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The Distribution of *Candida Species* from Clinical Specimens in Different Areas of Ondo State

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Abstract: **A total of 400 various clinical samples which included HVS, urine, stool and oral swabs were collected from patients in the four geographical locations of Ondo State, Akure, Ondo, Ikare and Okitipupa. *Candida species* were isolate from 63 out of the 400 samples after culturing. Of these total numbers of positive isolates, *C. albicans, C. Krusei, C. glabrata, C. parapsilosis* accounted for 73.2%, 12.3%, and 4.6% respectively. The results revealed a statistically significant relationship (P<0.5) between the candida infection and the debilitated state of individuals. There was no significant relationship (P>0.5) between the gender and the age. The incidence of the *Candida albicans* are often present in the absence of vaginitis, therefore isolation in healthy individuals does not necessarily imply infection.**

***Keywords*: Clinical isolates, *Candida species*, Geographical location, Ondo State.**

I. INTRODUCTION

Candida is a yeast like-fungus. Fungi as a group includes moulds, mushrooms and yeasts, which form a major entity of eukaryotic cells called eumycota. They are classified among the higher protista group of living cells, along with the protozoa and algae but differ by being achlorophilic (lacking chlorophyll) or differentiation of root, stem and leaves. They are therefore, incapable of photosynthesizing their own carbohydrates from CO_2 and H_2O like the green plants. This inability of the fungi makes them essentially heterotrophic for carbon compounds, synthesized by autotrophic organisms. This is why they are commonly termed saprophytes, living on dead organic matters, often found in the soil, but some are wide-spread pathogens in living animals and plants-unlike most microorganisms, fungi have mainly being of service to man, as in the making of bread, fermented drinks, cheese and more recently, useful organic chemicals including antibiotics [1].

Fungi have caused only a small proportion of infectious disease in the past, but with the increasing control of diseases caused by bacteria and viruses through sanitation, education, immunization and chemotherapy, they are becoming more important. Nowadays there are as many deaths from mycoses (caused by fungal infections) in the U.S.A as there from bacteria and malaria put together. In Nigeria, the *Candida* species of the yeast-like fungi are more prevalent in diarrheic children [2, 3].

Candida are similar to yeast in growth by forming blastospores, they are non-capsulated and the blastospores can grow into group of filamentous cells joined end to end called pseudohyphae or pseudomycellium. Several species of these candida occur naturally in humans but about 90% of the candida infections are due to *Candida albicans*, other species are *C. tropicalis, C. pseudotropicalis, C. krusei, C. parasilosis, C. stellatoidea, C. glabrata, C. lipolytical, C. norvegensis, C. rugose*. Some of these candidas have been found in the United State of America as nosocomial bloodstream pathogens causing fungemia and arthritis.

Candida albicans, has been a part of the normal flora of the mouth, intestine, vagina and occasionally adjacent skin. It occurs in 80% of human population under normal circumstances and may not exhibit any harmful effects. However overgrowth results in candidiasis. Ninety percent (90%) of the candida infection are due to *Candida albicans*. It ranges between harmless commensal, and a pathogen. It is an opportunistic pathogen, which is becoming increasingly important as a nosocomial pathogen in immunocompromised, intensive-care unit, post operative patients, thrush infection, diabetes and diarrheic infections [4, 5, 6].

The aim of this study therefore is to identify C*andida species* form clinical samples and their distribution in different parts of Ondo State.

II. MATERIALS AND METHODS

The clinical samples collected include urine, vaginal swabs, stool, and mouth swabs. The study population consisted of males and females, aged between 0-5years for infants and 20-50years for adult (mean 35years). The number of population sampled was 400 people. 40 infants aged 0-5years, (20 females and 20 males) with and without thrush. 140 women aged 20-40 years with (70 pregnant and 70 without pregnancy). 120 people aged 20-50 years with 60 (3o males and 30 females) having diabetes and 60 (30 males, 30 females) without diabetes. 40 people (20 males, 20 females) of the same age group without HIV/AIDS infection Ten individuals (5 males, 5 females) that were diarrhoiec and 10 people (5 males, 5 females), aged 20-50 years without diarrhea.

Samples Collection:

About 5mls of urine samples were collected from each patient with the aid of a sterile universal bottle, and centrifuged at 4000rpm for 5minutes using sterile test tube. The deposits were examined for yeast cells under the x40 objective of the binocular microscope. A loopful of the deposit was seeded on a sabouraud's dextrose agar culture plate for an overnight incubation at 37^{o}C. The plate was examined after overnight incubation for yeast growth and identified wit the conventional methods. Mouth swabs were collected with the aid of sterile swab sticks and sealed onto the culture plates for overnight incubation at 37^{o}C. The stool samples were also cultured and incubated at 37^{o}C. The growth on the culture plates were examined and identified as described below.

Analytical Methods:

Conventional methods for the identification of candida species were based on morphological characteristics, assimilation and fermentation tests [7, 8].

Gram Stains test:

A gram stain reaction was carried out on all the clinical isolates [9].

Germ tube test:

Candida albicans was identified by germ tube formation in serum owing to its ability to form chlamyclospores [10] .

Fermentation test:

Fermentation test were employed using sugars such as glucose, maltose, sucrose and lactose at 2-3% [11].

Data Analysis:

The data generated were analyzed using chi-square method. P value <0.05 was considered statistically significant while P value >0.05 was considered insignificant.

III. RESULTS

Out of the total 400 clinical samples collected from HVS, Urine, Stool and Oral swabs, 65 were found positive for candida (Table 1). Of these total number of positive cultures, *Candida albicans* accounted for 73.2%, C*andida krusei* 12.3% *Candida glabrata* 4.6% while *Candida parapsilosis* 4.6% (Table 2)

The percentage of positive cultures in relation to different clinical specimens is given in Table 3. Table 4 shows the prevalence of Candida species in relation to sex distribution while Table 5 shows the prevalence of Candida species in relation to age distribution. Table 6 shows the prevalence of Candida species in relation to morbidity

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TABLE 1: DISTRIBUTION OF *CANDIDA* SPECIES FROM 400 CLINICAL SPECIMENS COLLECTED IN DIFFERENT PART OF ONDO STATE

S/N	Samples Collected	No. Collected	No. of Isolate	Percentage (%)
1	Urine (pregnant women)	50	10	20
2	Urine (non-pregnant women)	50	2	4
3	HVS (pregnant women)	20	6	30
3b	Total (for pregnant women)	**70**	**16**	**22.8**
4	HVS (non-pregnant women)	20	2	10
5	Urine (diabetic)	60	11	16.9
6	Urine (non-diabetic)	60	1	1.66
7	Urine (HIV/AIDS+ve)	40	10	25
8	Urine (HIV/AIDS-ve)	40	0	0
9	Oral swabs (thrush)	20	3	15
10	Oral swabs (non-thrush)	20	0	0
11	Stool (diarrhoeic)	10	10	100
12	Stool (non-diarrhoeic)	10	10	100
		400	**65**	**16.25**

TABLE 2: *CANDIDA* SPECIES IDENTIFIED FROM POSITIVE CULTURES

S/N	Samples	Total Positive Cultures	*Candida Albicans*	*Candida Krusei*	*Candida Glabrata*	*Candida Parapsilosis*
1	Urine (pregnant women)	10	10	-	-	-
2	Urine (non pregnant women)	2	2	-	-	-
3	HVS (pregnant women)	6	6	-	-	-
4	HVS (non pregnant women)	2	2	-	-	-
5	Urine (diabetic)	11	8	3	-	-
6	Urine (non diabetic)	1	1	-	-	-
7	Urine (HIV+ve)	10	7	2	-	1
8	Oral swabs (thrush)	3	3	-	-	-
9	Stool (diarrhoeic)	10	6	2	1	1
10	Stool (non-diarrhoiec)	10	6	1	2	1
	Total	**65**	**51**	**8**	**3**	**3**
	Percentage	**16.25**	**78.5**	**12.3**	**4.6**	**4.6**

TABLE 3: 400 CLINICAL SPECIMENS SCREENED FOR *CANDIDA* SPECIES

S/N	Samples	No. Sample	Positive Isolate	(%) Positive
1	Urine (pregnant women)	50	10	20
2	Urine (non-pregnant women)	50	2	4
3	HVS (pregnant women)	20	6	30
4	HVS (non pregnant women)	20	2	10
5	Urine (male diabetic)	30	3	10
6	Urine (female diabetic)	30	8	26.7
7	Urine (male non-diabetic)	30	-	0
8	Urine (female non-diabetic)	30	1	33.3
9	Urine (male HIV/AIDS+ve)	20	8	40
10	Urine (female HIV/AIDS+ve)	20	2	10
11	Urine (male HIV/AIDS-ve)	20	-	0
12	Urine (female HIV/AIDS-ve)	20	-	0
13	Oral swabs (0-5yrs thrush)	10	2	20
14	Oral swabs (Adult thrush)	10	1	10
15	Oral swabs (0-5yrs non-thrush)	10	-	0
16	Oral swabs (Adult non-thrush)	10	-	0
17	Stool (male diarrhoeic)	5	5	100
18	Stool (female diarrhoeic)	5	5	100
19	Stool (male non-diarrhoeic)	5	5	100
20	Stool (female non-diarrhoeic)	5	5	100
	Total	**400**	**65**	**16.25**

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TABLE 4: PREVALENCE OF *CANDIDA* SPECIES IN RELATION TO GENDER DISTRIBUTION

Gender	No. Sample	No. Positive (%)	No. Negative (%)
Female	270	42 (15.6)	228 (84.4)
Male	130	23 (17.7)	107 (82.3)
Total	**400**	**65 (16.25)**	**335 (83.75)**

Calculated = 0.30

Tabulated = 2.71

P>0.05=not significant

TABLE 5: PREVALENCE OF *CANDIDA* SPECIES IN RELATION TO AGE DISTRIBUTION

Age (yrs)	No. Sample	No Positive (%)	No Negative (%)
0-5	20	2 (10)	18 (90)
20-50	380	40 (10.5)	340 (89.5)
Total	**400**	**42 (10.5)**	**358 (89.5)**

Calculated = 0.056

Tabulated = 2.71

P>0.05=not significant

Yrs=years

TABLE 6: PREVALENCE OF CANDIDA SPECIES IN RELATION TO MORBIDITY

Condition	No. Sample	No Positive (%)	No Negative (%)
Debilitated	200	50 (25)	150 (75)
Non-debilitated	200	15 (7.5)	185 (92.5)
Total	**400**	**65 (16.25)**	**335 (83.75)**

Calculated = 225.02

Tabulated = 2.71

P>0.05=not significant

IV. DISCUSSION

In this study, 16.25% of the total population studied was positive for *Candida* species. Out of the 200 debilitated subjects with HIV/AIDS, diabetics, pregnancy and diarrhoeic, 25% were positive while 7.5% were positive for healthy individuals. People living with HIV/AIDS infection were found to have species other than the *C. albicans. Candida* is capable of fermenting glucose. The presence of excess glucose causes an influx of the candida species from the primary locations and lead to an anatomical shift of the yeast/bacteria ratio. The numerous yeasts become invasive and turn opportunistic pathogens.

Diabetes is caused by deficiency of insulin which controls the conversion of glucose for the normal body use. The excess glucose in the body is filtered out along the urine and this accounted for the high prevalence of the *candida* species in diabetic patient. The presence of pregnancy alters the estrogen production and the helpful lactobacilli which lead to thrush and other infections, this account for the high prevalence of candida species in pregnant women. Patient with HIV/AIDS diseases has general disorders of their immune system. The change in the anatomical immune barriers, the use of cytotoxic drugs by these patients reduces the normal flora bacteria in the body and allows the normal flora yeast to thrive and become opportunistic pathogens. This accounted for the high prevalence of candida species in HIV/AIDS patient.

Other researchers have reported the prevalence of these *candida* species in immunodefiant individuals and 26.5%positively in organ recipient individuals. Some reported the prevalence of this *candida* species in generalized diseases such as leukaemia especially when treated with cytotoxic drugs. This was confirmed by Bon-Delabesse and Nho who reported the emergence of *C. Krusei* due to drug resistance in immunodefient subjects [12, 13, 14].

V. CONCLUSION AND RECOMMENDATION

From table I of this study, 25% of HIV/AIDS subjects were found to have candida infection. Pregnant women accounted for 22.8% of the infection, the diabetic patient 16.9% of the infection. Diarrhoeic individuals were found to have 100% of the *candida* species. This agrees with the findings of other researchers who reported that there is high prevalence rate of candidiasis among immune deficient or compromised individuals. It equally agrees that diarrhoeic patients that there is statistically no significant difference between the candida infections, in relation to sex and age (table 4 and 5). With the 16.25% positive of the population, 25% positive of the immuno-compromised individuals and 7.5% positive of healthy individuals, it will be advisable before treating a candida infection to first identify the underlying circumstances that brought about the infection. However this may be difficult in cases of HIV/AIDS patients.

This study also showed that the presence of candida species in healthy individuals suggests that commensal strains could assume pathogenic role in compromised host. I therefore recommend that more research work should be carried out to type out the strains that resist treatment. More precautions should be taken during laboratory operations in order to avoid laboratory acquired infections. Cytotoxic drugs should be used in order to minimize systemic infections.

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