{Prevalence}) (\text{Sensitivity}) + (1 - \text{Prevalence}) (1 - \text{Specificity})} \times 100$$

The higher the positive predictive value, the more frequently a positive test result correlates with the disease state.

Results

One hundred and fifty-nine women ranging in age from 18 to 40 years (mean 23 ± 6.3) were studied; 87 were white and 72 black. Since there were no statistically significant differences between white and black women for the variables considered, data were not subdivided by race.

We chose the breakpoints of more than four hours after micturition and more or less than 10 PMNL/hpf in the urethral smear by using discriminant analysis. This procedure let us select values that best identified patient groups—the numbers of PMNL and the presence or absence of pathogens in the urethra identified by culture.

The major reasons for attending the clinic were venereal infections in sexual partners (49%) and symptoms of genital infection (45%). The presenting symptoms of all 159 women are summarised in table 1. Overall, 45.5% of women had no PMNL in their urethral smear, 39% had 1-10 PMNL, and 15.5% more than 10 PMNL/hpf.

TABLE I Presenting symptoms in 159 unselected women attending an STD clinic

Symptom	No of patients*	%
Vaginal discharge	79	50.0
Dysuria without frequency	16	10.1
Frequency without dysuria	25	15.7
Both dysuria and frequency	15	9.4
Vulval or vaginal pruritus	53	33.3
Lower abdominal pain	12	7.5
Genital ulcer	3	1.9
Other	28	17.6
No problem	43	27.0

*Most patients had more than one presenting symptom

CULTURE

The micro-organisms isolated, with particular reference to those from the urethra, are given in table II. No significant differences were found in the

TABLE II Micro-organisms isolated from the endocervix and urethra of women attending an STD clinic

Organism isolated	No examined	% Infected at any site	% Infected at urethra only*
<i>Neisseria gonorrhoeae</i>	159	20.7	0
<i>Chlamydia trachomatis</i>	150	19.3	20.7
<i>Gardnerella vaginalis</i>	119	26.9	34.4
<i>Ureaplasma urealyticum</i>	119	65.5	35.9
<i>Mycoplasma hominis</i>	119	47.9	24.6
<i>Trichomonas vaginalis</i>	158	39.6	7.8
<i>Candida species</i>	159	17.6	3.6

*Percentage of those patients infected at any site

isolation rates of pathogens from the urethra of patients either with or without the symptoms of urethral inflammation, dysuria and frequency. Nor was there a significant correlation between symptoms and the number of PMNL found on urethral smear. However, there was a significant association between the presence of more than 10 PMNL/hpf and the isolation of *N gonorrhoeae*, *C trachomatis*, and *T vaginalis* (table III). *C trachomatis* and the

gonococcus were rarely isolated if there were no signs of urethral inflammation. The number of PMNL had no correlation with the isolation of any other micro-organisms.

In all patients fewer cultures gave positive results if micturition occurred less than four hours before examination (table IV), but this finding was

TABLE IV Comparison of urethral isolation rates with hours elapsed since last micturition

Urethral isolate	% Isolations from urethra	
	0-4 hours (n = 137)	>4 hours (n = 22)
<i>Neisseria gonorrhoeae</i>	10.2	27.3
<i>Chlamydia trachomatis</i>	10.6	33.3*
<i>Gardnerella vaginalis</i>	18.8	55.5
<i>Ureaplasma urealyticum</i>	52.0	55.5
<i>Mycoplasma hominis</i>	33.0	66.6
<i>Trichomonas vaginalis</i>	13.2	22.8
<i>Candida species</i>	17.5	18.2

*Statistically significant difference ($\chi^2 = 7.7$, $P = 0.02$)

statistically significant only for *C trachomatis*. In addition, numbers of PMNL in the urethral smear were significantly greater when the time-lapse between micturition and examination was more than four hours (table V).

TABLE V Relationship between the number of leucocytes in urethral smears and the hours elapsed since last micturition

No of hours since last micturition	PMNL in the urethral smear/hpf				
	0-10		>10		
	No of patients	%	No of patients	%	Total
0-4	90	92.8	47	75.8	137
>4	7	7.2	15	24.2	22
Total	97	100.0	62	100.0	159

 $\chi^2 = 9.2$, $p = 0.01$

TABLE III Correlation of urethral pathogens with urethral leucocyte counts

	Urethral leucocyte count/hpf						Statistical values	
	0		1-10		>10			
Urethral isolate	No of specimens	% Positive	No of specimens	% Positive	No of specimens	% Positive	χ^2 *	P
<i>Neisseria gonorrhoeae</i>	71	0.0	26	11.5	62	27.4	22.66	<0.001
<i>Chlamydia trachomatis</i>	71	5.6	22	4.6	57	26.3	13.43	<0.01
<i>Gardnerella vaginalis</i>	55	25.5	20	10.0	44	11.4	4.31	NS
<i>Ureaplasma urealyticum</i>	55	40.0	20	50.0	44	40.9	0.64	NS
<i>Mycoplasma hominis</i>	55	29.1	20	30.0	44	43.2	2.36	NS
<i>Trichomonas vaginalis</i>	70	21.4	26	34.6	62	43.5	7.44	<0.05
<i>Candida</i> species	71	16.9	25	16.0	62	8.1	2.43	NS
No pathogen isolated	55	21.8	20	25.0	44	9.1	3.62	NS

* χ^2 value with 2 df comparing 0, 1-10, and >10 PMNL/hpf with positive culture results

If the urethral PMNL count alone was used to discriminate between those with gonorrhoea (urethral or cervical or both), the positive predictive value of more than 10 PMNL/hpf in the urethral smear for the presence of gonorrhoea was only 49.6% and for identifying chlamydial and trichomonal infections 49.0% and 45.4% respectively. The positive predictive value depends heavily on the prevalence of the infections in the population tested. Our results should not be extrapolated to other population groups with radically different prevalences of infection.

The wet-mount examination of urethral material for the identification of *C. albicans* gave a positive result in only one patient out of 21 with positive urethral culture results. *T. vaginalis* was identified in the urethral wet mount in 67% of those with a positive culture result.

Ten per cent (15/149) of the women had $\geq 10^5$ bacterial colony-forming units/mm³ isolated from clean-voided urine specimens. The organisms isolated were *Escherichia coli* (8); *Klebsiella pneumoniae* (2); *Enterobacter aerogenes* (1); *Proteus mirabilis* (1); and group B streptococci (3). All these women also had one or more other urethral pathogens isolated at the same time. Of these 15 women with bacteriuria, eight were asymptomatic but six had more than 10 urethral PMNL/hpf. Of the seven with symptoms, five had more than 10 PMNL/hpf.

Discussion

Urethral symptoms are common in women attending our clinic, and urethritis—here defined as an inflammatory response within the urethra—is a distinct entity. Although there was poor correlation between the degree of inflammation found in urethral smears and symptoms, a potential or real pathogen could be isolated from most women with urethral leucocytosis. This is not surprising, since in men it is clear that urethral symptoms do not necessarily correlate with the degree of inflammation that is present. Men with significant urethral leucocytosis may be asymptomatic, and conversely symptoms can exist when there is no evidence of inflammation.

Oriel *et al*⁸ empirically chose 10 or more PMNL/hpf in a urethral smear or urine specimen as diagnostic of urethritis in men. Swartz *et al*¹ refined that number in men to four or more PMNL/hpf in a urethral smear and ≥ 5 /hpf in a urine specimen. It seems a logical step to attempt to apply this finding to women; indeed there appear to be close parallels. We found only one published report on the numbers of PMNL in urethral smears of women with urethritis;⁹ 10 PMNL/hpf was chosen as the cut-off

point, but no rationale was given for this decision. In this study we selected this figure on the basis of a statistical analysis of our population. Ten PMNL/hpf is the value which best divides women with urethral pathogens from those without. Women with classical non-sexually transmitted bacteriuria rarely had urethral leucocytosis. However, the finding of significant urethral leucocytosis is not an absolute indicator of urethral infection. About 3% of women with gonococci and 6% with *C. trachomatis* isolated from their urethras had PMNL counts of less than 10/hpf.

Relying on symptoms alone, we underestimated the number of women with urethral colonisation. We were able to culture potential pathogens from the urethras of 80% and to demonstrate evidence of inflammation in an additional 8% of women who had no urinary tract symptoms. We recovered *C. trachomatis* from the urethra alone in 21% (6/29) of those with chlamydial infection. Conversely, the gonococcus was not recovered solely from the urethra in any case, a finding which has been observed in other studies.^{10 11} The organism most frequently isolated was *U. urealyticum*. The pathogenic status of this organism remains to be defined, but some evidence exists that it is sometimes a cause of urethritis in men.^{12 13}

We examined the washout effect of urine on the numbers of PMNL and organisms in urethral specimens. There was a significantly lower number of PMNL/hpf in the smears of those women who had urinated less than four hours before examination. There was also a tendency towards lower isolation rates for all organisms, although this was significant only for *G. vaginalis*.

The possibility that some of these organisms were vaginal contaminants introduced into the urethra on the swabs cannot be excluded. Although the finding of more than 10 PMNL/hpf on the urethral smear is a helpful indicator of genital infection, it does not distinguish between aetiological agents. However, a negative result on Gram stain for the gonococcus and on a wet mount for *T. vaginalis* should markedly increase the suspicion of a chlamydial infection. The presence of more than 10 PMNL/hpf in a smear together with a history of a sexual partner with urethritis should further increase the likelihood of isolating either *N. gonorrhoeae* or *C. trachomatis* from the urogenital tract (table VI).

Finally, 10% of our study population had organisms often associated with bladder or upper urinary tract infections. We did not attempt to confirm this diagnosis by repeated cultures, nor did we examine the role of these organisms in causing acute urethritis. However, the finding of bacteriuria in 10% of this unselected population raises the

question of whether screening for common urinary tract pathogens should not be a routine investigation in an STD clinic.

TABLE VI Correlation of urethral leucocytosis and contact history with isolation of gonococci or chlamydia or both

<i>Isolation N gonorrhoeae or chlamydia or both</i>	<i>No of urethral PMNL/hpf</i>				
	<i>>10: partner with urethritis</i>		<i>≤10: partner without urethritis</i>		
	<i>No</i>	<i>%</i>	<i>No</i>	<i>%</i>	<i>Total</i>
Positive	41	49.4	10	13.2	51
Negative	42	50.6	66	86.8	108
Total	83	100.0	76	100.0	159

$\chi^2 = 23.9$, $P < 0.001$

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