

## ORIGINAL ARTICLE

# Prevalence of Four Sexually Transmitted Infection Pathogens Detected in Urine and Genital Tract Secretion in Hangzhou, China

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### SUMMARY

**Background:** Sexually transmitted infections (STIs) increase gradually and have become a public health problem in the world. UU, CT, NG, and MG are four common STI pathogens. Our retrospective study analyzed the clinical situation and the laboratory data of patients infected with the four pathogens. The prevalence of the four pathogens, detected in urine and genital tract secretion, was studied in Hangzhou, China.

**Methods:** A total of 3,168 male and female patients were randomly selected from February 2023 to February 2024. Urine and genital secretions were collected, and four STI pathogens were controlled for detection. Data were collected from the hospital's electronic medical records, and SPSS 25.0 software was used to perform a statistical analysis.

**Results:** Among 3,168 patients, a total of 1,527 were detected as positive, and the positive rate was 48.20%. The age of patients ranged from 13 - 98 years, with an average age of 45.6. The total of patients consisted of 2,191 males and 977 females, which had a significant difference ( $p < 0.05$ ). Specimens were mainly collected from the Department of Dermatovenereology, Urological Surgery, Obstetrics and Gynecology, and so on. The positive rate was statistically different between male and female patients ( $p < 0.05$ ). Single infection performed a main role and accounted for 79.57% of all of the positive patients. In the  $\leq 20$  age group, the positive rate was the highest and was as high as 77.65%. In detail, single infection caused by UU dominated, especially in the 21 - 30 age group. Double infection caused by UU and CT and triple infection caused by UU, CT, and NG were the majority, both especially in the 21 - 30 age group. There were significant differences in the positive rates in the different age groups and in the four pathogens ( $p < 0.05$ ). Quadruple infection was very rare and had only been detected in one patient.

**Conclusions:** The prevalence of the four pathogens in Hangzhou was different from other regions. More male than female patients, more single than multiple infections, and more single and multiple infections occurring in young people were the features in Hangzhou. The study would provide reference for prevention, diagnosis, and treatment of STI.

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### KEYWORDS

Ureaplasma urealyticum (UU), Chlamydia trachomatis (CT), Neisseria gonorrhoeae (NG), Mycoplasma genitalium (MG), prevalence, Hangzhou

### INTRODUCTION

STI is still a global public health concern [1]. *Ureaplasma urealyticum* (UU), *Chlamydia trachomatis* (CT),

*Neisseria gonorrhoeae* (NG), and *Mycoplasma genitalium* (MG) are four common STI pathogens [2-3]. *Ureaplasma* spp. are known as some of the smallest free-living and self-replicating microorganisms, which are a unique genus of bacteria due to their essential requirement for urea in the synthesis of ATP. UU is one of the most common *Ureaplasma* species, which causes persistent or repeated infection of the urogenital tract [4-5]. *Chlamydia trachomatis*, the obligate intracellular pathogen, is the causative agent of blinding trachoma. It is also the world's leading cause of bacterial STI [6-7]. *Neisseria gonorrhoeae* is the etiologic agent of gonorrhea [8-9]. *Mycoplasma genitalium* is an important STI pathogen, affecting both male and female [10-11]. It grows extremely slowly *in vitro* and needs demanding culture requirements. So molecular-based diagnostic methods for its detection in clinical specimens are widely in use [12-13]. Males infected with the four pathogens tend to have symptoms like urethritis, prostatitis, epididymitis, and so on. There are a number of asymptomatic patients. Females often suffer from some symptomatic diseases, i.e. urethritis, vaginitis, cervicitis, endometritis, and even infertility. And asymptomatic female patients are also common. Generally, NG infection and non-NG (UU, CT, and MG) infection are always being focused on, which are the two main categories, according to the symptoms. NG infection always has a more purulent secretion, and symptoms of non-NG infection are less severe, with urethral burning, itching and mild inflammation. The current study was conducted to analyze the prevalence of the four STI pathogens detected in urine and genital tract secretions of people from Hangzhou, Zhejiang Province, China.

## MATERIALS AND METHODS

### Study population

All 3,168 patients, including outpatients and inpatients from several departments in the Hangzhou Third People's Hospital from February 2023 through February 2024, were selected, and data were collected from the hospital's electronic medical records. Hangzhou Third People's Hospital is a dermatovenerology-specialized hospital, and the Department of Dermatovenerology is its largest department. By analyzing the social structure of the study population, students accounted for 63.53% (54/85) in patients aged  $\leq 20$ , and the ratio reached 43.01% (329/765) in the 21 - 30 age group. In other age groups, none of the patients identified as students. The proportion of patients who were married was 77.62% (2,459/3,168). The proportion of asymptomatic and symptomatic male and female patients was 1:4 and 1:6, respectively. Patients with NG infection were more symptomatic than those with an infection of another of the three pathogens. Among all patients, one in five patients had a positive test without clinical symptoms.

### Instruments and reagents

UU, CT, NG, and MG detection kits were purchased from Shanghai Rendu Biotechnology Co., Ltd. (Production batch numbers: 20183401647, 20183401646, 20183401648, and 20173404323), and AutoSAT automatic nucleic acid detection and analysis system were used.

### Specimen collection

The main types of specimens were urine and genital secretion. 500  $\mu$ L of the first urine in the morning or the first urine after a long time (at least 1 hour) without urination was mixed with 500  $\mu$ L of sample preservation solution, and the mixed sample was used for testing. The secretions of male urethral orifice, female cervix and vagina were collected with sterile cotton swabs. The sterile cotton swabs were placed at 1 cm to 2 cm of the inner side of urethral orifice, cervix, and vagina. After rotating it once or twice and remaining it in for about 10 seconds, the heads of the sterile cotton swabs were soaked thoroughly in 1,000  $\mu$ L of normal saline in tube and were then squeezed against the tube wall. 500  $\mu$ L of the soaked solution and 500  $\mu$ L of sample preservation solution were taken to form a mixed solution for detection.

The collected specimens could be stored at 2 - 8°C for 30 days, -20°C for 3 months, and -70°C for 6 months if they could not be pretreated in time. Repeated freezing and thawing of frozen specimens was avoided.

### Detection methods

RNA isothermal amplification method was applied. The whole nucleic acid detection process consisted of two steps: nucleic acid extraction and thermostatic amplification.

### Statistical analysis

Statistical analysis was conducted with SPSS 25.0 software. Data of categorical variables were expressed as rate (%), which were analyzed with the chi-squared test. A p-value less than 0.05 was considered significantly different.

## RESULTS

### Clinical characteristics of patients

A total of 3,168 patients were randomly selected, with an average age of 45.6 and an age range of 13 - 98 years. All patients included 2,191 males and 977 females, which had a significant difference (69.16% vs. 30.84%,  $p < 0.05$ ).

### Distribution of STI positive patients in different gender and department

Out of 3,168 patients, 1,527 were detected as positive, and the positive rate was 48.20%. There were 957 positive cases in all 2,191 male patients, and the positive rate was 43.68%. Out of 977 female patients, 570 were positive, and the positive rate was 58.34%. There was a

**Table 1. Distribution of STI positive patients in different gender and department.**

	Number of STI test patients	Number of STI positive patients	Positive rate (%)
<b>Gender</b>			
Male	2,191	957	43.68
Female	977	570	58.34
<b>Department</b>			
Dermatovenereology	1,127	610	54.13
Urological Surgery	1,042	547	52.50
Gerontology	438	24	5.48
Obstetrics and Gynecology	375	258	68.80
Emergency Surgery	165	78	47.27
Nephrology	7	2	28.57
Respiratory Medicine	6	2	33.33
Endocrinology and Hematology	2	2	100.00
General Medicine	1	1	100.00
Surgery	1	1	100.00
Pain Medicine	1	0	0.00
Cerebrovascular Diseases	1	1	100.00
Emergency Internal Medicine	1	0	0.00
Proctology Medicine	1	1	100.00
<b>Total</b>	<b>3,168</b>	<b>1,527</b>	<b>48.20</b>

**Table 2. Distribution of single and multiple infection in all STI positive patients.**

Type	Single infection	Double infection	Triple infection	Quadruple infection	Total
Number (rate %)	1,215 (79.57)	253 (16.57)	58 (3.80)	1 (0.06)	1,527 (100.00)

**Table 3. Analysis of STI positive rates in different age groups.**

Age (years)	Number of STI test patients	Number of STI positive patients	Positive rate (%)
≤ 20	85	66	77.65
21 - 30	765	478	62.48
31 - 40	887	462	52.09
41 - 50	560	314	56.07
51 - 60	283	145	51.24
≥ 60	588	62	10.54
<b>Total</b>	<b>3,168</b>	<b>1,527</b>	<b>48.20</b>

**Table 4. Detailed distribution of single and multiple infection in all STI positive patients.**

Type	Number of positive patients	Positive rate in all STI infections (%)
<b>Single infection</b>		
UU	847	55.47
CT	152	9.95
NG	142	9.30
MG	74	4.85
<b>Double infection</b>		
UU and CT	112	7.33
UU and NG	35	2.29
UU and MG	51	3.34
CT and NG	40	2.62
CT and MG	5	0.33
NG and MG	10	0.65
<b>Triple infection</b>		
UU, CT, and NG	28	1.83
UU, CT, and MG	20	1.31
UU, NG, and MG	5	0.33
CT, NG, and MG	5	0.33
<b>Quadruple infection</b>		
UU, CT, NG, and MG	1	0.06
<b>Total</b>	<b>1,527</b>	<b>100.00</b>

significant difference (43.68% vs. 58.34%,  $p < 0.05$ ). In the distribution of department, the top five departments were the Department of Dermatovenereology, Urology, Gerontology, Obstetrics and Gynecology, and Emergency Surgery, and the Department of Obstetrics and Gynecology had the highest positive rate, which was 68.80%, as shown in Table 1.

#### **Distribution of single and multiple infection in all STI positive patients**

1,215 (79.57%) patients had a single infection, which was dominant in all 1,527 STI positive patients. Patients with a single infection were more than those with multiple infections (79.57% vs. 20.43%,  $p < 0.05$ ), which was shown in Table 2.

#### **Distribution of STI positive patients in different age groups**

All patients were divided into six age groups, including  $\leq 20$ , 21 - 30, 31 - 40, 41 - 50, 51 - 60, and  $\geq 60$  age group. In each age group, the STI positive rate was 77.65%, 62.48%, 52.09%, 56.07%, 51.24%, and 10.54%. The highest positive rate was observed in the  $\leq 20$  age group, and the rate was as high as 77.65%. There were significant differences in the positive rates among the different age groups ( $p < 0.05$ ), as shown in Table 3.

#### **Detailed distribution of single and multiple infection in all STI positive patients**

Single infection consisted of four different types, representing infection with UU, CT, NG, and MG. Patients with single UU, CT, NG, and MG infection had statistical differences (69.71% vs. 12.51% vs. 11.69% vs. 6.09%,  $p < 0.05$ ). Multiple infections included three different types, representing infection with two to four different pathogens. Double infections were subdivided into six combinations, including UU and CT, UU and NG, UU and MG, CT and NG, CT and MG, and MG and NG. Triple infections were subdivided into four combinations, including UU, CT, and NG, UU, CT, and MG, UU, NG, and MG, and CT, NG, and MG. Quadruple infection had only one combination which was UU, CT, NG, and MG. For single infection, infections caused by UU were significantly more than those with the other three pathogens, and infections caused by MG had the lowest incidence. For double infection, infections caused by UU and CT were obviously dominating, and infections caused by CT and MG were very little. For triple infection, infections caused by UU, CT, and NG were the majority, and infections caused by CT, NG, and MG were the minority. For quadruple infection, only one patient had been detected, which was greatly rare, as listed in Table 4.

**Table 5. Detailed distribution of single and multiple infection in different age group.**

Type	Positive rate in all STI positive patients (%)						
	≤ 20	21 - 30	31 - 40	41 - 50	51 - 60	≥ 60	Total
<b>Single infection</b>							
UU	30 (1.96)	243 (15.91)	259 (16.96)	199 (13.03)	69 (4.52)	47 (3.08)	847 (55.47)
CT	5 (0.33)	53 (3.47)	57 (3.73)	20 (1.31)	13 (0.85)	4 (0.26)	152 (9.95)
NG	11 (0.72)	37 (2.42)	35 (2.29)	31 (2.03)	22 (1.44)	6 (0.39)	142 (9.30)
MG	2 (0.13)	22 (1.44)	33 (2.16)	10 (0.65)	5 (0.32)	2 (0.13)	74 (4.85)
<b>Double infection</b>							
UU and CT	8 (0.52)	53 (3.47)	8 (0.52)	30 (1.96)	19 (0.24)	2 (0.13)	120 (7.86)
UU and NG	2 (0.13)	16 (1.05)	8 (0.52)	4 (0.26)	4 (0.26)	1 (0.06)	35 (2.29)
UU and MG	3 (0.20)	16 (1.05)	16 (1.05)	13 (0.85)	3 (0.20)	0 (0.00)	51 (3.34)
CT and NG	2 (0.13)	14 (0.92)	6 (0.39)	13 (0.85)	4 (0.26)	1 (0.06)	40 (2.62)
CT and MG	0 (0.00)	2 (0.13)	1 (0.06)	1 (0.06)	1 (0.06)	0 (0.00)	5 (0.33)
NG and MG	0 (0.00)	1 (0.06)	3 (0.20)	3 (0.20)	3 (0.20)	0 (0.00)	10 (0.66)
<b>Triple infection</b>							
UU, CT, and NG	3 (0.20)	11 (0.72)	5 (0.33)	2 (0.13)	5 (0.33)	2 (0.13)	28 (1.83)
UU, CT, and MG	1 (0.06)	8 (0.52)	7 (0.46)	3 (0.20)	1 (0.06)	0 (0.00)	20 (1.31)
UU, NG, and MG	0 (0.00)	0 (0.00)	2 (0.13)	1 (0.06)	1 (0.06)	1 (0.06)	5 (0.33)
CT, NG, and MG	0 (0.00)	2 (0.13)	1 (0.06)	1 (0.06)	1 (0.06)	0 (0.00)	5 (0.33)
<b>Quadruple infection</b>							
UU, CT, NG, and MG	0 (0.00)	1 (0.06)	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	1 (0.06)

### Detailed distribution of single and multiple infection in different age groups

In the detailed distribution of single and multiple infection in different age groups, single infection caused by UU, double infection caused by UU and CT, and triple infection caused by UU, CT, and NG were all dominating in the ≤ 20 age group, which also occurred in the 21 - 30, 51 - 60, and ≥ 60 age groups. In the 31 - 40 and the 41 - 50 age groups, single infection caused by UU, double infection caused by UU and CT, and triple infection caused by UU, CT, and MG were the most common. STI caused by four pathogens was detected in the 21 - 30 age group, as shown in Table 5.

### DISCUSSION

STI are considered a common disease. Patients with no typical symptoms always lead to a wide transmission and under-diagnosis, and undiagnosed patients are the source of infection in crowd gatherings [14-15]. A quick, sensitive, and specific detection method is urgently needed, and it would actively contribute to the control of transmission of STI. Patients with STI, who do not receive a timely and effective treatment, would have various short-, medium-, or long-term complications, which consist of urethritis, cervicitis, pelvic inflammatory disease, and premature birth [16-17]. At present, a lot of different kinds of laboratory methods can identify STI pathogens, from culture in medium to molecular testing. Culture in medium is regarded as the most specific diagnostic method, but it takes too much

time, needs specialized equipment, and only viable bacteria can grow in medium [18-19]. Nucleic acid amplification tests are applicable to the detection of viable and non-viable bacteria, whose sensitivity and specificity are both high, compared with the other test methods. They are suitable for many types of specimens, such as urine, urethral secretions, vaginal secretions, and sperm [20-21].

In our study, out of 3,168 patients, 1,527 were positive, and the positive rate was 48.20%, which was in agreement with the positive rate of other areas in China of previous studies [22-23]. There were 957 positive cases in all 2,191 male patients, and the positive rate was 43.68%. Out of all 977 female patients, 570 were positive, and the positive rate was 58.34%, which was statistically different ( $p < 0.05$ ). STI are transmitted through sexual contact, and the genitourinary system is often the diseased organ. The genitourinary system is physically different in males and females. The genitourinary organs of females are more open than the ones of males. The characteristics of the body structure makes females more susceptible to STI. In the distribution of department, the main departments were the Department of Dermatovenereology, Urological Surgery, Gerontology, Obstetrics and Gynecology, and Emergency Surgery. Among the top five departments, as shown in Table 1, the Department of Obstetrics and Gynecology had the highest positive rate (68.80%) and was related to the vulnerable genitourinary system of women. STI distribution is always obviously different in region and population. Previous studies showed that  $\leq 25$  age group was the peak of the STI-positive population in Taizhou, China, and it was also reported that the main infection age group was the 20 - 39 age group in the Jiangsu Province [24-25]. In our study, the positive rate was the highest in the  $\leq 20$  age group in Hangzhou, and the positive rates were significantly different among the different age groups ( $p < 0.05$ ). It also showed that STI-positive was more frequent in younger people. Younger people are always more sexually active, and it is consistent with the clinical characteristics of STI. It also suggests that increasing the STI screening tests for younger people is desperately needed. In Hangzhou, single infection played a main role, and it accounted for 79.57% of all positive patients, which was consistent with the results of the research by Zeng et al, who reported that single infection was more than multiple infection in Xi'an, China [26]. There was a significant difference between single and multiple infection. Multiple infection included double, triple, and quadruple infection. Double infection was the most frequent among multiple infection. It was rare to be infected with all four pathogens simultaneously and only one patient had been detected. Among the single infection, patients with UU infection comprised the majority and were evidently more than the other three pathogens. Among multiple infection, patients with UU and CT infection were the most common for double infection, and patients with UU, CT, and NG infection were the most

common for triple infection. Both were related to the overwhelming majority of UU infection. For single infection, the positive rate was the highest in the 21 - 30 age group. And for multiple infection, the positive rates were both the highest in the 21 - 30 age group, in the double as well as in the triple infections. To sum it up, STI was the most frequent in the 21 - 30 age group in Hangzhou. The reason for this might be that Hangzhou had always been an economically developed area, and the improvement of the living standards and the open sexual minds might have made the incidence of unsafe sexual behavior increase greatly among the younger people [27-28].

## CONCLUSION

In our study, the prevalence of UU, CT, NG, and MG infection in Hangzhou was retrospectively analyzed, and it was different from other regions. More male than female patients, more single than multiple infections, and more single and multiple infections occurring in young people were the features in Hangzhou. The study offered value for facilitating the development of an effective prevention and intervention and provided reference for clinical diagnosis and treatment of STI diseases.

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### Data Availability Statement:

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

### Ethics Approval Statement:

The research is reviewed and approved by the Ethics Committee of the Hangzhou Third People's Hospital and is in accordance with the Declaration of Helsinki (reference number: 2024KA027).

### Declaration of Interest:

The authors have no conflicts of interest to declare.

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