

Original Research Article

Pathophysiological and sociological factors associated with infections of *Mycoplasma hominis* and *Ureaplasma urealyticum* in patients who came at the Military Hospital Region No.1 of Yaoundé, Cameroon

ABSTRACT

Aims : The general objective of this work was to highlight the physiopathological and sociological factors linked to uro-genital infections with *Mycoplasma hominis* (*Mh*) and *Ureaplasma urealyticum* (*Uu*), in patients who made a consultation at the Military Hospital Region N ° 1 in Yaoundé.

Place and Duration of Study: The research was carried out at at the Military Hospital Region N ° 1 in Yaoundé between 2017 and 2019

Methodology: Patients whose mycoplasma examination was done, were included in the retrospective study (2017 and 2018) and patients obtained in 3 months (January to march 2019) during the prospective study were also included. Parameters of patients were collected. These parameters were analyzed using SPSS 23 software and graphs were plotted using GraphPad Prism 7 software.

Results: From the 231 patients included, 152 (65.80%) were infected with MUG. The pathophysiological factors associated with *Mh* and *Uu* infections are young people aged 25 to 31 (39.47%), women (98.68 %), the use of hormonal contraceptives (15.79%), as well as other pathogens associated with mycoplasmas. The prospective study shows that, of the 20 patients obtained, 10 (50%) were infected with MUG. The most infected patients were women with a frequency of 80% and the most affected age group was 25 to 31 years old. The sociological factors noticed in these patients are the status of being single (70.39%), the type of professions such as students (21.70%), housewives (17.76%), women in uniform (13.82%), women entrepreneurs (11.84%) and the non-use of condoms during sexual intercourse.

Conclusion: The pathophysiological and sociological factors influencing the development of mycoplasma infection show that one can control the occurrence of the infection as well as infertility problems and spontaneous abortion in Cameroon.

Keywords : *Mycoplasma hominis* ; *Ureaplasma urealyticum* ; pathophysiological factors ; sociological factors.

1. INTRODUCTION

Mycoplasma hominis (*Mh*) and *Ureaplasma urealyticum* (*Uu*) are bacteria that are part of the commensal flora and are the source of infections leading to spontaneous abortions and sterility. They are responsible for many diseases such as pneumonitis, asthma, fever, arthritis, myocarditis, meningitis, non-gonococcal urethritis, infertility and premature delivery although their natural history and these sequelae clinics remain vague [1]. Uro-genital mycoplasmic infections (UGM) are statistically associated with several factors favoring young age, low socio-economic status, sexual activity, the multiple partners [2]. More recent researches have shown a high rate of this infection during pregnancy and after childbirth [3]. The means of combating these species in Cameroon remains the use of antibiotics belonging to the families of cyclins, macrolides and quinolones [4]. Another very effective means is abstinence, the use of condoms, loyalty to a single partner and above all good hygiene.

An epidemiological study carried out in Meknes in Morocco in subjects of two sexes showed an overall prevalence of *Ureaplasma urealyticum* infections of 32.29%. The overall prevalence of *Mycoplasma hominis* infection was 9.38% [5]. Another study carried out in two large hospitals in Yaoundé (Cameroon) in 2015 on patients with inflammatory pelvic disease showed that the most common microorganisms were genital mycoplasmas with a frequency of 54.3% [6]. These bacteria have a place in the natural history of HIV infections.

Certain socio-pathological factors can negatively influence the behavior of mycoplasmas in the urogenital tract causing infections which lead to the resurgence of spontaneous abortions and infertility. In this context, given the involvement of *Mh* and *Uu* in abortions and infertility in both sexes according to Amirmozafari *et al.* [7], and since no studies have been carried out so far on the pathophysiological and sociological factors that can influence the occurrence of urogenital mycoplasmas, we proposed to conduct this study. This study aims to highlighting the pathophysiological and sociological factors linked to the infections of *Mh* and *Uu* in patients coming for consultations at the Military Hospital Region N°1 (MHRN°1) of Younde.

2. MATERIALS AND METHODS

2.1. Inclusion criteria and procedure

We included patients treated at HMRN N ° 1 in Yaoundé for whom the urogenital myogenoplastic examination had been requested and had been done. As well as the patients who had taken the mycoplasma examination between January 01, 2017 and December 31, 2018 in this hospital. The patient's list was obtained from each patient's medical record. The data collected are the results of the mycoplasma test, clinical manifestations, treatment, demographic data (age and sex), contraceptives, other associated infections, marital status and occupation.

2.2. Analysis of statistical data

All the data collected above were analyzed using SPSS 23 software. The statistical software for social sciences and graphics were plotted using graphPad Prism 7. This analysis allowed us to determine the frequencies of all the different parameters :

$$\text{Frequencies} = \frac{\text{Number of patients infected with mycoplasmas of a parameter}}{\text{Total number of infected patients}} \times 100$$

It should be noted that only the results of examinations positive used for each parameter. After calculating the frequency of complaints and treatments, a comparison was made between these two parameters and the results of the patient diagnosis obtained, for the conduct of the retrospective study. To highlight the pathophysiological factors, a comparison was made between the parameters age, sex, contraceptives and other associated pathogens and the results of the diagnosis of the patients obtained after calculation of the frequency of these four parameters. After calculating the frequency of marital status and occupations, a comparison was made between these two parameters and the results of patient diagnosis obtained for sociological factors.

2.3. Methods for detecting urogenital mycoplasmas

When the samples (biological fluids) that were taken from the cervix of women and from the urethra of men reached the examination room, the culture was done instantaneously. This culture was carried out using the Mycoplasma System Plus kit which made it possible to carry out the research, the semi-quantitative counting, the identification and the antibiogram of *Mycoplasma hominis* (*Mh*) and *Ureaplasma urealyticum* (*Uu*) isolated from clinical sample [8]. The presence of urogenital mycoplasmas was confirmed by the color change from yellow to red.

The semi-quantitative count of *Mh* and *Uu* is indicated by the color change from yellow to red in wells 1 -GR +, 2-GR ++, 3-GR +++.

The presence of *Mh* is indicated by the color change from yellow to red in the Arginine 4-ADC test well.

The presence of *Uu* is indicated by the color change from yellow to red in the Urea 5-UR test well.

The antibiogram of *Mh* and *Uu* is evaluated by observing the color change in the wells from 7-TE to 24-AZM.

Sample preparation

The swab that was used to collect the cervix or urethra was immersed in a 7 ml ampoule of physiological solution contained in the kit. After 5 min, it was carefully pressed against the wall of the bottle to disperse the material evenly in the physiological solution. The sample suspension that was used to seed the wells was thus obtained.

2.4. Seeding of the medium

The medium proper was then removed from its packaging and placed at room temperature (25 °C). The patient identification number and the start date of the examination were noted. 0.2 ml of the suspension of the clinical sample was then introduced into each well of the medium and covered with three drops of paraffin oil for microbiological use. Finally, the medium was covered and incubated at 37 ° C for 18 to 24 h. When the presence of *Mh* or *Uu* was suspected, incubation was carried out for 24 hours at the same temperature.

2.5. Results interpretation

The colorimetric shift (from yellow to red) of the micro-wells during alkalization of the medium after 24 h or 48 h of culture allowed the identification and counting of the germs expressed in units of color change per milliliter (UCC / mL). The concentration varied from 0 to more than 105 CCU / ml. The quantification of these germs makes it possible to incriminate their responsibility in an infection. The pathogenicity threshold for a urogenital infection was fixed at 105 CCU / ml, which made it possible to define a colonization for a value less than 104 CCU / ml for a colonization and an infection for a value greater than or equal to 104 CCU / ml. At the end of the incubation, we base ourselves on the color change of the wells 1 -GR + to 5-UR and we interpreted the results using a table found in the protocol (counting and identification of mycoplasmas / ureaplasmas) [8].

3. RESULTS

3.1. Retrospective section

3.1.1. Prevalence

Data collected at the gynecology department of the region's No. 1 military hospital in Yaounde show that of the 231 patients who took part to research using a culture of urogenital mycoplasmas, 152 (65.80%) (were infected with UMG including 15 (6.49%) infected with *Mycoplasma hominis* (*Mh*), 80 (34.63%) infected with *Ureaplasma urealyticum* (*Uu*) and 57 (24.68%) infected with both species. of the 231 patients, 228 were women with 150 infected and 03 were men with 02 infected.

3.1.2. Mycoplasma infection complaints

The patients that were consulted at HMRN ° 1 in Yaounde between 2017 and 2018 and for whom an examination for MUG's presence was made had various complaints. The most frequent species of mycoplasma was *Ureaplasma urealyticum* (*Uu*), then the simultaneous presence of these two species and finally *Mycoplasma hominis* (*Mh*). Table 1 below presents these complaints and their frequencies.

3.1.3. Treatment of mycoplasmas

At HMRN ° 1 in Yaoundé, the 05 most used drugs are : levofloxacin 30 (19.74%), doxycycline 29 (19.08%), ornidazole + ofloxacin 21 (13.82%) azitromycin 20 (13.16%) and 17 (11.18%) josamycin see Figure 1 below.

3.1.4. Pathophysiological factors associated with urogenital mycoplasmas (MUG).

The pathophysiological factors considered in this study were age, sex, taking hormonal contraceptives, other associated germs and pregnancy.

3.1.4.1. Physiological factors associated with urogenital mycoplasmas

Associated physiological factors were age and gender.

3.1.4.1.1. Age associated with mycoplasma infections

We are in the presence of a young population and the most representative age groups are those of 25-31 years 91 (39.39%), 18-24 years 48 (20.78%) and 32-38 years 44 (19.05%), the most sexually active population studied. It seems that the age group most infected with urogenital mycoplasma is 25 to 31 years with a frequency of 60 (39.47%).

3.1.4.1.2. Sex associated with mycoplasma infections

According to the results obtained, the majority of patients infected with mycoplasmas are women with a frequency of 150 (98.68%). This frequency is 64.94% in the total study population. Men are the least infected 2 (1.32%). Table 2 below presents these physiological factors.

3.1.4.2. Pathological factors associated with urogenital mycoplasmas

The pathological factors associated with UMG infection are taking hormonal contraceptives, other associated diseases and pregnancy.

3.1.4.2.1. Patients using hormonal contraceptives

Of the 31 patients using hormonal contraceptives, 15.79% (24) were infected with urogenital mycoplasmas (MUG). *Ureaplasma urealyticum* remains the predominant species in all cases.

3.1.4.2.2. Other mycoplasma infections

Several other infections are associated with infection with urogenital mycoplasmas. The most common infections seen in patients are : *Gardnerella vaginalis*, *Chlamydiae trachomatis* and *Candida albicans* infections. We had 32 (21.05%) patients with *mycoplasma-Gardnerella vaginalis* associations, 25 (16.45%) patients with *mycoplasma-Chlamydiae trachomatis-Gardnerella vaginalis* associations, 16 (10.53%) patients with mycoplasma associations -*Chlamydiae trachomatis*, 10 (6.58%) patients with mycoplasma-*Candida albicans* associations and 9 (5.92%) patients with mycoplasma-*Gardnerella vaginalis-Candida albicans* associations.

3.1.4.2.3. Pregnancy associated with mycoplasma infections

It appears that from the 04 pregnant women who carried out a mycoplasma test, 2 (1.32%) were infected with urogenital mycoplasmas. Table 3 below presents the pathological factors.

3.1.5. Sociological factors associated with UMG infection

The sociological factors associated with UMG infection were the status of a single person, the professions of entrepreneur, teacher, men in uniform and student, and the non-use of condoms during sexual intercourse.

3.1.5.1. Marital status associated with UMG infection

Singles were the most important part of this sample. They were the most infected urogenital mycoplasmas with a population of 107 (70.39%).

3.1.5.2. Professions associated with UMG infection

Our study takes place in an area where female students are more frequent 50 (21.65%) then housewives 43 (18.61%) and finally men in uniform 30 (12.99%). The results showed that, the most infected with mycoplasmas were female students 33 (21.71%), housewives 27 (17.76%), men in uniform 21 (13.82%) and female entrepreneurs 18 (11.84%).

3.1.5.3. Patients not using a condom

Out of the 165 patients who did not use condom, 105 (69.08%) were infected with at least one species of MUG. Table 4 below presents the sociological factors.

3.2. Prospective part

In the prospective study, we had 20 patients, therefore 12 women and 08 men. This study showed a frequency of UMG infection of 10 (50%) ; 02 infected with *Mh*, 06 infected with *Uu* and 02 infected with both species. Women constituted of the 80% most infected (08) and men of 20% least infected (02). The age group that was mostly affected was that of 25-31 year olds with a frequency of infection.

Table 1 : Complaints related to mycoplasma infections

Significant patient complaints	Sick (%)	<i>Mh</i> (%)	<i>Uu</i> (%)	<i>Mh</i> and <i>Uu</i> (%)
Infertility problem	69 (45,39)	8 (5,26)	36 (23,68)	25 (16,45)
Dysmenorrhea	68 (44,74)	10 (6,58)	38 (25)	20 (13,16)
Abundant leucorrhoea with color variations + or – fetid	44 (28,95)	8 (5,26)	23 (15,13)	13 (8,55)
Dyspareunia	38 (25)	6 (3,95)	19 (12,5)	13 (8,55)

Vulvar and/or vaginal pruritus	35 (23,03)	5 (3,29)	15 (9,87)	15 (9,87)
Repeated miscarriages	4 (2,63)	0 (0)	2 (1,32)	2 (1,32)

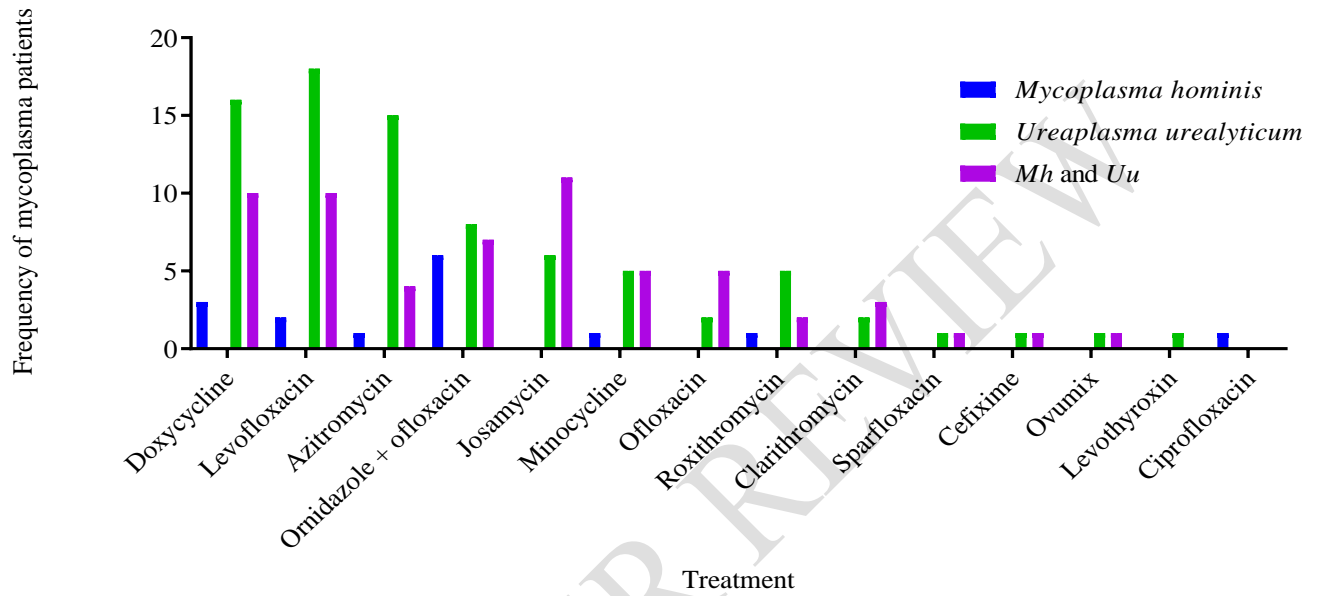


Figure 1 : Distribution of Mycoplasma patients according to treatment

Table 2 : Physiological factors associated with mycoplasma infections

Physiological factors	Sick (%)	<i>Mh</i> (%)	<i>Uu</i> (%)	<i>Mh and Uu</i> (%)
25 – 31	60 (39,47)	6 (3,95)	34 (22,37)	20 (3,16)
Feminine	150 (98,68)	15 (9,87)	79 (51,97)	56 (6,84)

Table 3 : Pathological factors associated with UGM

Contraception	Sick (%)	<i>Mh</i> (%)	<i>Uu</i> (%)	<i>Mh and Uu</i> (%)
No contraceptive	105 (69,08)	11 (7,24)	56 (36,84)	38 (25)

Hormonal contraceptives	24 (15,79)	2 (1,32)	11 (7,24)	11 (7,24)
Condom	20 (13,16)	2 (1,32)	11 (7,24)	7 (4,61)
No indication	3 (1,97)	0	2 (1,32)	1 (0,66)
Total	152 (100)	15 (9,87)	80 (52,63)	57 (37,5)
Other related infections	Sick (%)	<i>Mh</i> (%)	<i>Uu</i> (%)	<i>Mh</i> and <i>Uu</i> (%)
<i>Candida albicans</i>	10 (6,58)	1 (0,66)	6 (3,94)	3 (1,97)
<i>Chlamydiae trachomatis</i>	16 (10,53)	3 (1,97)	7 (4,61)	6 (3,94)
<i>Chlamydiae trachomatis</i> and <i>Gardnerella vaginalis</i>	25 (16,45)	4 (2,63)	9 (5,92)	12 (7,89)
<i>Gardnerella vaginalis</i>	32 (21,05)	3 (1,97)	18 (11,84)	11 (7,23)
<i>Gardnerella vaginalis</i> and <i>Candida albicans</i>	9 (5,92)	2 (1,31)	3 (1,97)	4 (2,63)
Patients	Sick (%)	<i>Mh</i> (%)	<i>Uu</i> (%)	<i>Mh</i> and <i>Uu</i> (%)
Pregnant woman	2 (50)	0 (0)	1 (25)	1 (25)

Table 4 : Sociological factors associated with UGM

Sociological factors	Sick (%)	<i>Mh</i> (%)	<i>Uu</i> (%)	<i>Mh</i> and <i>Uu</i> (%)
Single	107 (70,39)	11 (7,24)	54 (35,53)	42 (26,92)
Student	33 (21,71)	4 (2,63)	18 (11,84)	11 (7,24)
Housework	27 (17,76)	5 (3,29)	15 (9,87)	7 (4,61)
Woman in uniform	21 (13,82)	1 (0,66)	10 (6,58)	10 (6,58)
Contractor	18 (11,84)	1 (0,66)	10 (6,58)	7 (4,61)
No condom	105 (69,08)	11 (7,24)	56 (36,84)	38 (25)

4. DISCUSSION

In the retrospective study, patients complaining of infertility, dysmenorrhea, profuse leucorrhea, dyspareunia and vulvovaginal pruritus were the most infected with UGM. A study carried out on 44 patients complaining of infertility in Morocco showed that 15 (34.09%) were infected with UGM [9]. This result is significantly lower than ours for patients complaining of infertility. The rate of recurrent miscarriages noted in our study is very low compared to that of Cao *et al.* [10] Found in a study of 132 women complaining of spontaneous abortions and chorions where they had found frequencies of 44.7% and 33.3% for *Uu* and *Mh* respectively. These differences in results are due to the different populations, the high degrees of heterogeneity between the participants, the differences in detection methods, the types of samples studied and especially the low socio-economic levels of the population [5] ; [9]. In addition, the drugs most used in this study for the treatment of mycoplasmas belong to the family of fluoroquinolones. This means that there is usually bacterial resistance to first and second line drugs.

The pathophysiological factors associated with urogenital infections of *Mh* and *Uu* are young age, females, the use of hormonal contraceptives and the patient's immunological state (other associated diseases). Thus, an epidemiological study carried out in Morocco showed a rate of UGM infection in patients in the 26-35 age group 22 (42.31%) which is close to ours which was 60 (39, 47%) in the 25-31 age group. Another study conducted in Tehran in 2009 by Amirmozafari *et al.* [7] Showed 42 (20%) and 93 (44.3%) of the levels of *Mh* and *Uu* respectively isolated in women suffering from genital infection, so the average age is 31.9 years. This result is superior to ours. The rates of carriage of UGM obtained in our samples could be explained by the fact that patients in this age group are the most sexually active with irresponsible sexual behavior. This variability in the prevalence of rates reported in different countries could be due to the size of the samples, hygiene problems, socio-economic status that can push young people towards sexual disorders, alcohol and drugs, as well as " in the absence of regular screening, treatment and monitoring programs [7].

The retrospective study was conducted on 3 men against 128 women. From this study, it appears that the prevalence of UGM is 2 (1.32%) in men and 150 (98.78%) in women. Few Cameroonian studies have addressed the prevalence of UGM. However, our results on the prevalence of *Uu*, *Mh* and the co-infection *Mh + Uu* are higher in women and lower in men compared to those of Bouzaidi or that had found in men 8.33% ; 2.7% ; 2.7% for *Uu*, *Mh* and co-infection *Mh + Uu* respectively and in women 4% ; 0.5% ; 0.5%. Our results are different from those of Western countries but close to those observed in Tunisia, Morocco, Brazil or China. According to Bouzaidi [11] these differences in frequencies are due to different sample sizes, the types of samples used to perform the test, the populations studied and the countries.

We find in this study that 24 (15.79%) of patients using hormonal contraceptives are infected with MUG. It should be noted that patients using hormonal contraceptives are more sensitive to STIs [12]. The hormones that make up contraceptives behave like ovarian hormones, and these have an inhibitory effect on the immune system, thereby promoting infection by opportunistic microorganisms like UGM. According to our study, these contraceptives are used more by female students.

Other microorganisms isolated from UGM patients are responsible for immunosuppression. The most involved in our study population were *Gardnerella vaginalis* (*Gv*) which causes bacterial infections mainly in the gynecological mucosa. In our study, we observe a high prevalence of *Gv* associated with UGM. They are much more associated with Ureaplasmas than Mycoplasmas. Casari *et al.* [13] found a prevalence of *Gv* of 19.7% in a group of 396 women in Morocco therefore 3.79% were infected with *Uu* and 0.25% by *Mh*. Thus, the origin of bacterial vaginosis which is characterized by the replacement of the normal flore predominated by lactobacilli is *Gv*, bactericides so the species is Mobiluncus and UGM [13]. *Chlamydiae trachomatis* (*Ct*) is a strictly human pathogenic bacterium of worldwide distribution. In our study, we isolated it from 10.53% of patients infected with *Ct*-UGM. If not diagnosed in time, it is responsible for infections such as salpingitis, ectopic pregnancy and tubal infertility. In this study, 10 cases (6.58%) of *Candida albicans* (*Ca*) were isolated from patients infected with *Ca*-UGM. It is found in patients in the same way as UGM. It mainly causes fungal infections (candidiasis) in the digestive and gynecological mucosa. These infections weaken the immune system and thus promote the development of other opportunistic pathogenic species. Speaking of pregnant women, 2 (1.32%) are infected with UGM. Amine [5] study showed an infection rate of 1 (50%) in pregnant women. This result is superior to ours. The low enrollment rate observed in pregnant women suggests that this examination is not done regularly in these women. Pregnant women have a weak immune system and are therefore very susceptible to sexually transmitted infections.

It should be noted that the patients subjected to our study are mainly single. Among them, 107 (70.39%) are infected with urogenital mycoplasmas. This result is higher than that of Amine [5] where the rate of UGM infection in single patients was 4 (50%). She concluded that this infection rate could be explained by the fact that hygiene and prevention conditions are not really respected, unlike those reported by international studies carried out in Africa. It should also be remembered that not all of them are stable partners and that most of them are women. With regard to occupations, the high rate of infection among students, housewives, women in uniform and entrepreneurs may be linked to their irresponsible sexual behavior and poor hygiene. The multiplicity of sexual partners is therefore a risk factor among female students [14]. There are also hygiene problems with it, for example long nails which most often contain germs. In addition, housewives, who are generally married women, are like waste tanks during intercourse and most often do not use a condom. One can also observe in these hygiene problems for example the common towels in a house which a source of dissemination of germs, and the use of soaps for personal hygiene which is at the origin of the deterioration of the vaginal flora thus promoting infections [15]. As for entrepreneurs and women in uniform, they are constantly on the move and in turn meet new people. A high rate of infection is also seen in patients not using a condom. Unprotected sex increases the risk of sexually transmitted infections [4].

The results of the prospective study show that women are the most exposed to this infection and that the age group most affected is between 25 and 31 years old. These results go hand in hand with those of our retrospective study, with the only difference that the frequency in humans is 40% higher compared to 1.32% in the retrospective study. However, one might think that men very rarely go to gynecologists but discreetly consult other doctors.

5. CONCLUSION

In general, the pathophysiological factors associated with *Mh* and *Uu* infections are young age 25-31 years, female sex, use of hormonal contraceptives and the patient's immunological status (other associated infections). Sociological factors associated with *Mh* and *Uu* infections. Are celibate status and occupations such as students, housewives, women in uniform and entrepreneurs. Unprotected sexual intercourse also constitutes a sociological factor.

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