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A study of female genital swabs in a Nigerian Tertiary Hospital

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ABSTRACT

Objective: To detect some common microbial agents of vaginal discharge and improve the current syndromic management of abnormal vaginal discharge. **Methods:** A prospective study of female genital swabs collected from obstetrics and gynecology units of Aminu Kano Teaching Hospital, Kano, Nigeria and an analysed of microscopy, culture and sensitivity in the medical microbiology and parasitology laboratory of this hospital were conducted from December 2007 to December 2008. Data on epidemiologic indices were collected from the patients, using structured interviewer-administered questionnaires. **Results:** Eight hundred and forty *Candida* species were detected, constituting 60% (n=840) of 1 400 female genital discharge samples of microbial etiology in a total of 2 000 female genital samples received. The distribution of vaginal candidiasis was the highest in young adults aged 21 to 30 years with 43%(n=360) of the total 840 cases. Pregnant women that presented with vaginal candidiasis constituted 40% (n=360) of the total 840 cases. Other risk groups included the immuno-suppression with 24% (n=202), group on hormonal therapy with 15% (n=126) and broad spectrum antibiotics users with 16% (n=134). **Conclusions:** The results show that *Candida* is the most common cause of vaginitis and vulvo-vaginal candidiasis followed by *C. albicans* in the young adults aged 21 to 30 years, pregnant mothers, immuno-suppression, contraceptive and broad spectrum antibiotic users. Proper management of vaginal candidiasis and vulvo-vaginal candidiasis is recommended especially among the risk groups in order to avoid complications and reduce HIV transmission.

1. Introduction

Vaginal candidiasis(VC) is a common type of vaginitis, a gynecologic disorder that manifests with an odorless curdy white discharge ("cottage cheese") in the female lower reproductive tracts with pruritus, irritation, dysuria or dyspareunia[1]. It is a common complaint among women of different age groups in any society whether or not they are sexually active. Some studies have shown high preponderance of VC in infective vaginal discharge among which studies included 52.5% and 80% isolation rate respectively, of *Candida* species[1,2] .

Risk factors for VC are factors that do not seem to be a direct cause of the disease, but seem to be associated in some way. Having a risk factor for VC makes the chances of getting a condition higher but does not always lead to VC. Also, the absence of any risk factors or having a protective factor does not necessarily guard one against getting VC[3]. Some risk factors for VC include pregnancy, poorly controlled diabetes, oral contraceptive, antibiotics, immune suppression, douches, perfumed feminine hygiene sprays, topical antimicrobial agents, tight clothing, tight under-wears, thyroid disorders and corticosteroid[3].

VC is caused by the fungus *Candida albicans (C. albicans)* in approximately 85% of cases, while other species such as *C. glabrata, C. krusei*, *C. tropicalis* and *C. stellatoidea* rarely cause vaginitis[4]. *Candida* species are usually of endogenous origin and may be transmitted by sexual partners. Changes in the vaginal environment are usually necessary before the organism can induce pathological effects.

Although VC is both treatable and mild, when left untreated, is a possible risk for acquisition of HIV/AIDS as well as other complications[5]. Other complications include pelvic inflammatory disease, infertility, ectopic pregnancy, pelvic abscess, menstrual disorders, spontaneous abortion and premature birth. It is now well established that the presence of infective vaginal discharge greatly facilitates transmission and acquisition of HIV between sexual partners[6].

Therefore, there is a need for prevention, early diagnosis and prompt treatment of this common condition especially among the risk groups, in order to avert the complications

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and reduce the transmission of HIV. Laboratory support is necessary for a differential diagnosis or to confirm the clinical diagnosis of VC.

The purpose of this study was to investigate VC among female patients attending Aminu Kano Teaching Hospital Kano, north-western Nigeria, in order to achieve an effective medical management of this condition.

2. Materials and methods

This descriptive study was carried out from December 2007 to December 2008 in Aminu Kano Teaching Hospital, Kano Nigeria. Consent of the patients was received and they were assured of strict confidentiality of responses. Structured interviewer-administered questionnaire was then introduced. Questionnaire included serial number, date, age, address, educational status, marital status, occupation, last menstrual period, hormonal therapy(yes/no)(if yes, type?), antibiotics(yes/no)(if yes, type?), chronic illness(yes/no) (if yes, type?), douching ?(yes/no).

Data were collected from the patients using an interviewer-administered questionnaire. Endo-cervical and high-vaginal samples were collected from the patients for laboratory analysis.

2.1. Laboratory procedure

Endo-cervical and high vaginal swabs were collected following aseptic precautions[4]. The genital swabs were immediately sent to the genital bench of medical microbiology and parasitology laboratory, Aminu Kano teaching hospital where they were processed according to standard procedures[4]. Infection with *Candida* species was diagnosed by microscopy of a saline mount, Gram-stained smear of material from the vagina and colonial growth on Sabouraud's dextrose agar.

A saline mount of the vaginal discharge specimen was covered with a cover-slip, and examined microscopically at ×400 magnification, not only to detect yeast cells, but to exclude the presence of trichomonads and clue cells. Yeasts are round ovoid cells, 4ìm in diameter, showing typical budding (blastoconidia). Yeasts can easily be recognized on a Gram stained smear as Gram- positive cells.

Antibiotic treated selective Sabouraud dextrose agar (SDA) was used as a growth medium for the isolation of *Candida* species. After inoculation of the clinical specimen, plates were incubated at 36 ℃ for 2 days. Colonies of yeast cells are opaque white to creamy.

A simple test for the identification of *C. albicans* is the germ tube test. A colony was emulsified in 0.5-1 mL of sterile serum and incubated at 35-37 ℃ for 4 hours. *C. albicans* form germ tubes, short lateral hyphal filaments without any constrictions. It was examined every half hour for formation of germ tubes. A complete identification of *Candida* species was done by means of sugar assimilation test (auxanogram). The yeast was grown on a basal carbohydrate-free medium supplemented with essential vitamins and the test sugar. Each of the five test sugars, namely glucose (positive control), sucrose, trehalose, lactose and raffinose was dispensed into each plate. Plates were incubated at 30 ℃ for 48 hours. Growth produces an opacity in the medium and indicates the ability of the isolates to assimilate a sugar.

Trichomonas vaginalis was diagnosed by microscopy of a saline mount for the actively motile, spear shaped flagellates. *Gardnerella vaginalis*, an agent of bacterial vaginosis, was diagnosed by Whiff test and the evaluation of Gram stained vaginal smear at oil immersion power(×1 000) objective for Clue cells, usually representing at least 20 percent of vaginal epithelial cells.

Cervical specimens were Gram-stained and cultures were inoculated on plates of Chocolates and Thayer-Martins (Oxoid) media and incubated at 37 ℃ in a moisturized candle extinction jar for 24 to 72 hours. *Neisseria gonorrhoeae* was identified by typical colonial morphology, reactions to Gram-stain, positive oxidase test, and sugar fermentation. The antibiotic sensitivity of isolates was tested by the agar diffusion method on chocolate agar plates using oxoid multi-discs with standard antibiotic concentrations. The samples collection, transportation and processing including microscopy, culture and biochemical tests were carried out according to recommended standard[4].

The results were analyzed using SPSS 11.0 statistical software; chi-square(x^2) was used to compare association between proportions and P-values<0.05 were considered significant at 95.0% confidence level.

Approval of the study protocol was obtained from the Ethics Committee of Aminu Kano Teaching Hospital.

3. Results

Eight hundred and forty *Candida* species were detected, constituting 60% of 1 400 female genital discharge samples of microbial etiology in a total of 2 000 female genital samples received. *Candida* species were arranged according to the age groups of the patients ranging from 0-60 years. In the age ranged 0-10 years, 10 (1.1%) out of the 840 *Candida* species were detected, in age group of 11-20 years 206 (24.5%) *Candida* species were recorded, 360 (42.9%) aged 21-30 years, 200 (23.8%) aged 31-40 years, 60 (7.1%) aged 41-50 years and only 4 (0.5%) aged 51-60 years. The peak age bracket at risk was 21-30 years which constituted 360 (42.9%). There were multiple isolates in 40 genital samples (Table 1).

Pregnant women that presented with VC constituted 40% (n=336) of the total 840 cases. Other risk groups included the immunosuppression with 24% (n=202), group with hormonal therapy 15% (n=126), broad spectrum antibiotics users 16% (n=134) and undetermined factors 5% (n=42) (Table 2). In a population of 202 immunosuppressed patients with abnormal vaginal discharge, 102 (50%) with HIV/AIDS, 60 (30%) with diabetes mellitus, 20 (10%) with tuberculosis and 20 (10%) with cancer.

4. Discussion

VC was a leading cause of abnormal vaginal discharge in the study constituting 60% (n=820) of the 1 400 female genital discharge of microbial causes out of the 2 000 female genital samples. The result was similar to some earlier studies which recorded 52.5% and 80% respectively[1,2]. Predominance of candidiasis in the study was in the group aged 21-30. The age decade of 21-30 is the most sexually active age group with highest risk of pregnancies, indulgence in family planning pills and immunosuppression due to HIV/AIDS. The inflammation of the vagina, as in any inflammatory STI, increases the risk of acquisition of HIV[3,7]. Candidiasis is not usually sexually transmitted, though male contacts should be seen, firstly, if they have symptoms, and secondly, if the woman is having recurrence.

C. albicans isolated was 708(84%) out of the total 840 *Candida* species while Non-albicans contributed 16% (n=132). The result was in keeping with other studies, which

Table 1
Distribution of *Candida* organisms in the infective female genital discharge.

Age group in years	Abnormal female genital discharge	Determined microbial causes	Microbial Organisms		
			Candida albicans	Non-albicans	Other Microbial agents
0–10	20	12	8	2	2
11–20	440	346	186	20	140
21–30	780	558	300	60	198
31–40	540	420	160	40	220
41–50	180	60	50	10	–
51–60	40	4	4	–	–
Total	2 000	1 400	708	132	560

** 20 samples had multiple isolates.

Table 2
Distribution of VC in relation to pregnancy, hormonal therapy and broad spectrum antibiotics.

Age in years	VC	Pregnant women	Hormonal therapy	Antibiotics	Immune-Suppress ed
0–10	10	–	–	–	10
11–20	206	76	30	24	52
21–30	360	200	60	10	80
31–40	200	60	36		40
41–50	60	–	–	34	56
51–60	4	–	–	–	4
Total	840(100%)	336(40.0%)	126(15.0%)	134(16.0%)	202(24.0%)

reported that vulvo-vaginal candidiasis(VVC) is created by the fungus *C. albicans* in approximately 85% of cases, with *C. glabrata* being responsible for the remaining 15%[4]. An explanation for the predominance of *C. albicans* in the study may be that these species are more predominant than other *Candida* species in the environment and hence more in human body. More importantly, it is their favorable survival ability in a depressed human immune system.

Pregnant mothers with VVC in the study constituted 40% of the total cases seen. Some studies have implicated pregnancy as an important risk factor for vaginitis[2,4]. The relationship between pregnancy and VC indicates increase in hormonal influences and alteration in vaginal PH.

Immunosuppressed patients recorded 24% risk in the study. Systemic condition such as diabetic mellitus, HIV/AIDS, organ transplants and any chronic debilitating illness can increase the woman's chances of developing VVC[3,5]. Depressed cell mediated immunity provides a favorable condition for growth of *Candida* species such as in HIV/AIDS, where as dysfunction of neutrophils and monocytes favors candidal growth in diabetes mellitus[6,7].

Broad spectrum antibiotic users posed a 16% risk to VC in the study. Antibiotics and vaginal douching suppress normal bacterial flora and allow *Candida* organisms to proliferate. Of interest is that sulfonamide decrease neutrophil intracellular killing of *Candida* organisms and tetracyclines and amino glycosides have been shown to decrease neutrophil phagocytosis[7,8].

Patients on hormonal therapy contributed 15% risk to VVC. The study included patients on oral or injection or implants or intrauterine contraceptive device (IUCD). Hormonal therapy leads to an inter-play between hormonal influences and alteration in vaginal PH[8]. The normal mature vagina has a PH of 4.0.

Limitation in the study includes inability to identify most of the non-albicans species owing to lack of some test sugars.

We recommend prevention, early diagnosis and prompt treatment of VC and VVC especially among the risk groups in order to avert the complications and reduce HIV transmission.

In conclusion, the result shows that *Candida* species has assumed the role of the most common cause of vaginitis, with *C. albicans* as the most prevalent species. Vulvo-VC was common in young adults of age range of 21–30 years, pregnant mothers, immunosuppressed, contraceptive and broad spectrum antibiotic users. Proper management of VC and VVC is recommended especially among the risk groups in order to avert complications and reduce HIV transmission.

Conflict of interest statement

We declare that we have no conflict of interest.

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