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Research Article

Candida albicans Infection and Associated Risk Factors among Pregnant Women Attending Antenatal Clinic at Federal Medical Centre, Birnin Kudu, Jigawa State

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ABSTRACT

Candida albicans affects the quality of life and when present in pregnant women it causes complications especially in individuals that are immunocompromised and or poor attitude to hygiene. A total of 60 high vaginal swap (HVS) samples were taken from pregnant women and analyzed for the presence of *C. albicans* species. Swap sticks were used in the collection of white patches found around the posterior fornix of the vagina and inoculated in Sabouraud Dextrose Agar and incubated at 37°C for 24hours. Slides were prepared and viewed under reduced light microscope using ×10 and ×40 objective magnification. Germ tube test was carried out to distinguish *C. albicans* species from other *Candida species*, using test tubes containing 0.5ml human serum. The result shows that those in their second and third trimester have higher prevalence 12 (40.0%) of *C. albicans* and the secondi gravid 16 (53.3). Similarly, those in the age group of 15-20 years have prevalence 20 (66.7). Although, not significant (P> 0.05), third trimester, use of tights and age were important predisposing factors as revealed by the adjusted odd ratio of 3.3, 2.2 and 6.9 respectively. High prevalence of *Candida albicans* was observed and factors such as hygiene habit, age of the women, use of antibiotics, trimester and dressing among women played important role as risk factors. Destruction of epithelia cells of the vagina was significantly correlated with *C. albicans* infection.

Keywords: Candida albicans; Pregnant Women; Prevalence; Risk Factors; Jigawa State

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INTRODUCTION

Candida species are aerobic thin-walled, small yeast measuring about 4 to 6 micrometers and reproduce asexually by budding (Cheesbrough, 2000). They are members of the class Ascomycetes (Kwon-Chung,

1994; Guarro *et al.*, 1999). *Candida species* are ubiquitous yeasts and common residents of mucosal surfaces of the oral cavity, the gastrointestinal and the urogenital tracts of humans and animals (Hauman *et al.*, 1993; Wade, 1993; Prescott *et al.*, 2002; Holland *et al.*, 2003). In addition to their

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clinical importance, *Candida species* have many industrial applications, mainly in food industry (Maheswari *et al.*, 2016).

More than thirty years ago, only a few pathogenic species of *Candida* were known and 20 species have been identified inrecent years as being associated with human and animal infections (Ellis *et al.*, 2003; Fanci and Pecile, 2005; Hildago and Vazquez, 2005; Sirnivasan and Kenneth, 2006).

Candida species are part of the lower genital tract flora in 20-50 % of healthy asymptomatic women (McClelland et al., 2009; Akah et al., 2010). Carrier rates are higher in women treated with broad spectrum antibiotics (Singh, 2003), pregnant women, diabetic women (Donders, 2002) women with HIV/AIDS (Akah et al., 2010). Candida albicans is both the most frequent colonizer and responsible for most cases of VVC (Singh, 2003). The only well proven predisposing factors are pregnancy, diabetes mellitus (CDC, 2002), and the use of broad-spectrum antibiotics (Mardh et al., 2002) as well as oral contraceptive with high estrogen content (Akah et al., 2010) which kill the good and beneficial bacteria, allowing yeast overgrowth, poor dietary habits and poor personal hygiene. Many practitioners believe that nylon underwear and tight insulating clothing predispose to vaginal candidiasis by increasing the temperature and moisture of the perineum (Nwankwo et al., 2010; Alliet al., 2011).

A study among African women wearing tight clothes reported a higher prevalence of *Candida albicans* in Vulvovaginal candidiasis than those wearing loose clothing (Alliet al., 2011).

Poorly supported risk factors include use of sponge, intrauterine devices (IUDS), diaphragms, condoms, orogenital sex, douching and intercourse (Mardh *et al.*, 2002, Reed *et al.*, 2003) and diet with high glucose content (de Leon *et al.*, 2002; Akah *et al.*, 2010).

Infection due to *Candida* species accounts for approximately 80% of all fungal infections of the immunocompromised (Smith, 2004). *Candida* species have been implicated in cases of abortion, arthritis, osteomyelitis, endophthalmitis, endocarditis, myocarditis, fungemia, meningitis and peritonitis which are major causes of mortality in both tertiary care centres and community hospitals (Fanello*et al.*, 2006; Heljic *et al.*, 2005). Heljic *et al.* (2005) identified the transmission of *Candida species* from the maternal vagina and hands of hospital personnel as being responsible for invasive

candidiasis in neonates which is associated with high mortality. *Candida species* are the second most common cause of urinary tract infections and the fourth most common cause of nosocomial blood stream infections, which in turn are associated with considerable mortality (Ellis *et al.*, 2003; Smith, 2004; Fanello*et al.*, 2006). It has been reported that up to 75% of sexually active women experienced symptomatic vaginal candidiasis (Schroppei *et al.*, 1994; Lachke *et al.*, 2003).

MATERIALS AND METHODS

Experimental site

The experiment was conducted in Federal Medical Centre Laboratory, Birnin-kudu which is situated along Kano-Bauchi Road which links the north-western to the north-eastern states and extends up to neighboring countries of Cameroon and Chad. It has a catchment area that includes four states viz; Jigawa; Kano, Bauchi and Yobe.

Population study

The population study comprised of 60 pregnant women with complete data that came for antenatal check-up at federal medical center Birnin Kudu, Jigawa state (FMC) from the month of May-July 2021.

Sample Size

Collection of 60 samples were arrived at using the formula of Charan and Biswas, (2013).

 $n = 1.96^2 \times expected prevalence (1 - expected prevalence)$

 d^2

d = Precision Rate = 5%; allowing attrition rate of 5%

 $d^2 = (5\%)^2 = (0.05)^2 = 0.0025$

Expected prevalence = 4%

 $n = 1.96^2 \times 4\% (1 - 4\%)$

0.0025

 $n = 3.8416 \times 0.04 (1 - 0.04) = 100$

0.0025

Collection of data

The socio-demographic data of the patients such as age, sex, patient's history were recorded with the aid of a questionnaire.

Ethical Clearance and Informed Consent

Before the commencement of the research, ethical clearance was with assigned number FMC/HREC/APP/CLN/001/201 was obtained from Health Research Committee, Federal Medical Centre, Birnin Kudu, Jigawa State. Informed consent was obtained from each person that participated in the study.

Selection Criteria

Patients for study were selected based on the following criteria informed.

- Informed Consent: The patients that gave their consent and meet other selection criteria were selected.
- Place of residence: Pregnant women resident in Birnin Kudu locality who attend antenatal clinic at FMC Birnin Kudu were selected.
- iii. Drug Treatment: Those who are on antifungal agents were excluded, while those who are on other antibiotics treatment were selected.

Collection of specimen

High virginal swap (HVS) was obtained by a qualified nurse who has a knowledge on HVS collection from each participant using sterile swap sticks. The sample was collected using ID number of study participant. Specimens were collected from symptomatic and apparently healthy individuals. HVS was inoculated in a medium or directly smeared on a clean slide using an inoculation loop or swab. Slides were covered with cover slips and were placed in an incubator for growth.

Culture medium

All culture media was prepared in accordance with manufacturer's prescription. The basic culture media used in isolating clinical *Candida species* is Sabouraud Dextrose Agar (SDA). The swap stick was inoculated on the Sabouraud Dextrose Agar (SDA) plates. The media plates were covered and then placed in an incubator for 24hours. A little drop of normal saline was added in the swap stick container, shaken and smeared on a clean glass slide, covered with a cover slip and viewed under microscope using x40 magnification.

Morphological Test

Presumptive identification of *C. albicans* was done with the germ tube test, where the clinical samples were incubated in 0.5mL human serum for 2 to 3 hours at 37°C. Filamentous extension from yeast cell and no constriction at the neck was considered as germ tube positive, isolates with no extension or extensions with constrictions at the neck were considered germ tube negative.

C. albicans was confirmed when short, slender, tubelike structures (germ tube) were observed under the microscope (Mackenzie, 1962). However, Lactophenol cotton blue together with Potassium hydroxide solution (10-20% KOH and 10% glycerine) stains were used as aid to view yeasts in specimens.

Staining

A sterilized wire loop was used to pick colony from the cultured plate smeared on a clean glass slide, dried and fixed by passing it over a Bunsen flame and flooded with 1%Crystal violet stain and allow for 2minutes. The smear was decanted and flooded with Lugol's iodine for 1minute, the smear was washed with clean tap water and decant excess water, decolorize with acetone until no more colour comes out. The smear was flooded with dilute Carbon fuchsin for two minutes, washed with tap water and viewed under microscope.

RESULTS

Women in second and third trimester have higher prevalence 12(40.0), with P-value 0.67 which showed no significant difference. Also, those in their secondi gravidity have high prevalence 16(53.3) than those in primigravid and multigravida, hence the P-value 0.60 shows that there's no significant difference of *C. albican* in association with gravidity. Those under the age group of 15-20 have high prevalence of 20(66.7), with no significant difference of *C. albicans* in association with age (Table 1).

Preliminary findings showed that those who use water only to wash their vagina have prevalence of 16 (53.3) with no significant difference (P-value 0.18) for *C. Albicans* in terms of association with hygiene. Also, those pregnant women that use cotton type of pants have prevalence of 22 (35.0) with no significant difference (P-value 0.67) for *C. albicans* in terms of association with the type of pants used. Lastly those that do not experience itching and excessive discharged have prevalence of 18 (60.0) with significant difference (P-value 0.02) for *C. albicans* in terms of association with itching experience and excessive discharge (Table 2).

A very strong significant positive correlation between the destruction of epithelia cells of the vagina and the presence of yeast exited. Similarly, positive correlation with no significant difference in the relationship between the destruction of epithelia cells of the vagina and knowledge about *Candida albican* infection also existed (Table 3).

Pregnancy in the second and third trimester (AOR, 95%CI, 2.8 and 3.3 respectively) appear to be a risk factor for *C. albicans* infection compared to those in their first trimester. Similarly, the secondi gravid and multigravida (AOR, 95%CI, 1.1 and 1.6 respectively) appear to be predisposing or risk factors of *C. albicans* infection compared to the primigravid pregnant women in the study. Also, pregnant women that use tight frequently were more predisposed to *C. albicans* infection than those that don't use tight frequently (AOR, 95%CI, 2.2), soare

those pregnant women that use antibiotics more predisposed to *C. albicans* infection than those that do not use antibiotics. The pregnant women that uses water and soap are more predisposing to C. albicans infection than those that use water only. Also in the type of pants used, those women who use nylon were more predisposed to C. albicans infection than those that use cotton and any type of pants. Furthermore, pregnant women that have the knowledge of C. albicans were more predisposed to C. albicans infection than those that have no knowledge of *C. albicans*. Also pregnant women that have previous history of infection were more predisposed to C. albicansthan those that don't have any previous history of the infection. Those pregnant women under the age categories of 33-38 years were more predisposed to C. albicans than other age categories (Table 4).

Table 1. Prevalence of *C. albicans* and its association with Trimester, Gravidity and Age

Variables	Negative	Positive	Total
Trimester			
First	4(13.3)	6(20.0)	10(16.7)
Second	15(50.0)	12(40.0)	27(45.0)
Third	11(36.7)	12(40.0)	23(38.3)
χ^2		0.77	
P-value		0.67	
Gravidity			
Primigravid	8(26.7)	5(16.7)	13(21.7)
Secondi gravid	13(43.3)	16(53.3)	29(48.3)
Multigravida	9(30.0)	9(30.0)	18(30.0)
χ^2		1.00	
P-value		0.60	
Age			
15-20	20(66.7)	20(66.7)	40(66.7)
21-26	6(20.0)	8(26.7)	14(23.3)
33-38	4(13.3)	4(13.3)	6(10.0)
χ^2		0.95	
P-value		0.62	

Table 2. Prevalence of *C. albicans* and its association with Hygiene, Types of Pants, Itching and Excessive Discharge from the Vagina Germ tube test

Variables	Negative	Positive	Total
Hygiene			
Use of water only	21(70.0)	16(53.3)	37(61.7)
Use of water and soap	9(30.0)	14(46.7)	23(38.3)
X^2	1.76		
P-value	0.18		
Types of pants			
Any type	4(13.3)	2(6.7)	6(10.0)
Cotton	21(70.0)	22(35.0)	43(71.7)
Nylon	5(16.7)	6(20.0)	11(18.3)
X^2	0.78		
P-value	0.67		
Experience itching			
and Excessive			
Discharge			
No	26(86.7)	18(60.0	33(73.3)
Yes	4(13.3)	12(40.0)	16(26.7)
X^2	5.45		
P-value	0.02		

Table 3. Relationship between Destruction of Epithelia cells of the vagina and Presence of Yeast and Knowledge of *Candida albicans* infection

Variables	Destruction of Epithelia Cells of the Vagina	
Presence of Yeast	0.66**	
P-value	0.00	
Knowledge of <i>C. albicans</i>	0.10	
P-value	0.45	

^{**}Very strong positive correlation

Table 4. Predisposing Factors of *C. albicans* Infection among Pregnant Women attending ANC in Birnin Kudu

Variables	AOR (95%CI)	P-Value
Trimester		
First	1.0	
Second	2.8(19.7-0.4)	0.27
Third	3.3(1.9-0.0)	0.24
Gravidity		
Prim gravid	1.0	
Secondi gravid	1.1(1.2-0.0)	0.08
Multi gravid	1.6(9.8-0.2)	0.57
Frequency in the use of tight		
Not Always	1.0	
Always	2.2(1.2-0.0)	0.09
Use of Antibiotics		
No	1.0	
Yes	0.1(0.9-0.0)	0.04
Hygiene Cleanliness		
Use of water only	1.0	
Use of water and soap	0.1(2.1-0.0)	0.05
Type of Pants		
Any type	1.0	
Cotton	0.1(2.1-0.0)	0.16

Nylon	0.4(2.8-0.0)	0.39
Knowledge of Candida		
No	1.0	
Yes	0.1(1.1-0.0)	0.06
Previous History of Infection		
No	1.0	
Yes	1.1(6.8-0.1)	0.89
Age Categories		
15-20	1.0	
21-26	5.6(53.6-0.5)	0.13
33-38	6.9(87.1-0.5)	0.13

DISCUSSION

The findings this study did ascertained the prevalence of Candida albicans among pregnant women attending antenatal care clinic at the FMC, Birnin Kudu, Jigawa state. The study became necessary because documents regarding factors responsible for prevalence of Candida albicans among pregnant women had not been reported at least in Birnin Kudu and its environs. The mean age of the study participants was 22.8 years, which is contrary to, a higher mean age of 31.5 years reported by Sasikala et al. (2013). Most of the participants were within the range of 15-20 years, and 33-38 years, which does not correlate with the previous reports reasons been that the current study geographical area have high young girls that are married and pushed to the family way, and majority of the participants had only basic education, poor educational background may correlate with poor personal hygiene and/or low economic status, which may in turn, make the pregnant women prone to Candida albicans infection. However this was not statistically significant (P=0.62; P>0.05). The highest frequency of Candida albicans among the pregnant women was under the age group of 15-20 yearswith prevalence of 20 (66.7), although the association was not statistically significant, the majority of Candida albicans infection confirmed participants were in their second and third trimester, which is similar to reports of study by Babic and Hukic, (2010). This is likely because in the third trimester of pregnancy, mostly the high level of estrogen results in higher glycogen deposits in the vagina, and this provides a good source of carbon, which supports the proliferation of Candida spp. Moreover, estrogen increases the affinity of Candida to the yeast cytosol receptor in vaginal epithelial cells; this indicates a very strong significant positive correlation in the relationship between the destruction of the epithelial cells of the vagina and the presence of the yeast. All participants infected with Candida

albicansin this study are married. This was in contradiction to what Bitew and Abebaw, (2018) reported; in which the majority of their study participants were divorced.

Clinical presentations reported by study included the hygiene level of the patients, the use of antibiotics, previous history of infection, frequency in the use of tight and type of pants used, knowledge of Candida albicans, itching and creamy/whitish/yellowish vaginal discharge, though the knowledge of Candida albicans, frequency in the use of tight, previous history of infection, types of pant used and hygiene cleanliness had no statistical association with Candida albicans. practitioners believe that nylon underwear and tight insulating clothing predispose to vaginal candidiasis by increasing the temperature and moisture of the perineum (Nwankwoet al., 2010). A study among African women wearing tight clothes reported a higher prevalence of *Candida* albicans Vulvovaginal candidiasis than those wearing loose clothing (Elegbe and Botu, 1982). Most participants had one or more of the three clinical presentations, but laboratory outcomes did not confirm them as having Candida albicans. This outcome simply means that clinical presentations of Candida albicans are not pathologic due to the facts that other vaginal conditions may establish such presentations. Clinical presentations reported by study subjects such as the use of antibiotics, had statistical association with Candida albicans (P < 0.05). This outcome simply means that excessive use of antibiotics tends to kill the good bacteria in the system giving chance for the proliferation of the yeast Candida albicans.

CONCLUSION

A relatively high prevalence of *Candida albicans* was observed in FMC Birnin-Kudu, Jigawa state. This study has shown that factors such as age of the women, use of antibiotics, stage of pregnancy and

dressing among women would be responsible for high prevalence of *Candida albicans*. This study shows that pregnant women who always frequently wear tight are 2.2 times more likely to be infected with *C. albicans* than those who do not frequently wear tight. However, the findings of this study indicated that the presence of *Candida albicans* and *Candida species* among these subjects was high and could be associated with an increased risk for sexually transmitted diseases.

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CONFLICT OF INTEREST

The authors of this manuscript declare that they have no conflict of interest whatsoever.

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