HPV with genital pathogens and antibiotics resistance genes among female sex worker

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Abstract

Context: Genital infections and Sexually Transmitted Infections (STIs) remain a real public health problem in the world predominantly in sub-Saharan Africa. The purpose of this study was to determine co-infection of HPV, *Neisseria gonorrhoeae* (*NG*); *Chlamydia trachomatis* (*CT*); *Mycoplasma genitalium* (*MG*) and *Trichomonas vaginalis* (*TV*) among female sex workers in West Africa and to search antibiotics resistance genes. This study could serve as a support for the management of patients infected.

Methods: The study took place in Ouagadougou in July 2019 and from June to July 2020. It was a cross-sectional study with descriptive and analytical aims.

A total of 182 samples from sex workers of West and Central African origins, were analyzed by real-time PCR and resistance genes by classical PCR after DNA extraction.

Data were entered and analyzed using the IBM SPSS software in its 21 version and Epi Info 6. Tables and figures were produced using IBM SPSS Statistics version 20 and Microsoft Excel 2007. Chi-square and fischer tests were used for comparisons Epi info version 7. with a significant difference for $p \leq 0.05$.

Results: These women, who came from nine different countries, were aged 17–50 years with an average age of $27.12 \pm 6.09$ years and had an average of $415.9 \pm 75.2$ sexual partners per year. HPV and vaginosis co-infection (*NG, CT, MG and TV*) was 85%. The prevalence of bacteria was: *NG* 13.74%, *CT* 11.54% and *MG* 11.54%. Among the HPV co-infections the most common were HPV/*NG* (15%), HPV/*MG* (12%), and HPV/*CT* (8%). The antibiotic resistance genes identified are: $bla_{QNR}$ 24%, $bla_{GES}$ 22%, $bla_{SHV}$ 17%, $bla_{CTX-M}$ 13%; $bla_{QNR}$ 1%.

Conclusions: This study showed that the majority of sex workers of West and Central African origin working in Ouagadougou were infected with multiple STIs. This confirms that the presence of genital infections and STIs remains a real public health problem. The scale of these infections and the detection of associated resistance genes require increased surveillance of the molecular epidemiology of these pathogens.

1. Introduction

For years, genital infections in women were considered a minor ailment, but nowadays, given the reality of their consequences, they represent a matter of great public health concern. They can facilitate infection with HIV and many other sexually transmitted infections (STIs) such as *Neisseria gonorrhoeae* (*NG*), *Chlamydia trachomatis* (*CT*), *Mycoplasma genitalium* (*MG*) and *Trichomonas vaginalis* (*TV*) (1–4). Sexually transmitted infections are still prevalent in developing countries, particularly those south of the Sahara. STIs also pose serious public health problems around the world because of their frequency and complications (5). According to the WHO, more than one million people a day worldwide contract STIs (6,7). The global burden of morbidity and mortality associated with sexually transmitted pathogens threatens the quality of life of populations, their sexual and reproductive health and the health of newborns and children. Similarly, STIs indirectly promote the sexual transmission of HIV and cause cellular changes that lead to certain cancers. STIs place considerable pressure on household budgets and national health systems in low- and middle-income countries and have a negative impact on the overall well-being of individuals (5–8). In Africa, the tropical climate is a paradise for vaginal pathogens that proliferate and cause endemic pathologies (1). Among the STIs encountered are *Chlamydia trachomatis*, *Neisseria gonorrhoeae*, *Mycoplasma genitalium* and *Trichomonas vaginalis* infections. *Chlamydia trachomatis* is often the most common agent of STIs. The majority of women are at risk (9–11). In 2016, according to the World Health Organization (WHO), an estimated 131 million people will contract chlamydia each year (12). Today, it is estimated that there are approximately 162 million people infected worldwide, 6 million of whom are blind (13). Chlamydia can, however, lead to an ascending disease of the female genital tract which can result in cervicitis, endometriosis and then salpingitis (14). As for *Neisseria gonorrhoeae*, it remains a pathogenic bacterium of considerable health risk especially in developing countries. According to studies, it has a prevalence of 14.3% and 16.8% (15, 16). On examination of the vaginal swab, vaginosis and greenish-grey purulent mucus in the endocervix are signs suggestive of gonococcal infection. It should be noted that many forms are asymptomatic: more than 60% of female urogenital localizations
and more than 90% of pharyngeal localizations have no translation (14). The three bacterial species: *Neisseria gonorrhoeae, Chlamydia trachomatis*, and *Mycoplasma genitalium* are able to cross the cervical barrier and infect glandular crypts, leading to endocervicitis. Acquired sexually, they infect the endocervix and at the same time, the urethra (17). To this effect, the objectives of the study were to characterize by real-time PCR the pathogenic germs such as *Neisseria gonorrhoeae, Chlamydia trachomatis, Mycoplasma genitalium* and *Trichomonas vaginalis*, responsible for STIs in female sex workers in West and Central Africa with known HPV status and to determine the prevalence of these pathogenic germs in the genital tract and also to detect antibiotics resistance genes. This study could serve as a support for the management of patients infected by these pathologies.

2. Material And Methods

2.1 Setting of the survey

The study took place in Ouagadougou in the Molecular Biology Laboratory of the Pietro Annigoni Center for Biomolecular Research (CERBA) in Burkina Faso. It was a cross-sectional study with descriptive and analytical aims, which took place in July 2019 and June to July 2020.

Data was collected using an individual collection sheet administered to each sex worker included. Statistical analyses were carried out using IBM SPSS software version 21 and Epi Info 6. Tables and figures were produced using IBM SPSS Statistics version 20 and Microsoft Excel 2007 for pivot tables using SPSS 25.0 software. Chi-square and File tests were used for comparisons using Epi info version 7. with a significant difference for $p \leq 0.05$.

2.2 Sampling

Following recruitment and obtaining written informed consent, each patient was placed in a gynecological position on the examination table. After highlighting the cervix and ano-genital areas using a disposable speculum, a simple inspection of the cervix was performed. Subsequently a cotton-tipped swab was inserted into the endocervix and ectocervix at the squamocolumnar junction area and rotated at least three times in a counterclockwise direction. Each patient's specimen was coded for anonymity. The collected specimen was placed in a transport medium (SACACE Amplification Kit Transport Medium) and stored at -20 °C pending DNA extraction.

This study consisted of the analysis of data from 182 female commercial sex workers in whom endocervical cervical sampling was performed in 2017 for determination of HPV infection status (18). Included were all women who met the recommended conditions for good vaginal sampling (VSP), who were required to give informed consent, and whose specimens after HPV testing were properly stored at minus eighty (-80 °C) for further molecular analyses including those in this study. The HIV status of the study population was known simply by patient response.

3. Molecular Characterization

3.1 DNA extraction

DNA extraction was performed at CERBA /LABIOGENE using the DNA-Sorb-A kit from SACACE biotechnologies® (reference K-1-1/A /100, lot 23B16A155), following the protocol provided by the manufacturer.

3.2 Real-Time PCR Amplification

The real-time PCR performed was multiplexed with four tubes for each sample. The reaction mixture yielded a volume of 25 µL, i.e. 10 µL of the volume of DNA and 15 µL of the preparation of PCR mix-1-FL, PCR-mix-2-FRT and polymerase (TaqF) (10 µL of PCR mix-1-FL, 5 µL of PCR-mix-2-FRT and 0.5 µL of polymerase (TaqF). The mixture was vortexed and distributed to each well. The program used is the one from Sacace™ for the urogenital "STD" assays proposed by the manufacturer. This amplification program was: 1 cycle of 95 °C for 10mn followed by 5 cycles of 95 °C for 05 s; 60 °C for 20 s; 72 °C for 15 s and finally 40 cycles of 95 °C for 05 s; 60 °C for 30 s and 72 °C for 15 s. An internal amplification control and negative and positive controls were used.
The principle of real-time PCR is based on the cycle-by-cycle monitoring of the enzymatic amplification reaction using a fluorescent reporter molecule capable of emitting, under well-defined conditions, fluorescent radiation whose intensity will be directly measured at some point during each PCR cycle (19).

### 3.3. Identification of resistance genes by classical PCR

For the detection of antibiotic resistance genes, specific primers were used (Table 1). The PCR was performed in a 20 µL reaction volume consisting of Master mix 5x HOT FIREPol@ Blend: 4 µL, Primer F: 0.6 µL, Primer R: 0.6 µL, H2O: 10.8 µL, DNA: 5 µL).

For the amplification of the desired genes several programs were used (Table 2).

Antibiotic resistance genes were detected by amplification using the GeneAmp System PCR 9700 thermal cycler (Applied Biosystems, California, USA). Migration was performed by electrophoresis on a 2% agarose gel at 120 volts for 40 min. The visualisation was possible using Ethidium Bromide (BET). A 100 bp molecular weight marker was used as a molecular weight index.

| SPECIFIC PRIMERS (5’- 3’) | RESISTANCE GENES | EXPECTED SIZES | REFERENCES |
|----------------------------|------------------|----------------|------------|
| TEM-F: ATAAAATTCTTGAAGACGAAA | bla TEM | 1000 Pb | (20) |
| TEM-R: GACAGTTACCAATGCTTAATCA | bla TEM | 1000 Pb | (20) |
| CTX-MF : GTTACAATGTGTGAGAAGCAG | bla CTX-M | 1000 Pb | (21) |
| CTX-M-R: CGGTTTCCGCTATTACAAC | bla CTX-M | 1000 Pb | (21) |
| PER-F; ATGAATGTCATTATAAAAGC | bla PER | 925 Pb | (22) |
| PER-R: AATTTGGGCTTAGGAGC | bla PER | 925 Pb | (22) |
| QNR A-F: ATTTCTCAGCCAGATTGT | bla QNR A | 846 Pb | (23) |
| QNR A-R : GATCGGCAAAGGTAGGTA | bla QNR A | 846 Pb | (23) |
| QNR B-F: GATCGTGAAGCCAGAAAGG | bla QNR B | 769 Pb | (23) |
| QNR B-R : ACGATGCCGTGGTATTTGTCC | bla QNR B | 769 Pb | (23) |
| QNR S-F: ACGACATTGTCAACTGCA | bla QNR S | 566 Pb | (24) |
| QNR S-R : TAAATTGGCACCCCTGTAGGC | bla QNR S | 566 Pb | (24) |
| SHV-F : ATG-CGTATATTCCGCTGTG | bla SHV | 875 Pb | (25) |
| SHV-R : TCAGCGTTGCCAGTGTC | bla SHV | 875 Pb | (25) |
| GES-F : ATGGCGTCTCACCCGAC | bla GES | 863 Pb | (26) |
| GES-R : CTAATTGTCCGTCGTCAGG | bla GES | 863 Pb | (26) |
Table 2
PCR program according to gene type

| Setting          | bla TEM | bla SHV | bla CTX-M | bla QNR A | bla QNR B | bla QNR S | bla PER | bla GES |
|------------------|--------|--------|----------|----------|----------|----------|--------|--------|
| Initial denaturation | 96 °C/5 mn | 96 °C/5 mn | 96 °C/5 mn | 95 °C/12 mn | 95 °C/12 mn | 96 °C/5 mn | 96 °C/5 mn |
| Denaturation      | 96 °C/1 mn | 96 °C/1 mn | 96 °C/1 mn | 95 °C/1 min | 95 °C/1 min | 95 °C/1 min | 96 °C/1 mn |
| Matching          | 58 °C/1 mn | 60 °C/1 mn | 96 °C/1 mn | 55 °C/1 min | 55 °C/1 min | 55 °C/1 min | 96 °C/1 mn |
| Elongation        | 72 °C/1 mn | 72 °C/1 mn | 72 °C/1 mn | 72 °C/2 min | 72 °C/2 min | 72 °C/2 min | 72 °C/1 mn |
| Final elongation  | 72 °C/10 mn | 72 °C/10 mn | 72 °C/10 mn | 72 °C/7 min | 72 °C/7 min | 72 °C/7 min | 72 °C/10 mn |
| Number of cycles  | 35     | 35     | 35        | 35        | 35        | 35        | 35     |

3.4. Interpretation of real-time PCR results

Interpretation of the results was performed using the Microsoft Excel Real-Time Program provided by the manufacturer. The sample result is invalid in the absence of any fluorescence signal. The result is valid if the Negative Amplification Controls do not have a positive fluorescence signal. The sample result is negative if all four tubes contain only positive signal in Cy5 (red). The specimen result is positive if the positive signal appears in Fam (green) and/or Joe (yellow) and/or Rox (orange) regardless of the Cy5 (red) result. *Neisseria gonorrhoeae* is detected on the FAM/Green channel, *Chlamydia trachomatis* on the JOE/Yellow/HEX/Cy3 channel, *Mycoplasma genitalium* on the ROX/Orange/Texas Red channel, *Trichomonas vaginalis* on Cy5.5/Crimson and IC DNA on the Cy5/Red channel.

3.5. Data analyses

Data were entered and analyzed using the IBM SPSS software in its 21 version and Epi Info 6. Tables and figures were produced using IBM SPSS Statistics version 20 and Microsoft Excel 2007. Chi-square and fisher tests were used for comparisons Epi info version 7. with a significant difference for p ≤ 0.05.

4. Results

4.1 Socio-demographic, Clinical and Biological Characteristics of the Study Population

Our study population consisted of 182 women aged 17 to 50 years with an average age of 27.12 ± 6.09 years. This group of female sex workers was composed of 9 nationalities namely: Burkina Faso, Nigeria, Cameroon, Benin, Togo, Senegal, Ghana, Côte d'Ivoire and Mali. The Nigerian nationality is the most represented with 43.96% of the total number of patients. It is followed by Burkina Faso with 43.41%. Senegalese nationality is the least represented with 0.55% of the study population. Nigerian women are 1.9 times more likely than Burkinabe women to contract an infection with OR 1.9 (CI 1.01–3.96) p = 0.06.

Single women were 160 or 87.91%. Married women came next with 7.14% or 13 women followed by 8 divorced women or 4.40%.
In our study population, 99 patients had a secondary level education and forty-one women had a primary level. 36 patients had no schooling while 6 women had a university level of education, i.e. 3.30%. The level of secondary schooling is the most represented (54.40%) and is also the most affected (30.76%) by the pathogenic germs encountered. (Table 3).

Table 3
Socio-demographic, clinical and biological characteristics of study population

| Pathogens Germs (NG, CT, MG) | Positive | Negative | P value | HPV |
|-------------------------------|----------|----------|---------|-----|
| N = 56 (%)                    | N = 126 (%) |          |         |     |
| AGES CLASSES                  |          |          |         |     |
| ≤21                           | 3 (1.6)  | 16 (8.8) | 0.002   | 13 (7.1) c |
| 21–30                         | 17 (9.3) | 101 (55.5)| 0.0001  | 63 (34.6) d |
| ≥30                           | 1 (0.5)  | 44 (24.2) | 0.0001  | 24 (13.2) e |
| SCHOLASTIC LEVEL              |          |          |         |     |
| Not attending school          | 7 (3.8)  | 29 (15.9)| 0.019   | 16 (8.8) c |
| Primary                       | 12 (6.6) | 29 (15.9)| NS      | 19 (10.4) |
| Secondary                     | 35 (19.2)| 64 (35.2)| 0.038   | 61 (33.5) d |
| University                    | 2 (1.1)  | 4 (2.2)  | NS      | 4 (2.2)  |
| SEX BUDDY                     |          |          |         |     |
| <=500                         |          |          |         |     |
| 53 (29.1) a                   | 116 (63.7)| 0.0001  | 94 (51.6) c |
| >500                          | 3 (1.6) b| 10 (5.5) | NS      | 6 (3.3) d |
| MARITAL SITUATION             |          |          |         |     |
| Unmarried                     | 49 (26.9) a| 111 (61.0)| 0.0001  | 91 (50.0) c |
| Married                       | 6 (3.2) b| 8 (4.4)  | NS      | 5 (2.7) d |
| Divorcees                     | 1 (0.5)  | 7 (3.8)  | NS      | 4 (2.2) e |
| NATIONALITIES                 |          |          |         |     |
| Burkinabè                     | 19 (10.4)| 60 (33.0)| 0.001   | 42 (23.1) c |
| Nigerian                      | 31 (17.0)| 49 (26.9)| NS      | 46 (25.3) d |
| Others                        | 6 (3.3)  | 17 (9.3) | NS      | 12 (6.6) e |
| Totals                        | 56 (30.8)| 126 (69.2)| 0.0001  | 100 (54.9) |

a – b: p < 0.001 c – d: p < 0.001

d – e: p < 0.001

NS = Not significant c – e: p < 0.001

4.2 HIV status of patients
Negative HIV status was reported by female sex workers in 92.86% of cases. While 3.85% of female sex workers reported HIV-positive status. In addition, 3.29% had an unknown serological status.

4.3 Symptoms associated with the infections found in this study

Symptoms associated with the STIs encountered in our study, 97 or 53.30% had reported the frequent presence of leucorrhoea with or without bleeding. 66 or 36.26% were not noticeable and 19 or 10.44% reported ulcerations in the vaginal area (Fig. 1).

4.4 Distribution of the population according to the prevalence and epidemiological estimate of the pathogenic agents encountered

In our study population, 56 women out of 182 were infected with at least one of the pathogenic germs encountered, either a prevalence of 30.70%. For the bacteria, 25 women or 13.74% of the study population were infected with NG; 21 women or 11.54% were infected with CT. Similarly, 21 women or 11.54% were infected with MG. Of the four pathogenic germs sought: Neisseria gonorrhoeae (NG); Chlamydia trachomatis (CT); Mycoplasma genitalium (MG) and Trichomonas vaginalis (TV), three were present at varying rates. Trichomonas vaginalis was not found in this study.

Co-infection with the microorganisms was encountered in ten (10) patients. Of these, six (6) patients were co-infected with CT and MG, three (3) patients were co-infected with NG and CT, and one patient (1) was infected with NG, CT and MG for a co-infection rate of 5.49 (Table 4).

Table 4: Frequency of pathogens germs from the manipulation by real time PCR

| Pathogens Germs | Positive (%) | Negative | P-value |
|-----------------|--------------|----------|---------|
| Only one Infection | Fishers exact test |
| MG | 21 (11.5) | 161 (88.5) | <0.0001 |
| CT | 21 (11.5) | 161 (88.5) | <0.0001 |
| NG | 25 (13.7) | 157 (86.3) | <0.0001 |
| TV | 0.0 | 182 (100) | - |
| Total (at least one infection) | 56 (30.7) | 126 (69.3) | <0.0001 |
| Co-Infections | | | |
| MG/CT | 6 (3.3) | 176 (96.7) | <0.0001 |
| MG/NG | 2 (1.1) | 180 (98.9) | <0.0001 |
| CT/NG | 4 (2.2) | 178 (97.8) | <0.0001 |
| MG/NG/CT | 1 (0.5) | 181 (99.5) | <0.0001 |

Legend

MG = Mycoplasma Genitalium; NG = Neisseria Gonorrhoeae; CT = Chlamydia Trachomatis; TV = Trichomonas Vaginalis

Taking into account the accumulation of pathogenic germs such as NG and or CT and or MG and or TV, we note a co-infection of these germs with HPV in 85% of cases. Table 5 shows the co-infection with HPV and NG, CT, MG, TV. There is a statistically significant difference between HPV/NG and HPV/NG/MG co-infections.

Tableau 5: Co infection HPV, NG, CT, MG and TV among sex professionals in our study
| Co infections | HPV+ N=100 | HPV- N=82 | P value |
|---------------|------------|------------|---------|
| NG+           | 15 (15%)   | 10 (85%)   | <0.0001 |
| NG-           | 85 (12.2%) | 72 (87.8%) | <0.0001 |
| CT+           | 8 (8%)     | 13 (15.9%) | 0.099   |
| CT-           | 92 (92%)   | 69 (84.1%) | 0.099   |
| MG+           | 12 (12%)   | 9 (11%)    | 0.830   |
| MG-           | 88 (88%)   | 73 (89%)   | 0.830   |
| NG+ CT+       | 2 (2%)     | 2 (2.4%)   | 0.759   |
| NG-CT-        | 98 (98%)   | 80 (97.6%) | 0.759   |
| NG+ MG+       | 2 (2%)     | 0 (0%)     | <0.0001 |
| NG-MG-        | 98 (98%)   | 82 (100%)  | <0.0001 |
| CT+ MG+       | 2 (2%)     | 4 (4.9%)   | 0.506   |
| CT-MG-        | 98 (98%)   | 78 (95.1%) | 0.506   |
| Vaginal infections | 85 (85%) | 69 (84.1%) | 0.874 |
| No Vaginal infections | 15 (15%) | 13 (15.9%) | 0.874 |

Legend: Vaginal infections: Cumulative NG/CT/MG/TV

### 4.5 Identified resistance genes

The resistance genes identified as a function of isolated bacteria are shown in Fig. 2. A total of eight (08) resistance genes were sought, of which five (5) tested positive. These are \( \text{bla}_{\text{CTX-M}} \) 13%, \( \text{bla}_{\text{SHV}} \) 17%, \( \text{bla}_{\text{QNR-B}} \) 24%, \( \text{bla}_{\text{QNR-S}} \) 1%, and \( \text{bla}_{\text{GES}} \) 22%. Of the bacterial species studied, Mycoplasma Genitalium was the only species to simultaneously host the five resistance genes found: \( \text{bla}_{\text{CTX-M}} \), \( \text{bla}_{\text{HVS}} \), \( \text{bla}_{\text{GES}} \), \( \text{bla}_{\text{QNR-B}} \) and \( \text{bla}_{\text{QNR-S}} \). The percentage of antibiotic resistance genes found was 77% with a predominance of \( \text{bla}_{\text{QNR-B}} \).

### 5. Discussion

By age, our study population consisted of 182 women aged 17 to 50 years with an average age of 27.12 ± 6.09 years. Our study population appears young compared to that of Mamadou et al. who found in 2000, during their study of 529 female professionals in Niamey, an average age of 29 years and age extremes ranging from 16 to 65 years (15). Our cohort is composed of 9 different nationalities. Most of the countries concerned border on Burkina Faso and therefore this has facilitated easy access to these women. They migrated to Ouagadougou because they found easy access and also because they did not want to be recognized in their own country as commercial sex workers. Many of them could be seeking easy gain and also lack the necessities for their survival and perhaps for that of their families. Many of them may have entered the sex trade for economic reasons, as it may be the only employment opportunity, or the one that pays the best. Others may have entered the sex trade because they are trapped in violence, trafficking or debt. Some, especially adults, may have freely chosen the profession of commercial sex work. Entry into the sex trade may have socially entrenched causes, referring to traditions, beliefs and norms that perpetuate gender inequalities (27).

The number of sexual partners and marital status (single and married) were statistically associated with the presence of the pathogens NG, CT, MG: (p ≤ 0.001). Also, age group, number of sexual partners and marital status were statistically associated...
with HPV carriage (p = 0.001). Indeed, in our study population, single women were dominant with 160 (87.91%), followed by married women with 7.14% or 13 women and finally 8 divorced women or 4.40%. Among the single women, 31.88% were infected, i.e. 28.02% of the study population. It could be said that the high number of single women could be justified by the fact that such women are more likely to engage in commercial sex work in order to provide for their needs.

As for the level of education of the study population, it was found that women with secondary education were in the majority and those with university education were in the minority. Ninety-nine (99) patients had a secondary level; 41 had a primary level. The education group most exposed to the infections encountered was those with secondary school education (62% of the infected population and 19.78% of the study population), followed by those with primary school education (20% of the infected population or 06.59% of the study population). These results are different from those of Mamadou et al. whose study population were sex workers who had parallel activities: bar waitresses and petty traders, and 27% of the study population were enrolled in school (15).

Of the four pathogens investigated in our study, *Neisseria gonorrhoeae* (NG); *Chlamydia trachomatis* (CT); *Mycoplasma genitalium* (MG) and *Trichomonas vaginalis* (TV), three were present at varying prevalence.

NG was positive at 13.74%; the prevalence of *Neisseria gonorrhoeae* was lower in our study than the 20.5% and 22% reported in other studies conducted in Cotonou, Benin in 1999 (28) and Dakar in 2000 (29). Another study in Niamey, Niger, reported a prevalence of 5.67% of N. gonorrhoeae which was isolated from 30 endocervical specimens, and identified only on direct examination by the presence of Gram-negative diplococci, in “coffee bean”, intra- and extra-leukocytic in 7 other cases. We note here that the Niamey study is different from ours due to the classical detection technique used and the results obtained (15).

The prevalence of CT infection in this study was 11.54% or 21 positive women. This rate is higher than those found in other studies in Cotonou in 1999 (5.1% (29)), Accra in 2000 (10.1%) (30) and Yaoundé in 1998 (12%) (31). Studies of low-risk women in different African countries have shown a prevalence of *Chlamydia trachomatis* infections ranging from 0.6% among women attending antenatal clinics in Tunisia to 5.5% in the general female population in the Gambia (32). However, the Mamadou study in Niger found higher results than ours (68.2% or 361/529 cases of active *Chlamydia trachomatis* infection in antigen detection) (15). Similarly, in Indonesia, studies among sex workers have shown prevalence rates of *Chlamydia trachomatis* infection ranging from 12–39%, while in Bangladesh, among sex workers in brothels, the prevalence rate was 15.5% (32). The prevalence of these STIs is higher in at-risk groups such as female sex workers than among women in the general population. Female sex workers are therefore said to be a reservoir for transmission to partners of those who use their service. Surveillance for these germs should therefore be strengthened.

MG also gave a prevalence of 11.54%. In their study, Mamadou et al. did not obtain *Mycoplasma Genitalium* but rather *Mycoplasma hominis*, which infected about 37% of the female sex workers in their study; it is involved in bacterial vaginosis sometimes complicated by endometritis and salpingitis (15). In this study, TV was not found. However, 10 patients were co-infected, including six (6) co-infections with CT and MG; three (3) co-infections with NG and CT and one co-infection with NG, CT and MG, i.e. a percentage of 5.49 co-infections. In a study conducted by Forward on the risk of co-infection with *Chlamydia trachomatis* and *Neisseria gonorrhoeae* in Nova Scotia, the co-infection rate was 31 out of 1495 patients for NG and CT (33). In another study by PETER et al, in patients infected with MG, co-infection of up to 40% with other microorganisms such as CT, NG or TV was found. This may draw our attention to co-screening for MG infection. The study population in this study is different from their study population, which justifies the difference in the exposure rate to the microorganisms (34, 35).

In terms of HIV status, most of the patients in this group of female sex workers had negative HIV status. While 6 had unknown HIV status, 7 were HIV positive. There is a low prevalence of HIV-positive status; however, several studies have shown that genital infections and STIs can increase HIV infectivity and/or transmission during sexual intercourse (3, 36, 37). This low rate of positive HIV status may be explained by the fact that most sex with female sex workers is protected or patients may not have answered truthfully to the question about their HIV status. In a study conducted in Niger in 2003, of 529 female sex workers enrolled, 30.1% were infected with HIV (15).
HPV-associated vaginosis co-infection was noted in 85% of cases in this study. This high rate of co-infection has the following combinations: HPV/NG; HPV/CT; HPV/MG; HPV/NG/CT; HPV/NG/MG and HPV/CT/MG with a statistically significant difference for HPV/NG and HPV/NG/MG. This association of STIs among sex workers in this investigation would increase the risk for their sexual partners. Indeed, although genital infections are often asymptomatic in women, the presence of a low genital infection would increase the risk of contracting HPV. For example, bacterial vaginosis is believed to be associated with high levels of anaerobic organisms that can damage the vaginal epithelium and thus increase the risk of HPV infection. In addition, Coudray and Madhivanan reported that bacterial vaginosis may increase the risk of many sexually transmitted infections (STIs) such as human immunodeficiency virus (HIV), *Neisseria gonorrhoea* (NG), *Chlamydia trachomatis* (CT), *Trichomonas vaginalis* (TV), and herpes simplex-2 virus (HSV-2) (38).

In addition, female sex workers constitute a "reservoir" of STIs, which would further increase the risk of genital infection for the general population. HPV co-infection with pathogens such as NG, CT, MG and TV have been reported by other authors such as Coudray and Madhivanan in 2020 in the USA and Lv and al. 2019 in China (38, 39).

HPV is the leading cause of cervical cancer, which is the leading cause of cancer death in women in Africa. It is therefore a real public health problem, especially since the circulating genotypes are not covered by existing and available vaccines (40). From the perspective of integrated control, it would be beneficial to perform multiplex PCR for the molecular diagnosis of these pathogens (HPV, NG, CT, MG, TV) both in sex workers and in the general population. Thus, the treatment of these pathogens detected would make it possible to break the chain of transmission of these genital infections for the health of populations.

With regard concerned to resistance genes, our study revealed the presence of several genes hosted by the different species studied. These resistance genes were also identified in previous studies in Burkina Faso but in other biological samples such as urine, faecal matter and pus (41–43). In contrast to our study in which *bla*<sub>QNR B</sub> is predominantly represented, the study by Metuor Dabiré and al.; in 2013, showed a predominance of *bla*<sub>CTX-M</sub> (43). The coexistence of several resistance genes observed in the present study was also detected in previous studies in Burkina Faso (44, 45).

This coexistence of several resistance genes within bacteria is believed to confer multidrug resistance to antibiotics.

**Conclusion**

Beyond all the control that has been done so far against genital infections and STIs, with the administration of antimicrobials in order to destroy their reserve foci, and to consider means of control for their definitive eradication, this study conducted showed that the majority of female sex workers of West and Central African origin, working in the city of Ouagadougou in Burkina Faso, were infected with NG, MG, CT and HPV. This confirms that the presence of genital infections and STIs remains a real public health problem. This magnitude of genital infections and STIs among female sex workers in West and Central Africa and the detection of associated resistance genes calls for increased surveillance of the molecular epidemiology of these pathogens. Notwithstanding the relatively high cost of PCR, it should be recommended for the detection of these pathogens and the identification of their resistance genes.

**List Of Abbreviations**

HPV: Human papillomavirus; CT *Chlamydia trachomatis*.
NG: *Neisseria gonorrhoeae*; MG: *Mycoplasma genitalium*
TV: *Trichomonas vaginalis*; PCR: Polymerisation Chain Reaction
TEM: Teimonera; CTXM: Céfotaxime-Munich
PER: Pseudomonas Extended Resistance; QNR A: Quinolone Resistance A
QNR B: Quinolone Resistance B; QNR S: Quinolone Resistance S
**SHV:** Sulfhydryl Variable; **GES:** Guyana Extended Spectrum Beta-lactamase

## Declarations

### Ethical Aspect

This study does not involve any risk to the health, physical and moral integrity of the participants. It is based on a study that was approved by the National Ethics Committee of Burkina Faso (Ref. N°2017-1026/MS/RCEN/DRSC).

**Consent for publication:** “Not applicable”

### Availability of data and material:

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

### Competing interests

All authors declare no conflict of interest.

### Financement: “Not applicable”

### Authors’ contributions:

- **Study concept and design:** SFT, TMZ, LS, CO and JS.
- **Sampling and Laboratory analysis:** SFT, TMZ, AMD, RAO, RYT, AKO, ATY, PAS, RI and FWD.
- **Statistical analysis and interpretation of data:** TMZ, DO and FWD.
- **Drafting of the manuscript:** FWD, DOY, LS, CO, SFT and JS.
- **Critical revision of the manuscript for important intellectual content:** SFT, FWD, DOY, CO and JS.
- **Administrative, technical, and material support:** CO, LS and JS.
- **Study supervision:** SFT and JS.

All authors have read and approved the manuscript.

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Figures
Figure 1

Associated signs of genital infection amongst sex professionals in our study

Figure 1

Associated signs of genital infection amongst sex professionals in our study
Figure 2

Antibiotics resistance genes as a function of bacteria