Original Article

Microbial Milieu and Antibiogram of Female Genital Infections in a Tertiary Health Facility in Nigeria

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Abstract

Background: Vaginal infections constitute a significant health challenge for women and lead to long-term complications if not promptly and adequately treated. We aimed to determine the prevalent organisms in vaginal infections in our women and the antibiotic susceptibility of the offending organisms.

Methodology: This was a prospective cross-sectional study of 635 consecutive women attending the Obstetrics and Gynaecology clinic of the University of Port Harcourt Teaching Hospital from 1st January 2017 to 31st December 2018. Data obtained were analysed using SPSS version 19.

Results: Four hundred and eight (64.3%) of the women were gynaecological patients, while the remaining 227 (35.7%) were obstetric patients. One hundred and ninety-one(30.1%) out of the total 635 study population did not have any growth of the organisms tested for in the culture assay, while the remaining 444 (69.9%) had. One hundred and forty-four of the women(22.2%) had growth of candida Albicans, 130 (20.5%) had staphylococcus aureus, 78 (12.3%) - Klebsiella species (spp), 48 (7.6%) - Escherichia coli, 30 (4.7%) - Pseudomonas species, 8 (1.3%) - Proteus species and 6 (0.9%) of the women had Streptococcus species. The commonest pathogen involved in vaginal infections in women was Candida albicans. The commonest bacterial isolate was Staphylococcus aureus and then Klebsiella species. The bacteria were susceptible to Ceftriaxone, Cefpodoxime, Cefotaxime, Cefixime, Ciprofloxacin, Dorepenem and Ampicillin. They were mainly resistant to cefuroxime, ceftazidime, Augmentin, Erythromycin and Meropenem.

Conclusion: Candida albicans is prevalent in our women. Staphyloccocus aureus is the commonest bacterial organism in women with vaginal infections. Empirical antibiotic treatment using Cetriazone, Cefpodoxime and Ciprofloxacin, available in our setting, may be beneficial while awaiting the culture results in women with suspected vaginal infections.

Keywords: Microbial; Antibiogram; Genital infections; Antibiotics; Nigeria.

Introduction

Globally, vaginal infections constitute a major health challenge for women.[1] Infection and subsequent inflammation of the vagina (vaginitis) is commonly encountered in obstetrics and gynaecological practice.[2] Candidiasis, bacterial

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How to cite this article: Amadi SC, Mkpe A, Awopeju ATO, Iwo-Amah RS, Olobuah AC. Microbial Milieu and Antibiogram of Female Genital Infections in a Tertiary Health Facility in Nigeria. Niger Med J 2022;63(5):348-355

vaginosis (BV) and Trichomoniasis have been observed to be majorly responsible for the vaginal infections.[2, 3, 4, 5]

Various aetiologic agents of vaginal infection result in a number of gynaecological complications and amplify Human Immuno-deficiency virus (HIV) and Herpes simplex virus-1 (HSV-1) transmissions.[6] Vaginal infections are associated with a significant risk of morbidity in women.[4, 6] If untreated, they can lead to pelvic inflammatory disease (PID), which can cause long-term sequelae, such as tubal infertility, ectopic pregnancy, reproductive dysfunction and adverse pregnancy outcomes (e.g. preterm labour and delivery and low birth weight).[4] Cervical dysplasia, increased risk of post-operative infection, HIV and Herpes simplex virus (HSV) -1 acquisition and transmission are associated with vaginal infections.[2, 7, 8, 9, 10]

In low-income and developing countries like Nigeria, screening and treatment of symptomatic sexually transmitted infections (STIs) are based on syndromic case management (SCM) (without culture for the causative organism) because of the unavailability of inexpensive point-of-care diagnostic assays.[10, 11] Introduction of additional parameters in the syndromic diagnosis of non-viral sexually transmitted infections in low-resource settings and hence improved management has been advocated but still far-fetched.[11]

Overuse and misuse of antibiotics in STIs are not new; the emergence of antimicrobial or multidrug-resistant organisms is well reported by several study groups worldwide.[12, 13, 14, 15, 16, 17]The prevalence of multidrug-resistant infections varies widely across different regions of the world. Still, some of the highest levels of infection have been found in low income, middle-income, and underdeveloped countries.[8, 9] The reasons are complex and include poor quality of health services, high burden of disease, and lack of accessible, accurate and confirmed diagnostic assays, regulatory oversight and overuse of antibiotics, inappropriate dosing, and lack of knowledge about the risks of microbial resistance.

Abnormal vaginal discharge due to bacterial organisms in our environment commonly manifests as sexually transmitted infections with their sequelae. In our setting, most clinicians adopt the syndromic case management approach for aetiologic diagnosis and targeted effective treatment due to a lack of resources (equipment, funds, personnel and patient-related factors). Hence, broad-spectrum antibiotics (commonly Ceftriazone - intravenously, oral Cefuroxime, Metronidazole, Augmentin and Ciprofloxacin) are usually commenced on the patient's first visit while awaiting the result of the vaginal or cervical swab microscopy/culture/sensitivity (m/c/s). Unfortunately, the result may take 72 hours or more to be available.

This study thus set out to prospectively review the results of the high vaginal swabs (HVS) and endocervical swabs (ECS) microscopy, culture and sensitivity (MCS) to ascertain the prevalent pathogens in our patients with suspected vaginal infections and determine the antibiotic susceptibility of the bacterial organisms. The findings of this study will add to the existing body of knowledge and aid clinicians in choosing appropriate broad-spectrum antibiotics for the empirical treatment of vaginal infections before the availability of vaginal culture results. That will reduce the problem of antibiotic resistance and its sequelae and form the basis for further research.

Materials and Methods

Study design, timing and location

The study was a prospective cross-sectional study carried out in the gynaecological and antenatal clinics of the University of Port Harcourt Teaching Hospital (UPTH) from 1st January 2017 to 31st December 2018. The University of Port Harcourt Teaching Hospital (UPTH) is an 800-bed tertiary hospital located in Port Harcourt, Rivers State, Nigeria. This health institution provides all levels of health care services for Rivers State and the Niger Delta area's catchment states. The antenatal and gynaecological clinics hold from Monday to Friday, and a team oversees each day. The patients are registered and sent in for a consultation at the presentation. In addition, the UPTH controls a modern Medical Microbiology Department equipped with facilities for culture assay.

Determination of the sample size

The sample size was calculated using the Sample size formula for cross-sectional studies with a definite outcome.

$n = Z_{\alpha/2}^2 \; P(1-P) / d^2$ where

$Z_{\alpha/2}$ = Standard average deviation at 95% confidence interval = 1.96.

P – Expected proportion in population based on previous studies. P was taken to be 50% due to paucity of Nigerian studies that dealt with the subject matter.

d = Absolute error or precision = 0.05.

Therefore $n = 1.96^2 \times 0.5 (1-0.5) / 0.05^2$

$= 3.8416 \times 0.5 \times 0.5 / 0.0025 = 384.16$

The required number of patients for the study was, therefore, 384.16. Giving allowance for an attrition rate of 10%, the final power for the study was 10/100 x 384 + 384 = 422.56. However, 635 patients were recruited for the study.

Inclusion Criteria

Patients with suspected vaginal infections in the gynaecological and obstetric clinics of the UPTH were eligible for the study.

Exclusion Criteria

They include patients who had been on antibiotics within seven days of the swab collection date, those who douched their vaginas with chemicals, and those who did not give consent.

Participant Recruitment

The patients who met the inclusion criteria were recruited into the study using the consecutive sampling method until the sample size was attained.

Data collection

Research assistants (5) and a dedicated medical laboratory scientist in the Medical Microbiology Department were recruited and trained for the study. The research assistants and the researchers consecutively recruited the patients who met the inclusion criteria daily from the gynaecological and antenatal clinics. The Medical Microbiologist supervised the specimen analysis in the Medical microbiology laboratory. The recruited participants were counselled, and informed consent was obtained. The data in their antenatal cards and folders (age of the patients, the clinic attended, presenting symptoms), were transferred into the proforma. High vaginal and endocervical swabs (HVS and ECS) were taken aseptically from the consenting consecutive obstetrical and gynaecological patients who were suspected of having vaginal infections. The researchers or the research assistants collected the specimen as described below. The result of the microscopy, culture and sensitivity were also entered in the proforma.

Procedure for HVS/ECS

The woman was asked to empty her bladder. The equipment needed for the procedure (sterile gloves, bivalve speculum, water-based lubricant, good light source [torch or lamp], sterile bacterial swab stick [blue top]) and a Chaperon were ensured to be in place. She was then taken to the procedure room, where both auditory and visual privacies were maintained. The chaperone guided the participant to undress and lay down in a dorsal position on the examination bed in the procedure room. The researcher's hands were washed, and sterile gloves were worn. The bivalve speculum was lubricated with a water-based lubricant. The speculum was gently passed into the vagina, and the cervix was located with the aid of a good light source. Using the sterile swab stick, the specimen was collected from the posterior vaginal fornix (for HVS) or the endocervix/cervical canal (for ECS). The swab stick is immediately put in the tubular container and sealed.

The HVS or the ECS was labelled with the woman's name, Hospital number, date and clinic. The microbiology request form was completed. The patient's drug/antibiotic allergies, if any, were entered in the clinical information section on the request form so that the laboratory could perform appropriate sensitivity testing with a positive result. The HVS/ECS was placed in a biohazard plastic bag and sent to the medical microbiology laboratory. The speculum was washed (reusable speculum) and sent to Central Sterilising Department (CSD) for re-sterilisation. The disposable speculum, when used, is discarded appropriately. Other materials / equipment used were appropriately disposed of. The

hand gloves were removed, and the hands were washed. The specimen collection was documented in the patient's folder or her antenatal card.

Statistical analysis

Data were collected and analysed on a Statistical package for social sciences software SPSS version 19. Simple proportions were used in the descriptive analysis. Quantitative data were summarised and presented as mean and standard deviation, while qualitative data were presented as numbers and percentages. The Chi-square test was used to compare categorical variables with a p-value of 0.05 or less taken as being statistically significant.

Ethical consideration

The study was carried out in compliance with the international ethical guidelines for biomedical research involving human subjects. Ethical approval was obtained from the UPTH ethics committee. Participants were counselled, and informed consent was obtained. All the information collected from individual patients was available for clinical use and research purposes. Privacy rules were maintained, and confidentiality was observed at all levels of dealing with patients' data.

Results

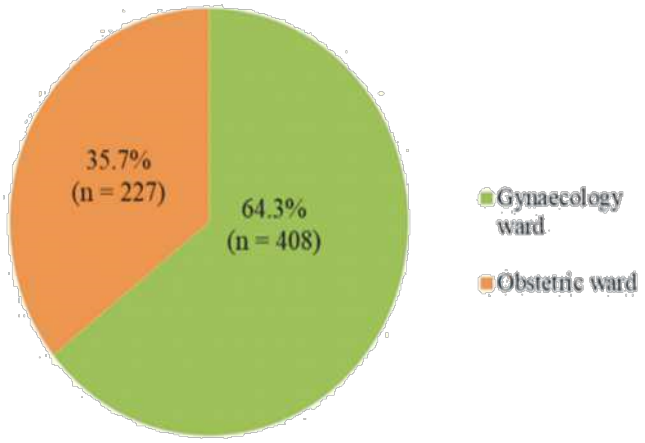

Figure 1: Clinical ward/Unit of study participants

The data of the six hundred and thirty-five (635) patients who underwent culture assay of high vaginal/endocervical swabs were involved in this study. Four hundred and eight (64.3%) of the women were gynaecological patients, while the remaining 227 (35.7%) were obstetric patients. (Fig 1)

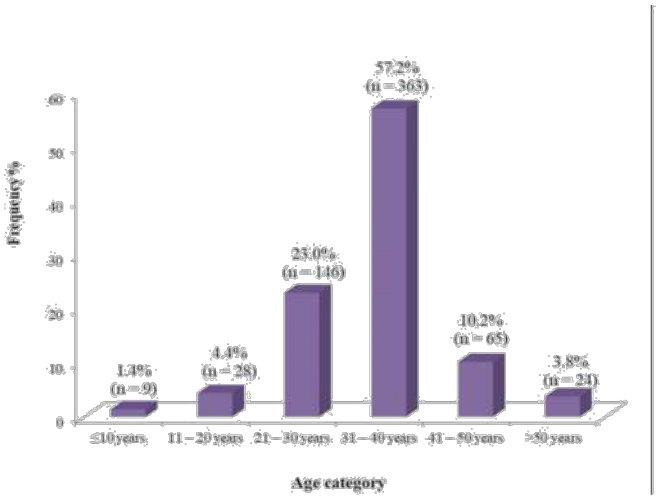

Figure 2: Age category of study participants

The Mean age of the study participants was 33.31±9.17years. (Fig 2)

Table 1: Microbial isolates from the HVS and ECS

	Test specimen		
	HVS	ECS	Total
Isolates	n (%)	n (%)	n (%)
No growth	185 (6.9)	6 (3.1)	191 (100.0)
Candida albicans	141 (97.9)	3 (2.1)	144 (100.0)
Staphylococcus aureus	127 (97.7)	3 (2.3)	130 (100.0)
Klebsiellaspp	72 (92.3)	6 (7.7)	78 (100.0)
Escherichia coli	47 (97.9)	1 (2.1)	48 (100.0)
Pseudomonas aeruginosa	27 (90.0)	3 (10.0)	30 (100.0)
Proteus spp	7 (87.5)	1 (12.5)	8 (100.0)
Streptococcus spp	6 (100.0)	0 (0.0)	6 (100.0)
Total	612 (96.4)	23 (3.6)	635 (100.0)

HVS = High vaginal swab; ECS = Endocervical swab.

One hundred and ninety-one 191 (30.1%) out of the total 635 study population did not have any growth of the organisms tested for in the culture assay, while the remaining 444 (69.9%) had. The general distribution of micro-organism growth according to the test specimen among the study population was as follows: 144 (22.2%) out of the total 635 patients had growth of candida Albicans, 130 (20.5%) had staphylococcus aureus, 78 (12.3%) - Klebsiellaspp, 48 (7.6%) - Escherichia coli, 30 (4.7%) - Pseudomonas species, 8 (1.3%) - Proteus species and 6 (0.9%) of the women had Streptococcus species (Table 1). The age distribution of tested patients and those with significant microbial growths were as shown in table 1.

Table 2: Age distribution of tested patients and those with significant microbial growths.

	Gynaecological patients		Obstetric Patients	
category (years)	Patients tested n (%)	Patients with Significant growth (%)	Patients tested (%)	Patients with Significant growth (%)
	2 (0.5)	1 (0.5)	7 (3.1)	5 (5.0)
11-20	19 (4.7)	6 (3.0)	9 (4.0)	4 (4.0)
21-30	83 (20.3)	33 (16.6)	63 (27.8)	23 (22.8)
31-40	235 (57.6)	119 (59.8)	128 (56.4)	61 (60.4)
41-50	49 (12.0)	27 (13.6)	16 (7.0)	5 (5.0)
> 50	20 (4.9)	13 (6.5)	4 (1.8)	3 (3.0)
Total	408 (100.0)	199 (100.0)	227 (100.0)	101 (100.0)

The participants in the age range of 30 – 40 years had a more positive culture for microbial isolates (Table 2).

Table 3: Sensitivity of bacterial isolates to drugs

Antibiotics	Bacterial isolates					
	Staphylococcus aureus n (%)	Klebsiella spp n (%)	Escherichia coli n (%)	Pseudomonas aeurugmosan (%)	Proteus spp n (%)	Streptococcus spp n (%)
Ampicillin	118 (90.9)	39 (50.0)	16 (33.3)	15 (50.0)	8 (100.0)	0 (0.0)
Gentamicin	56 (43.5)	51 (65.6)	30 (61.8)	17 (55.0)	5 (66.7)	2 (25.0)
Moxifloxacin	95 (73.3)	70 (90.0)	36 (75.0)	15 (50.0)	(0.0)	0 (0.0)
Ofloxacin	76 (58.3)	62 (79.2)	30 (61.5)	15 (50.0)	8 (100.0)	4 (67.0)
Cefactor	93 (71.4)	39 (50.0)	16 (33.3)	17 (55.0)	0 (0.0)	3 (50.0)
Cephalexine	104 (80.0)	13 (16.7)	5 (10.0)	15 (50.0)	MD	MD
Cefuroxime	69 (51.4)	28 (35.4)	17 (35.3)	6 (20.0)	MD	0 (0.0)
Ceftazidime	66 (50.9)	48 (61.5)	26 (54.2)	23 (75.0)	8 (100.0)	3 (50.0)
Ciprofloxacin	80 (61.3)	47 (60.6)	40 (82.4)	25 (82.4)	8(100.0)	4(67.0)
Cefotaxime	122 (93.8)	20 (25.0)	33 (69.2)	20 (67.0)	7 (90.0)	5 (83.0)
Cloxacillin	19 (14.3)	*7 (17.0)*	*0 (0.0)*	15 (50.0)	4 (50.0)	0 (0.0)
Augmentin	32 (25.0)	*3 (4.5)*	*5 (11.1)*	6 (20.0)	2 (25.0)	0 (0.0)
Erythromycin	54 (41.7)	*11 (14.3)*	*2 (4.8)*	2 (6.7)	3 (42.9)	2 (25.0)
Cefixime	96 (73.9)	31 (40.0)	29 (60.0)	18 (60.0)	6 (75.0)	3 (50.0)
Ceftriazone	85 (65.5)	41 (53.1)	33 (69.0)	25 (84.6)	8 (100.0)	4 (67.0)
Piperacillin	61 (47.3)	39 (50.0)	10 (20.0)	27 (90.0)	6 (75.0)	5 (83.0)
Meropenem	62 (48.0)	65 (83.3)	30 (62.5)	25 (85.7)	4 (50.0)	6 (100.0)
Cefepime	113 (86.7)	55 (70.0)	34 (71.4)	30 (100.0)	8 (100.0)	6 (100.0)
Dorepenem	119 (92.3)	51 (65.0)	44 (90.9)	26 (85.7)	8 (100.0)	6 (100.0)
Amoxycilin	106 (82.4)	5 (6.7)	32 (66.7)	15 (50.0)	8 (100.0)	4 (67.0)
Clotrimazole	NT	NT	NT	NT	NT	NT
Tetracycline	NT	NT	NT	NT	NT	NT
Streptomycin	NT	NT	NT	NT	NT	NT
Nitrofurantoin	NT	NT	NT	NT	NT	NT
Nalidixic Acid	NT	NT	NT	NT	NT	NT
Chloramphenicol.	NT	NT	NT	NT	NT	NT

NT – Not tested; MD = Missing data

The most prevalent bacterial isolate (Staphylococcus aureus) was sensitive to Ampicillin, Amoxycillin, Ceftriaxone, Cefotaxime, Cefixime, Ciprofloxacin, Ofloxacin, Dorepenem and Moxifloxacin. However, it was primarily resistant to Augmentin, Cefuroxime, ceftazidime, Erythromycin and Meropenem. All the other organisms in the study were resistant to Cefuroxime, Erythromycin and Augmentin but sensitive to Gentamycin, Ofloxacin, Cefotaxime, Ceftriaxone, Meropenem, and Dorepenem (Table 3).

Table 4: Hierarchical Stratification of antibiotic sensitivity of the isolated micro-organisms

S/N	Isolated Micro-organisms					
	Staphylococcus aureus Antibiotic (N%MSTI)	Klebsiellaspp Antibiotic (N%MSTI)	Escherichia coli Antibiotic (N%MSTI)	Pseudomonas aeurugmosa Antibiotic (N%MSTI)	Proteus spp Antibiotic (N%MSTI)	Streptococcus spp. Antibiotic (N%MSTI)
1	**Cefotaxime** 122 (93.8)	**Dorepenem4** 4 (90.9)	**Ciprofloxacin4** 0 (82.4)	**Cefepime30** (100.0)	Ampicillin 8 (100.0)	**Meropenem** 6 (100.0)
2	**Doripenem** 119 (92.3)	Moxifloxacin 70 (90.0)	Moxifloxacin 36 (75.0)	**Piperacillin** 27 (90.0)	**Ofloxacin 8(100)**	**Cefepime** 6 (100.0)
3	**Ampicillin** 118 (90.9)	**Meropenem 65 (83.3)**	**Cefotaxime** 33 (69.2)	**Meropenem** 25 (85.7)	**Ceftazidime 8(100)**	**Piperacillin** 5 (83.0)
4	**Cefepime** 113 (86.7)	**Ofloxacin 62 (79.2)**	Gentamicin 30 (61.8)	**Doripenem** 25 (85.7)	**Ciprofloxacin 8(100)**	Cefotaxime 5 (83.0)
5	**Amoxycillin** 106 (82.4)	**Cefepime 55 (70.0)**	Ofloxacin 30 (61.5)	**Ceftriazone25** (84.6)	**Ceftriaxone 8(100)**	Ofloxacin 4 (67.0)
6	**Cephalexine1 04 (80.0)**	**Amoxycilin3** 2 (66.7)	**Ceftazidime** 26 (54.2)	**Ciprofloxacin** 25 (82.4)	**Cefepime 8(100)**	**Ciprofloxacin 4** (67.0)
7	Moxifloxacin 95 (73.3)	Gentamicin 51 (65.6)	Ampicillin 16 (33.3)	**Ceflazidime23** (75.0)	**Doripenem 8(100)**	**Ceftriazone** 4 (67.0)
8	**Cofactor** 93 (71.4)	**Ceftazidime** 48 (61.5)	**Cofactor** 16 (33.3)	**Cefotaxime** 20 (67.0)	**Cefotaxime** 7 (90.0)	**Amoxycilin4** (67.0)
9	Cefuroxime 69 (51.4)	Ciprofloxacin 47 (60.6)	**Piperacillin** 10 (20.0)	**Cefixime** 18 (60.0)	**Cefixime** 6 (75.0)	**Cofactor** 3 (50.0)
10	**Ceftriaxone** 85 (65.5)	Ceftriaxone 41 (53.1)	Augmentin 5 (11.1)	Gentamicin 17 (55.0)	**Piperacillin** 6 (75.0)	**Ceflazidime** 3 (50.0)

N%MSTI = number and percentage of Micro-organism sensitive to it.

Discussion

In this study, most patients had microbial isolates in their culture assay. The absence of microbial growth in some of the culture assays of the patients may be due to prior treatment of the patients with potent drugs, as is mostly obtainable in our setting or due to the actual absence of offending organisms in the specimen. The complaints warranting the culture assay may also have been due to viral aetiology or other organisms which were not assayed in this study.

Candida albicans were the most prevalent pathogen in the patients in the study. That is similar to findings from other studies from Vietnam, Ethiopia, Nepal and Bangladesh where candida Albicans was the most prevalent organism.[1, 3, 18, 19] The findings in the present study differ from the findings in other studies in India and Shandong.[20, 21] The difference may be due to difference in the hygiene practices obtainable in different environments or cultural practices.

The most typical bacterial isolate in the study was Staphylococcus aureus, followed by Klebsiellaspp, Escherichia coli, Pseudomonas aeruginosa, Proteus spp and Streptococcus spp in decreasing order of frequency. That is similar to the study by Mumtaz et al. in which Staphylococcus aureus was the most prevalent vaginal pathogen.[22] This finding is at variance with the conclusion of a survey by Tariq et al. in which Enterococcus species were the most pervasive vaginal pathogen.[23] The difference may be accounted for by the different hygienic protocols in the other study populations and the common pathology for which the investigation was requested in each study setting.

This study did not test the sensitivity of the predominant fungal vaginal infection (candida Albicans) to antifungal agents. The commonly used empirical antibiotics in the study setting include Ceftriaxone, Cefuroxime, Augmentin, Ciprofloxacin, Ofloxacin, Gentamycin, Erythromycin and, in severe cases, Meropenem. The finding of significant resistance of Staphylococcus aureus to Cefuroxime, Gentamycin, Augmentin, Erythromycin, and Meropenem is a critical point to note in the choice of antibiotics in our setting while waiting for the result of the culture test.

The sensitivity of the predominant vaginal bacterial pathogen (Staphylococcus aureus) and the other isolated micro-organisms to different antibiotics was shown in table 4, in which the preferred antibiotics were outlined in decreasing order of sensitivity from up to down. Among the drugs staphylococcus Aureus was sensitive were Ceftriaxone, Cefotaxime, Ciprofloxacin, Ofloxacin, Amoxicillin, and Doripenem. Therefore, the drugs should be preferable as empirical antibiotics of choice while waiting for the culture test result in women with suspected vaginal infections. However, these findings are similar to those from other studies.[3, 22, 24]All the different organisms in the study were highly resistant to Cefuroxime, Erythromycin and Augmentin. So, using these drugs in cases of suspected vaginal infections in our setting may not be very beneficial.

The Klebsiella species was highly resistant to Cefixime, Amoxicillin and many other drugs. All the other organisms were sensitive to Cefotaxime, Ceftriaxone, Ofloxacin and Dorepenem; applying these drugs as empirical treatment for patients with suspected vaginal infections in our setting may produce a good clinical outcome for the patient. These patterns of antibiotic sensitivity are similar to findings by other researchers.[23]They are different from results in other studies.[22] The differences between bacterial susceptibility to antibiotics in this study and other studies may be due to the antibiotic usage in the different study settings and the bacterial resistance to those commonly used drugs.

Conclusion

This study suggested that some patients who had vaginal swab culture assays may not have infections. Vaginal candidiasis was the most common vaginal infection in our setting. The bacterial pathogen's antibiogram suggested that using Ceftriaxone, Cefotaxime, Ofloxacin and Dorepenem as empirical antibiotics while waiting for culture test results will reduce the morbidity associated with vaginal infections in our environment.

Regular research to check the antibiogram of organisms is important to ensure that appropriate potent drugs are used for empirical treatment of infections in places where culture tests are not done at all or the culture results needed to determine the causative organism of any infection requires up to 72 hours or more for the assay result to be available.

Limitations

This was a single Centre study. Nonetheless, the findings of this study, within limits of acceptable error, may be generalisable to the entire population as the UPTH is located in the metropolitan city of Port Harcourt and attends to patients from the rural

and urban areas of the neighbouring States within the Niger Delta region.

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