



PREVALENCE OF FUNGI ASSOCIATED WITH URINARY TRACT INFECTION: A CASE STUDY OF STUDENTS OF THE JOSEPH SARWUAN TARKA UNIVERSITY, MAKURDI, BENUE STATE



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Received: August 25, 2021 Accepted: November 11, 2021

Abstract: The rising trend of fungal urinary tract infection (FUTIs) is of great concern. Five hundred students were recruited into this study to a certain the prevalence of fungi associated with urinary tract infections among students. Each student recruited into the research completed a questionnaire that explores information on their age, hostel block, risk and predisposing factors. Urine samples collected from the students were analyzed in the laboratory according to the clinical laboratory standards. RapID yeast system (identification kits) was used for confirmation of isolates, which inferred the presence of *Candida albicans* 310 (68.9%) and *Candida glabrata* 140 (31.1%) in the urine samples of the students. From the overall prevalence result 45 (90%) positive. Persons aged 22-26 had the highest prevalence (55.6%) while the age range of 27-31 (13.3%) and above had the least prevalence. The use of antibiotics, sexual activities and wearing tight fitted nylon and cotton under wears were significant at 99% $P < 0.001$ and identified as risk factors for fungi associated with urinary tract infections. This study reveals high prevalence of fungi associated with urinary tract infection among students in Joseph Sarwuan Tarka University, Makurdi.

Keywords: Candidiasis, *Candida albicans*, HVS, CLSI, RapID yeast plus system

Introduction

Urinary tract infections (UTIs) are the inflammatory disorders of the urinary tract caused by the abnormal growth of pathogens (Martin *et al.*, 2019). Urinary tract infections are caused predominantly by bacteria and fungi species that live in the digestive tract, vagina, or around the urethra. In Nigeria, fungi species such as; *Candida albicans*, *Candida tropicalis* and *Candida glabrata* are usually implicated in UTI patients as observed in studies carried out in Enugu, Yola, Zaria and Ife, shows that these fungi are the same etiological agents (Iregbu and Nwajobi, 2013). It is estimated that 150 million urinary tract infections occur yearly on a global basis resulting in more than 6 billion dollars in direct health care expenditure (Foxman, 2012).

The urinary tract consists of the urethra, prostrate glands (in males), kidney, ureter and bladder. The foci of the infection may be kidney, bladder, urethra or ureter (Vasudevan, 2014). Anatomically, urinary tract infection whether caused by fungi or bacteria are categorized into two (lower and upper tract infection) and this may occur in asymptomatic or symptomatic forms. The infection is named based on the site of infection, the infection of the urethra and ureter is referred to as urethritis and ureteritis respectively whereas cystitis and pyelonephritis correspond to infection of the bladder and kidney (Vasudevan, 2014). Cystitis is a common type of infection whereas the infection associated with renal damage (kidney) is an issue of serious concern (Vasudevan, 2014).

The urethra is shorter and closer to the anus in females than in males and is more readily transverse by microorganism; this is why urinary tract infection is common in females than in males (Coyle *et al.*, 2017). The urinary tract system is structured in such a way that it helps ward off infection and the bladder and urethra normally prevent backward flow of urine from the bladder to ureter which causes vesicoretral reflux (Nagler *et al.*, 2011). The flow of urine from the bladder helps wash out pathogens from the body. Urinary tract infections may occur as a result of lapses in hygiene standard both individual and communal level (Sobel *et al.*, 2014). The treatment of an infected individual is based on the antimicrobial sensitivity pattern of the uropathogen isolated (Agada *et al.*, 2017).

The rising trend of urinary tract infections (UTIs) among students of the Joseph Sarwuan Tarka University, Makurdi is gradually becoming a cause of concern. The predisposition of

women, especially in the hostel to the infection is because of their short urethra, and certain behavioral factors which include delay in micturition, sexual activities, use of oral contraceptives, prolong use of antibiotic, use of diaphragms and spermicides which promote colonization of the periurethral area with pathogens. The prevalence of UTIs in men is significantly lower than in women, occurring due to sexual intercourse, in men with urological structural abnormalities and in older adult men. The infection is mostly proliferated in women and in most cases it often results from perineal or periurethral pathogens that enter the urethral and ascend into the bladder often in association with sexual activities, wiping from back to front after using the toilet or due to mechanical instrumentation such as catheterization (Agada *et al.*, 2021)

Up to 60% of young women have at least one symptomatic UTI during their life time. Around 10% of young women in Nigeria have one or more episodes of symptomatic UTIs each year. Hence there is a need to effectively study the prevalence of the fungi associated with UTIs in the student Hostels and it is hoped that data obtained in this research will be useful in the control and prevention of UTIs in the University student Hostels.

Materials and Methods

Study area

This study was carried out at Joseph Sarwuan Tarka University, Makurdi the Capital of Benue state in Nigeria. The City is located in North Central Nigeria along the Benue river it is home of the Joseph Sarwuan Tarka University and the Benue state university. The major ethnic groups are the Tivs, Idomas and Igede.

Makurdi town lies between latitude 7°44'N and longitude 8°32'N covering an area of 820 km square with an estimated population of 348,990 people (National Population Commission of Nigeria, 2011). The vegetation type in Makurdi is guinea savannah with annual rainfall between 150 – 180 mm and temperature of 26-29°C (NPCN, 2011). The school is divided into three major cores: south core, north core and middle core. There are four major hostels for male and female students: block A and B for females and block C and D for male student, respectively.

Ethical approval

The Ethical approval for this study was obtained from the Ethical Committee on Research of Infectious Diseases of the Federal Medical Center, Makurdi, Benue State, Nigeria. Consent was also obtained from the female patients that presented themselves for medical treatment in the University Clinic before sample collection. The approval was on the agreement that participants' anonymity will be maintained, good laboratory practice/quality/control ensured, and that every finding would be treated with utmost confidentiality and for the purpose of this research only. However, patients who desire information on our findings would be given (verbally) free of charge.

Demographic data collection

A well-structured questionnaire were used to collect relevant demographic, clinical and laboratory information of patients.

Sample size determination and sample collection

The sample size was determined based on the prevalence rate of a study carried out by Sapkota *et al.* (2010) as follows:

$$N = \frac{Z^2 P(1-P)}{d^2}$$

Where: N= patients to be sampled; Z= the standard normal deviation corresponding to a chosen level of confidence = 1.96; P= expected prevalence v (0.2); d= the degree of accuracy desired (2.5%) = 0.025

In our calculation, we used $Z = 1.96$, $P = 0.2$ and $d = 0.025$. This calculation resulted in a sample size of 504. This sample size was reduced to 500 samples to account for the clustered nature of the study design. This total sample size was divided by the number of clusters. This method of dividing the sample equally among clusters was in accordance with "generic cluster sample" design methods previously described by the WHO Department of Vaccines and Biologicals (Sapkota *et al.*, 2010).

Samples collection

A total of 500 urine samples were collected from student of the Joseph Sarwan Tarka University, Makurdi living on Campus (Hostel). 25 samples from female students and another 25 samples from male students, students who indicated their interested were recruited for the study with no inclusion or exclusion criteria. Participants were issued questionnaire which contains questions such as age, personal hygiene, sexual activities, number of those taking broad spectrum Antibiotics and those taking oral contraceptives, type of under dress (pants), those using soap to wash the vulva.

Macroscopy

The urine samples collected were physically examined /viewed with the naked eye for colour, turbidity and to detect if some samples had blood in them (Agada *et al.*, 2021).

Culture media

The culture media used is Sabouraud Dextrose Agar (SDA). It is a classic medium recommended for most studies (Cheesbrough, 2006). All the media used were prepared according to manufacturer's instructions.

Media preparation (SDA)

Approximately 31.0 grams of dehydrated Sabouraud Dextrose Agar powder was added into 250 mls of distilled water and an antibiotic (tetracycline) was added to inhibit the growth bacteria contaminants. The suspension was homogenized,

sterilized at 121°C for 15 min, cooled to about 45 – 50°C, 20 mls of the solution was poured into a Petri dish and allowed to cool and set as described by Agada *et al.* (2017).

Samples inoculation

Pour plate method was adopted, 1 ml each of the urine sample was poured into different Petri dishes and the prepared media after allowing to cool to 45 – 50°C 20 mls each was poured into the Petri dishes and swirled clock wise and anti- clock wise, up and down, left and right for proper mixing of the sample in the medium and allow to set then incubated for 24– 72 h at 37°C (Agada *et al.*, 2021).

Staining

After about 24 – 72 h of incubation at 37°C to observe for fungal growth on the agar plate; a smear of small amount of the colonies was made on a clean glass slide using a sterilized wire loop. It was then slightly heat fixed and stained using lacto phenol cotton blue.

Microscopy

After staining using lacto phenol cotton blue, the slide is covered with a cover slip and viewed using the microscope at x10 and x40 objective lens to view fungi cells and hyphae (Cheesbrough, 2006).

Germ tube test

Germ tube test as a method of identification of *C. albicans* was also carried out on suspected yeast colonies. Using a sterile wire loop small portion of the colonies were inoculated into peptone water for 24 h and incubated at 37°C to obtain a pure culture. The pure culture was inoculated in sterile test tubes containing 0.5ml of human serum which was incubated for 3 – 4 h. A drop of the mixture was placed on a grease free slide with a drop of lacto phenol blue stain and covered with a cover slip. It was then examined microscopically at X10 and X40 objective lenses (Cheesbrough, 2006). *C. albicans* formed germ tubes with short lateral hyphae filaments without any constrictions while non- *C. albicans* species did not

Biochemical test

Rapid yeast plus system was used to identify *Candida* species. Pure culture of the yeast was inoculated into the rapid yeast plus system panel containing fungi food substrates and enzymes for 4 h. Following the manufacturer's instructions, Antigen A and Antigen B were added to their corresponding wells on the panel and further reactions were observed and matched with the manufacturers color chart. The result shows the presence of *Candida albicans* and *Candida glabrata* in urine samples of the students (Agada *et al.*, 2021).

Results and Discussion

This section presents the findings of this study which was carried out based on the objective of the study. Table 1 shows the cultural, morphological, biochemical characteristics and percentage prevalence of the fungi species. Out of the 50 samples collected some were colourless, others were pale yellow and amber colors some of the samples were turbid. After culturing and incubation for 24 – 72 h, the colonies appear smooth pasty and creamy with oval shaped single budded cells with a short hyphae when viewed microscopically. Biochemical test using the rapid yeast plus system indicated the presence of *Candida albicans* and *Candida glabrata* with 35 (70%) of the total sample to be *C. albicans* while 15(30%) were *C. glabrata*.

Table 1: Microbiological analysis

Sample macroscopy	Culture macroscopy	Microscopy/ morphology	Biochemical test	Inference	% prevalence
Colorless, amber, pale yellow and turbid samples	Smooth, pasty creamy colonies	Oval shaped single budded cells with a short hyphae	Rapid Yeast Plus System	<i>Candida albican</i>	350 (70%)
Colorless, amber, pale yellow and turbid samples	Smooth, pasty, creamy colonies	Oval shaped single budded cells with a short hyphae	Rapid Yeast Plus System	<i>Candida glabrata</i>	150 (30%)

Table 2 shows the prevalence of fungi urinary tract infections (FUTIs) among the student of Joseph Sarwuan Tarka University, Makurdi, with respect to their age. Age group 17-21 with a percentage prevalence of 34% are statistically significant at 99% $p < 0.01$, while the age group 22-26 with percentage prevalence of 54% are statistically significant at 99% $p < 0.01$, the age group 27-31 with percentage prevalence of 12% are statistically significant not significant at 99% $p > 0.05$. Table 3 it shows the prevalence of FUTIs among studentsof Joseph Sarwuan Tarka University, Makurdi,with respect to hostel block A and D with percentage prevalence of 26% and 24% are statistically significant at 99% $p < 0.01$ while block B and C are not statistically significant at 99% $p > 0.05$.

Table 2: Prevalence of fungi associated with urinary tract infection among students of Joseph Sarwuan Tarka University, Makurdi, with respect to age

Age Group	Observed N	Expected N	Chi-square	Sig
17-21	170	85	7.118	0.008*
22-26	270	135	19.593	0.000*
27-31	60	60		
Total	500			

* = $P < 0.01$, statistically significant at 99%

Table 3: Prevalence of fungi associated with urinary tract infection among students of Joseph Sarwuan Tarka University, Makurdi, with respect to Hostel

Hostel Blocks	Observation N	Expected N	Chi-square	Sig
Block A	130	65	9.308	0.002*
Block B	120	120		
Block C	130	65	3.769	0.052
Block D	120	60	8.333	0.004*
Total	500			

** = $P < 0.001$; statistically significant at 99%

Table 4 shows the prevalence of FUTIs among studentsof Joseph Sarwuan Tarka University, Makurdi, with respect to their gender. Both male and female with percentage prevalence of 50% are statistically prevalent at 99% $p < 0.01$. Table 5 shows the prevalence of FUTIs among studentsof Joseph Sarwuan Tarka University, Makurdi, with respect to poor personal hygiene. It had a percentage prevalence of 42% which is not statistically significant at 95% $p > 0.05$. Table 6 it shows the prevalence of FUTIs among studentsof Joseph Sarwuan Tarka University, Makurdi, with respect to sexual activities. It had a percentage prevalence of 66% which is statistically significant at 95% $P < 0.05$.

Table 4: Prevalence of fungi associated with urinary tract infection among students of Joseph Sarwuan Tarka University, Makurdi, with respect to gender

Gender	Observation N	Expected N	Chi-square	Sig
Male	250	125	11.560	0.001*
Female	250	125	21.160	0.000*
Total	500			

** = $P < 0.001$; statistically significant at 99%

Table 5: Prevalence of fungi associated with urinary tract infection among students of Joseph Sarwuan Tarka University, Makurdi, with respect to poor hygiene

Poor Hygiene	Observation N	Expected N	Chi-square	Sig
Yes	210	250	1.280	0.258
No	290	250		
Total	500			

$P < 0.05$; not statistically significant at 95%

Table 6: Prevalence of fungi associated with urinary tract infection among students of Joseph Sarwuan Tarka University, Makurdi, with respect to sexually active

Sexually Active	Observed N	Expected N	Chi-square	Sig
Yes	330	250	5.120	0.024
No	170	250		
Total	500			

$P < 0.05$; statistically significant at 95%

Table 7: Prevalence of fungi associated with urinary tract infection among students of Joseph Sarwuan Tarka University, Makurdi, with respect to tight fitted under wears

Tight fitted under wares	Observed N	Expected N	Chi-square	Sig
Yes	150	250	2.880	0.005*
No	350	250		
Total	500			

* = $P < 0.01$, statistically significant at 95%

Table 7 shows the prevalence of FUTIs among studentsof Joseph Sarwuan Tarka University, Makurdi, with respect to wearing tight fitted nylon and cotton under wear which had a percentage prevalence of 30% is statistically significant at 95% $P < 0.01$.

Table 8 shows the prevalence of FUTIs among studentsof Joseph Sarwuan Tarka University, Makurdi, with respect to the use of contraceptives which had a percentage prevalence of 24% is not statistically significant at 95% $P > 0.05$. Table 9 shows the prevalence of FUTIs among students of Joseph Sarwuan Tarka University, Makurdi, with respect to douching which had a percentage prevalence of 38% is statistically not significant at 95% $P > 0.05$. Table 10 shows the prevalence of FUTIs among studentsof Joseph Sarwuan Tarka University, Makurdi,with respect to prolong use of antibiotics which had a prevalence of 22% is statistically significant at 95% $P < 0.05$.

Table 8: Prevalence of fungi associated with urinary tract infection among students of Joseph Sarwuan Tarka University, Makurdi, Benue State with respect to use of contraceptives

Use of Contraceptives	Observation N	Expected N	Chi-square	Sig
Yes	230	250	.320	0.572
No	270	250		
Total	500			

$P < 0.05$, not statistically significant at 95%

Table 9: Prevalence of fungi associated with urinary tract infection among students of Joseph Sarwuan Tarka University, Makurdi, Benue State with respect to douching

Douching	Observation N	Expected N	Chi-square	Sig
Yes	190	250	2.880	0.090
No	310	250		
Total	500			

Table 10: Prevalence of fungi associated with urinary tract infection with respect to antibiotic consumption among students of Joseph Sarwuan Tarka University, Makurdi, Benue State

Antibiotics	Observation N	Expected N	Chi-square	Sig
Yes	110	250	15.680	0.000*
No	310	250		
Total	500			

* = P<0.05, statistically significant at 95%

Table 11 shows the overall percentage prevalence of FUTIs (*Candidiasis*) among students of Joseph Sarwuan Tarka University, Makurdi. Those within the age group 17-21, 22-26, those students living in block A and D hostels, male and

Table 11: Prevalence of fungi associated with urinary tract infection among students of Joseph Sarwuan Tarka University, Makurdi, Benue State

<i>Candidiasis</i> infection	N	Mean	Std. Deviation	Minimum	Maximum	Chi-square	Sig
17-21	170	1.18	0.393	1	2	7.118	0.008*
22-26	270	1.07	0.267	1	2	19.593	0.000*
27-31	60	1.00	0.000	1	1		
Block A	130	1.08	0.277	1	2	9.308	0.002*
Block B	120	1.00	0.000	1	1		
Block C	130	1.23	0.439	1	2	3.769	0.052
Block D	120	1.08	0.289	1	2	8.333	0.004*
Male	250	1.16	0.374	1	2	11.560	0.001*
Female	250	1.04	0.200	1	2	21.160	0.000*
Poor Hygiene	500	1.58	0.499	1	2	1.280	0.258
Sexually Active	500	1.34	0.479	1	2	5.120	0.024*
Tight Fitted Underwear	500	1.70	0.463	1	2	8.000	0.005*
Use of Contraceptives	500	1.54	0.503	1	2	.320	0.572
Use of Soap to Wash Vulva	500	1.62	0.490	1	2	2.880	0.090
Antibiotics	500	1.78	0.418	1	2	15.680	0.000*

* = P<0.001, statistically significant at 99%

The result of this study has established the existence of fungi urinary tract infection (FUTIs) among students of Joseph Sarwuan Tarka University, Makurdi with a high prevalence rate of 90% (45/50) at p<0.001. In this study *Candida albicans* was identified as the predominant fungi specie causing FUTIs with a high prevalence of 310 (68.9%) followed by *Candida glabrata* which had a prevalence of 140 (31.1%) these were also the same fungi specie identified by Schulz *et al.* (2016) and Voltan *et al.* (2014).

A high prevalence rate was recorded in this study among students who are between the age group of 22-26 years (55.6%), followed by 17-21 years (31.1%) and 27-31 years 6(13.3%) had the least prevalence. This revealed that student especially women in their reproductive years are more prone to FUTIs this is consistent with the findings of Emerebe *et al.* (2015) and Mbim *et al.* (2017) according to them this is because estrogen which induces the lining of the vagina to mature contain glycogen a substrate which *Candida* specie thrives in and in men could be due to sexual intercourse with an infected person or due to poor personal and environmental hygiene (Behzadi *et al.*, 2015). A high prevalence rate was recorded in both gender, female had a high prevalence rate of 24(53.3%) P<0.001 this is in consistent with the findings of Emerebe *et al.* (2015) according to them 60-75% of female are infected with a FUTIs, male student had a prevalence of 21(46.7%) P<0.001 this is also in consistent with Behzadi *et al.* (2017) and Emerebe *et al.* (2015) according to them 12-15% of men have been reported to develop a FUTIs following sexual intercourse with an infected person or due to poor personal and associated risk factors.

female students, those who are sexually active, those who wear tight fitted nylon and cotton under wears and those who are on prolong use of antibiotics are statistically significant at 95 and 99% P<0.001 while those living in block B and C are, those within the age group of 27-31, those practicing poor personal hygiene, those who use contraceptives and those who use soap to wash the vagina are all not statistically significant at both 95 and 99%, respectively P>0.05.

The relationship between the prevalence of FUTIs and some associated risk and predisposing factors were evaluated in this study. The results revealed that poor personal hygiene is statistically not significant at 99% P>0.05 this agrees with the finding of Sobel *et al* (2014) that revealed that urinary tract infections may occur as a result of lapses in hygiene standard both at individual and communal level, it disagrees with the findings of Behzadi *et al.* (2008); according to them this infection correlates with individual hygiene.

Sexual activities in this study is statistically significant at 95% P<0.05 this agrees with the findings of Behzadi *et al.*, 2011 and Ackhar *et al* 2013 who reported that sexual activities increase the risk of FUTIs, similarly Behzadi *et al.* (2017), Emerebe *et al.* (2015) and Mbim *et al.* (2017) correlates this factors as a cause of FUTIs. Wearing tight fitted nylon and cotton underwear in this study was statistically significant at 99% P<0.001 this agrees with the findings of Behzadi *et al.* (2008) who reported that his tight fitted underwears foster fungal growth this is because this tight fitted under wears discourage aeration of the urinary tract increasing the moisture and making the region dark and warmth.

Douching, use of contraceptives and soap to wash the vulva as risk factors identified in this study are statistically not significant not significant at 95% P>0.05 this findings agrees with that of Agada *et al.* (2017) and Mbim *et al.* (2017) who reported that the use of contraceptives, douching and soap to wash the urinary tract causes FUTIs. Prolong use of antibiotics in this study was statistically significant at 99% P<0.01 this agrees with the findings of Behzadi *et al.* (2015) and Sobel *et al.* (2014). Prolong use of antibiotics can destroy

the bacterial flora of the intestine giving room for fungi species, similarly broad spectrum antibiotics can destroy the normal flora of the vagina which suppresses the growth of fungi specie particularly *Candida*. This normal flora have been known to secrete acidic materials which keep the pH of the vagina under check, alteration in the normal pH influence the overgrowth of this fungus leading to FUTIs.

Conclusion

This study reveals that the student of Joseph Sarwuan Tarka University, Makurdi had a high prevalence of FUTIs 90%. *Candida albicans* and *Candida galabrata* are the fungi specie implicated, with *C.albicans* as the major causative agent accounting for about 68.9% of the isolates. Both genders have high prevalence rate and the age group of 22-26 had the highest prevalence rate 55.6% followed by 17-21 31.1% the age group 27-31 had the least prevalence of 13.3%. Risk and predisposing factors such as sexual activities, wearing of tight fitted underwear and prolong use of antibiotics are statistically significant and identified as causes of FUTIs.

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