Prevalence and Antimicrobial Susceptibility Profile of Fungi Isolated From the Environment of Two Major Hospitals in Calabar Metropolis, Nigeria

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Abstract

The hospital environments of two hospitals were investigated for fungi prevalence and their susceptibility profile to commonly used antifungal drugs. A total of 240 swab samples were collected from the laboratory coats, sinks, door handles and table tops of the hospital wards, pharmacies, blood banks theatres and intensive care units. The isolates were aspectically manipulated, mean load determined and their susceptibility determined using disk diffusion assay. This study revealed that General Hospital (GH) and Infectious Disease Hospital (IDH) recorded total mean loads of 386cfu and 416cfu, respectively. Air had the highest count range of 160 – 201cfu with GH having the highest while the pharmacy unit of the GH had no fungi. Fungi prevalence rates of 17(58.6%) and 12(41.4%) were observed in General hospital and IDH, respectively. The highest prevalence of 5(29.4%) and 6(50%) was seen in the male wards of both locations. Most frequent organisms in the pharmacy, intensive care, theatre, and ward units of General Hospital were *Candida species* and *Aspergillus species* from the laboratory unit, and *Penicillum species* in the blood bank. On the other hand, *Rhizopus species* dominated the pharmacy unit and *Candida species* in ICU. In the ward unit of IDH, *Penicillum species*, *Candida species* and *Rhizopus species* were most frequent. *Penicillum species* exhibited varying degrees of resistance to all the antifungal agents employed in this study. This study revealed gross contamination of the environment of both hospitals environments and further confirms that hospital environment plays a significant role in hospital associated infections.

KEYWORDS: Nosocomial, fungal, hospital, Calabar, susceptibility

INTRODUCTION

Fungal infections have been reported to be on the increase in hospital settings due to rapid influx and efflux of people and contribute significantly to the 5,000 deaths recorded yearly from hospital related infections (Stichler, 2007 and Perlroth *et al.,* 2007). Once regarded as laboratory contaminants, they are now implicated in the pathogenesis of most health care associated infections (Shrishari *et al*., 2012). These infections range from urinary infections, respiratory infections, surgical site infections to systemic infections. The commonly implicated fungi in nosocomial infections include *Candida species*, *Aspergillus species*, *Penicillum species*, *Fusarium species*, *Mucorales* and other molds such as *Scedosporium species* (Bereket *et al*., 2012 and Yang *et al*., 2013). These infections have been shown by studies to be aggravated by the rapid development of resistance by fungi to commonly used antifungal agents including the azoles and polyenes (Perlroth *et al.,* 2007).

Their incidence as evaluated by researches stem from the fact that risk factors associated with these infections are multifaceted. Commonly reported reservoirs of

fungi in hospital settings include moist surfaces, construction and renovation activities, inflow of unfiltered outside air, backflow of contaminated air, air filters, fireproofing materials, air conditioners and duct systems, as well as dust above false ceiling (Haidexen, 2009). Construction and renovation activities are ever constant activities in the health care settings that have been identified as a major reservoir capable of dispersing fungal spores (Kanamori *et al*., 2015). Once dispersed, they can gain entry through contact (either direct or indirect) or become airborne or vectored by common vehicles such as food or water, medications, devices/equipment and also living vectors (Bereket *et al*., 2012). As far as fungal infections with primary site being the lower respiratory tract are concerned, air is an important route (Kanamori *et al*., 2015). More worrisome is the fact that a study has shown that the emergence of Acquired Immunodeficiency Syndrome (AIDs) has altered the incidence and prevalence of fungal infections in hospital settings and most fungal infections found in hospitalized patients are often severe and progress to chronic stages and become difficult to treat (Shrishari *et al*., 2012).

Despite progress in public health and hospital care, reports from Saka *et al* (2011) and Naidu *et al* (2014) has it that nosocomial infections have continued to be a menace in hospitalized patients globally, with the impact more severe in resource-poor settings where the rate of infection is estimated to range from 25% to 40% (Uneke and Ijeoma, 2010). This challenge is further compounded by the lack of data in sub Saharan Africa including Nigeria on prevalence of these fungi in the hospital environment and makes it almost impossible to monitor the resistance pattern and the effectiveness of disinfection protocols in these hospitals. Therefore, this research was aimed at evaluating the prevalence and antimicrobial susceptibility profile of fungi isolated from the environment of two major hospitals in Calabar Metropolis, Nigeria.

MATERIALS AND METHODS

Study area/site

This study was undertaken in two major hospitals including General Hospital and Infectious Disease Hospital all in Calabar Metropolis. Calabar metropolis is commonly referred to as "Canaan City", and is the capital of Cross River State in south-eastern Nigeria (Antigha *et al*., 2015). See map of the study area below.

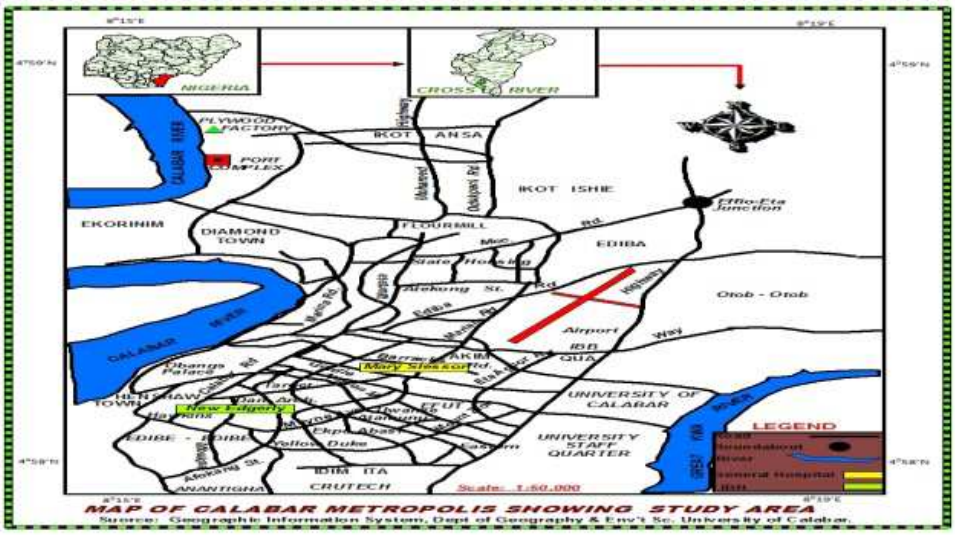

Figure 1: Map of Calabar Metropolis showing Study Areas

Source and size of sample

A total of 240 swab samples were collected from two hospitals with 20 from each of the following units; wards, pharmacies, blood banks, theatres, laboratories and intensive care units. The swab samples from these units were collected from table tops (10), laboratory coats (5), sinks (3) and door handles (2). In addition, samples for air qualities of the units were collected using settle plate technique.

1.3 Media/Reagents and antifungal agents

The media and reagents used for the study included sabouraud dextrose agar (SDA), crystal violet, 70% alcohol, safranin, Gram's iodine, cotton blue in lactophenol, human serum, distilled water, methylated spirit and hydrogen peroxide solution were purchased from a reputable dealer (TM media, Nigeria). The antifungal agents used in this study were ketoconazole (K) (10µg), fluconazole (F) (25µg), griseofulvin (GF) (25µg) (Rosco Diagnostica, Denmark), amphotericin B (AP) (20µg) and nystatin (NS) (100units) (Hi Media Laboratories, India).

Collection and inoculation of swap samples

This was done following procedures described by Murray *et al* (2003). Briefly, sterile swab sticks soaked in physiological saline were used to swab all the aforementioned sample sites, labelled, packaged and transported to the microbiology laboratory of the University of Calabar within 24hrs. These samples were immediately inoculated into plates containing freshly prepared sabouraud dextrose agar, using the streak plate technique after which the plates were incubated aerobically at 37^0C for 48hrs as previously described (Benson, 2001).

Microbiological analysis of the hospital air

This was performed following procedures described by Center for Disease Control and Prevention (CDC, 2003). Briefly, plates containing freshly prepared SDA in triplicates were exposed to the air of each hospital unit for 1hr and then immediately transported to the laboratory and incubated for 48hrs at 37^0C.

Enumeration of fungal and macroscopy

After incubation, discrete colonies on each plate were counted as described previously by Benson (2001). Each of the discrete colonies was then transferred using a sterile inoculating loop into plates containing freshly prepared SDA and incubated 37^0C for 48hrs. After incubation, the colony morphologies of the isolates were recorded and compared with descriptive features (Benson, 2001).

Purification and storage of fungal isolates

This was done following methods described by Benson (2001). Briefly, isolates were sub-cultured into plates containing freshly prepared sabouraud dextrose agar and incubated at 37^0C for 48hrs. Discrete colonies of the isolates were sub-cultured into bijou bottles containing sabouraud dextrose agar slants and incubated for 72hrs at 37^oC.

Slide culture and staining

This was carried aseptically as described by Matnani et *al* (2012). Briefly, a pair of forceps was used to aseptically place a filter paper in a sterile petri dish and 4mls of distilled water used to moisten the filter paper. Then, a U-shaped glass rod was placed on the filter paper and a sterile grease free slide placed on the U-shaped rod. Using a scalpel, 5mm square block of freshly prepared SDA was cut and placed at the centre of a microscope slide. With a sterile inoculating needle, the block of medium was inoculated with the test organism on all the four sides of the block and then a sterile cover slip was placed on the upper surface of the agar cube and the petri dish was covered and incubated at room temperature for 48-72 hours. After incubation, the slide and cover slip were heat fixed and stained with a drop of cotton blue in lacto-

phenol stain while the block of medium containing the inoculum was removed from the slide and discarded. Both stained slides were then viewed under the microscope and the growths compared.

Germ tube test for *Candida species*

This was performed following methods described by Matnani *et al* (2012). Briefly, 0.5ml of human serum was dispensed into a test tube and using a sterile wire loop, a colony of the test organism suspected to be *Candida species* was inoculated into the serum. The tube was then incubated at 35-37^{0}C for 2-3 hours. After incubation, a loopful of yeast inoculated serum was placed on a slide, covered with a cover-slip and observed under low and high magnification for germ tubes. A positive test gave presumptive identification of *C. albicans*.

Preparation of the antifungal disks and inoculum

All antifungal drugs were obtained as standard powders and prepared by dissolving them in specific solvents (DMSO, water, and ethanol), after which, they were loaded into blank paper disks at the following potencies: griseofulvin (25µg/disk), fluconazole (25µg/disk) amphotericin B discs (20µg) and ketoconazole (10µg/disk) according to antifungal disks potency of Rosco Diagnostica Company (Neosensitabs, Denmark). The isolates from SDA slants were sub-cultured to plates to enhance sporulation. Seven day-old cultures were covered with 1ml distilled water and the colonies were probed with the tip of a sterile Pasteur pipette to obtain a mixture of mycelium and conidia. The suspensions were transferred to sterile tubes and allowed to sediment for 30 minutes and then adjusted to match with 0.5 McFarland standards (CLSI, 2002).

Disk Diffusion Assay

All the tests were performed and criteria of susceptibility and resistance of antifungal disks were measured according to CLSI (2002).

2.3.3 Minimum inhibitory and bactericidal concentrations (MICS AND MBCS) of antifungal agents against fungi

Fungal isolates were subjected to antifungal sensitivity testing using the methods already described by CLSI (2002) with some modifications in the medium used. Briefly, ketoconazole, amphotericin B, griseofulvin and nystatin in their various potencies were diluted in 5ml of diluents to prepare stock solutions. Exactly 2ml from each stock solution was used to run a 4 fold doubling dilution which were (4, 8, 16, 32, µg/ml) in tubes containing 2ml of diluents respectively. Then, 0.1ml of the various antifungal concentrations was placed in tubes respectively. About 0.1ml of DMSO and water were also used to prepare controls in separate tubes. Within 15mins after the inoculum was standardized, 0.9ml of the inoculum suspension was added to each tube in the dilution series and mixed, resulting in a 1:10 dilution of each antifungal concentration and a 10% dilution of the inoculum. The tubes were then incubated at 35^{0}C for 46-72hours. After incubation period, the results were recorded and compared with the control to determine the minimum inhibitory concentration (MIC) of the antifungal used. This procedure was then repeated for all isolates.

RESULTS

The results of the study are presented in the tables below. Table 1 shows the mean load of fungi isolated from both hospital locations. The results showed that only the sampled laboratory coats in pharmacy unit of general hospital was free of fungi. The rest of the units and location showed the presence of fungi with the air environment having the highest mean load. The cultural and microscopic characteristics and the

presumptive isolates are presented in tables 2 and 3, respectively. Both tables showed the presence of Aspergillus species, *Penicillum species*, *Candida species* and *Rhizopus species*. Table 4 and 5 showed the prevalence rate of the fungi in the sampled sites and units of both hospitals respectively. Table 6 shows the resistance of the fungal isolates to routinely used anti-fungal agents.

Table 1: Mean loads of fungi isolated from General Hospital and IDH according to sampled sites/units

General Hospital Units	Table tops	Laboratory coats	Door handles	Sinks	Air	Mean Load/unit	%
Pharmacy	4	-	2	1	18	25	6.5
Theatre	2	7	5	5	12	31	8.0
Laboratory	10	5	12	7	60	94	24.4
Male Ward	4	22	4	10	40	80	20.7
Blood bank	13	6	13	16	26	74	19.2
ICU	15	2	8	12	45	82	21.2
Mean load/site	48	42	44	51	201	386	
Percentage (%)	12.4	10.9	11.4	13.2	52.1		
Infectious Disease Hospital units							
Pharmacy	26	10	3	35	40	114	27.4
Laboratory	12	8	7	9	34	70	16.8
Male Ward	14	5	14	21	20	74	17.8
Blood bank	27	2	9	10	35	83	20.0
ICU	21	7	5	11	31	75	18.0
Mean load/site	100	32	38	86	160	416	
Percentage (%)	24.0	7.7	9.0	20.7	38.5		

Table 2: Probable isolates using Cultural characteristics on SDA

General hospital	**IDH**	**Presumptive organisms**
$W_{sp,}$ L_{sp}	-	*A. niger*
ICU_{sp}	-	*A. fumigatus*
P_8	-	*A. flavus*
$BB_{sp,}$ ICU_{sp} W_{sp}	W_{19}, W_{sp} , L_{sp}	*Penicillum species*
$W_{17,}$,L_{sp}	L_{17}	*Penicillum species*
W_3, W_{sp}, P_{sp}, P_{20}, T_{sp}, ICU_{sp}, ICU_{sp}, L_{sp}.	L_{18}, W_{19}, W_{11}, ICU_{sp}.	*Candida species*
-	W_5, P_7, W_{18},	*Rhizopus species*

P_{20}.

KEY: P -Pharmacy, ICU-Intensive care unit, L-Laboratory, W-Ward, BB-Blood Bank, T-Theatre, 1-10-Table tops, 11-15-Laboratory coat, 16-17-Door handles, 18-20-sinks, sp-settled plates.

Table 3: Probable isolates using microscopic Characteristics of Fungal Isolates

General Hospital	IDH	Probable organism
$W_{sp,}$ L_{sp}	-	*A. niger*
P_8	-	*A. fumigatus*
ICU_{sp}		*A. flavus*
-	W_5, P_7, W_{18}, P_{20}.	*Rhizopus species*
W_3, W_{sp}, P_{sp}, P_{20}	L_{18}, W_{19}, W_{11},	*C. albicans*
T_{sp}, ICU_{sp}	-	*C. tropicalis*
L_{sp}.	ICU_{sp}	*C. glabrata*
ICU_{sp}	-	*C. krusei*
W_{17},L_{sp}	L_{17}	*P. notatum*
BB_{sp}, ICU_{sp}, W_{sp}	W_{19}, W_{sp}, L_{17}	*P. chrysogenum*

KEY: P -Pharmacy, ICU-Intensive care unit, L-Laboratory, W-Ward, BB-Blood Bank, T-Theatre, 1-10-Table tops, 11-15-Laboratory coat, 16-17-Door handles, 18-20-Sinks and sp-Settled plates.

TABLE 4: Prevalence of fungi per sampled site/material from both hospitals

Organisms	Table tops	Sinks	Laboratory Coat	Door handle	Air
GH (n -17,58.6%	(n -2, 11.8%)	(n -1, 5.9%)	(n -0, 0.0%)	(n -1, 5.9%)	(n-13,76.5%
Penicillum species (5)	0(0.0)	0(0.0)	0(0.0)	1(20.0)	4(80.0)
Candida species(8)	1(12.5)	1(12.5)	0(0.0)	0(0.0)	6(75.0)
Aspergillus species(4)	1(25.0)	0(0.0)	0(0.0)	0(0.0)	3(75.0)
IDH (n -12, 41.3%)	(n -2, 16.7)	(n -3, 25.0%)	(n -1, 8.3%)	(n -1, 8.3%)	(n -5, 41.7%)
Penicillum species (4)	0(0.0)	1(25.0)	0(0.0)	1(25.0)	2(50.0)

Candida species(4)	0(0.0)	2(50.0)	1(25.0)	0(0.0)	1(25.0)
Rhizopus species(4)	2(50.0)	0(0.0)	0(0.0)	0(0.0)	2(50.0)

TABLE 5: Prevalence of fungi in sampled units of both hospitals

Organisms	Pharmacy	ICU	Blood bank	Theatre	Laboratory	Ward
GH (n -17, 58.6%)	(n -3, 17.6%)	(n -4,23.5%)	(n -1, 5.9%)	(n -1,5.9 %)	(n -3,17.6 %)	(n -5, 29.4 %)
Candida species (8)	2(25.0)	2(25.0)	0(0.0)	1(12.5)	1(12.5)	2(25.0)
Penicillum species(5)	0(0.0)	1(20.0)	1(20.0)	0(0.0)	1(20.0)	2(40.0)
Aspergillus species (4)	1(25.0)	1(25.0)	0(0.0)	0(0.0)	1(25.0)	1(25.0)
IDH (n -12, 41.3%)	(n -2, 16.7%)	(n -1, 8.3%)	(n -0, 0.0%)	NA	(n -3, 25.0%)	(n -6, 50.0%)
Penicillum species (4)	0(0.0)	0(0.0)	0(0.0)	NA	2(50.0)	2(50.0)
Candida species(4)	0(0.0)	1(25.0)	0(0.0)	NA	1(25.0)	2(50.0)
Rhizopus species(4)	2(50.0)	0(0.0)	0(0.0)	NA	0(0.0)	2(50.0)

Table 6: Resistance pattern of fungal isolates to antifungal agents

ORGANISMS	K	F	GF	AP	NS	ORGANISMS	K	F	GF	AP	NS
	GENERAL HOSPITAL						**INFECTIOUS DISEASE HOSPITAL (IDH)**				
Candida albicans(4)	3(75.0)	1(25.0)	1(25.0)	2(50.0)	2(50.0)	*C. albican (3)*	2(66.6)	1(33.3)	0(0.0)	0(0.0)	3(100)
C. tropicalis (2)	0(0.0)	0(0.0)	1(50.0)	1(50.0)	0(0.0)	*C. glabrata(1)*	1(100)	1(100)	1(100)	1(100)	0(0.0)
C. glabrata (1)	1(100)	0(0.0)	0(0.0)	1(100)	1(100)	-	-	-	-	-	-
C. krusei (1)	0(0.0)	1(100)	0(0.0)	1(100)	1(100)	-	-	-	-	-	-
Penicillum notatum (2)	2(100)	1(50.0)	2(100)	1(50.0)	1(50.0)	*Penicillum notatum* (1)	1(100)	1(100)	0(0.0)	1(100)	1(100)
Penicillum chrysogenum	3(100)	2(66.7)	0(0.0)	1(33.3)	2(66.7))	*P. chrysogenu*	2(66.7)	2(66.7)	1(33.3)	0(0.0)	1(33.3)

(3)						*m* (3)					
Aspengillus niger (2)	2(100)	1(50.0)	0(0.0)	1(50.0)	2(100)	-	-	-	-	-	-
Aspergillus flavus (1)	1(100)	1(100)	0(0.0)	0(0.0)	1(100)	-	-	-	-	-	-
Aspergillus fumigatus (1)	0(0.0)	0(0.0)	1(100)	1(100)	0(0.0)	*Rhizopus species (4)*	2(50.0)	1(25.0)	3(75.0)	2(50.0)	3(75.0)

KEY: K- Ketoconazole, F-Fluconazole, GF-Griseofulvin, AP-Amhotericin B, NS-Nystatin

DISCUSSION

The hospital environment has been observed to be a complex one containing virtually all kinds of microorganisms with bacteria and fungi being the dominant ones (Abbas, 2013). This study revealed that General Hospital (GH) and Infectious Disease Hospital (IDH) recorded total mean loads of 386cfu and 416cfu, respectively. The mean fungal counts for both hospitals revealed that air had the highest count range of 160 – 201cfu with General Hospital having the highest and was higher than 122.1cfu/m^3 previously reported (Sautour *et al*., 2009). This high mean count observed in air of the hospitals studied is unconnected to the fact that air although it does not readily support their growth is a good vector for fungi and other microorganisms. An earlier study confirmed that air borne molds and yeast are a potential risk to patients due to possible inhalation of conidia (Caggiano *et al.*, 2014). Microbial quality of air is considered a reflection of the hygienic condition of the hospital (Pasquarella *et al*., 2010) and as noted by Alireza *et al* (2012), the index of microbial air contamination increases during work activities. This may have influenced the high microbial contamination observed in this study. The presence or finding of fungal growths in other sampled sites / materials besides air is in accordance with Khan and Karuppayil (2012) assertion that fungi possess ability to grow on almost any material especially if the materials in question are wet or hygroscopic. The presence of fungi in should-be sterile areas is of great concern. The ICU of both recorded similar mean fungal counts while blood bank mean fungal count was higher in IDH (83 cfu/hr) and this was lower than pharmacy in the same hospital.

Fungi prevalence rates of 17(58.6%) and 12(41.4%) were observed in GH and IDH, respectively. The 41.4-58.6% prevalence rates observed in this study is higher than the 32% reported by Matnani *et al* (2012). Fungal contamination of the hospital environment is mostly due to factors including presence of construction activity as well as favourable micro climate such as the presence of warmth, moisture and construction activity (Cristina *et al*., 2006).

Evaluation of the sample sites/materials revealed that General hospital air recorded fungi prevalence range of 13(76.5%), table tops 2(11.8%) and (5.9%) from door handles and sinks, respectively. In IDH air recorded 5(41.7%), sinks 3(25.0%), table tops 2(16.7%) and 8.3% from laboratory coats and door handles, respectively. The prevalence of fungal contamination in the sampled sites/materials of both

hospitals were much lower than the 96% earlier reported (Gniadek and Macura, 2007) but lower than the 13.1% reported by Caggiano *et al* (2014). An evaluation of the sampled units revealed that the male ward unit of GH recorded the highest prevalence of 5(29.4%) followed by ICU 4(23.5%), pharmacy and laboratory units recorded 3(17.6%), respectively while the blood bank and theatre units recorded the least with a frequency of 1(5.9%) each. The male ward of IDH had the highest prevalence of 6(50.0%) followed by laboratory unit 3(25.0%), pharmacy 2(16.7%) and ICU 1(8.3%). Our findings for theatre are consistent with reports of Sautour *et al* (2009) and Cristina *et al* (2006).

Most frequent organisms in the pharmacy, intensive care, theatre, and ward units of General hospital were *Candida species, Aspergillus species* from the laboratory unit and *Penicillum species* in the blood bank. On the other hand, *Rhizopus species* dominated the pharmacy unit and *Candida species* in ICU. In the ward unit of IDH, *Penicillum species, Candida species* and *Rhizopus species* were most frequent. In total the fungal species identified in all sampled units were 41.4% and 58.6% yeast and moulds, respectively. Although the *Candida species* were isolated from indoor environment in five selected units from five tested locations of hospital surfaces and air., studies have shown that candidiasis is an established endogenous and exogenous infection (Lutz *et al*., 2003) and have been shown to survive in air up to 150 days and capable of causing both surgical site and urinary tract infections (Garcia-cruz *et al*., 2012). The high prevalence of *Aspergillus and Penicillum species* found in the indoor environment tested in this study conforms to the report of Abbas (2013) and those of Caggiano *et al* (2014) and Verde *et al* (2015) respectively. The former is a potential pathogen in immune-compromised person but not in immuno-competent individuals and can pose respiratory risk as toxins and allergens (Abbas, 2013).

Candida species as noted by Tscherner *et al* (2011) is the fourth leading cause of hospital acquired systemic infections with mortality rates of up to 35-40% and with various levels of resistance to antifungal agents. *C. albicans* isolated from General hospital exhibited 75% resistance to ketoconazole, 50% to amphotericin B and nystatin and 25% to fluconazole and griseofulvin. Those from IDH exhibited 100% resistance to nystatin, 66.6% to ketoconazole and 33.3% to fluconazole. The 25-75% resistance to azoles observed in this study is consistent with reports of Ramesh *et al* (2011) who ascribed their resistance to either mutation or over expression of *ERG11* which encode *14α* demethylase in ergosterol biosynthetic pathway (Watamoto *et al*., 2013). The range of resistance to azoles recorded in this study is consistent with 11.9-41.1% reported by Ramesh *et al* (2011) but higher than the 6.67% recorded by Biernasiuk *et al* (2013) and 1-2.1% by Kanafani and Perfect (2008). In this study, most *C albicans* strains resistant to ketoconazole also showed resistance to fluconazole and this was consistent with those of Sheik *et al* (2013).

C. tropicalis isolated only from General hospital recorded 50% resistance to griseofulvin and amphotericin B. This is extremely higher than the 1.4-6.6% reported by Kanafani and Perfect (2008). *C. glabrata* recovered from both hospitals exhibited 100% resistance to ketoconazole, amphotericin B and nystatin. Tscherner *et al* (2011) revealed that *C. glabrata* possess intrinsic resistance to azoles especially when subjected to prolonged therapy with these drugs.*C. krusei* recovered only from General hospital exhibited resistance to fluconazole, amphotericin B and nystatin. The resistance of *C. krusei* to fluconazole employed in this study is consistent with 66.6%

reported by Badiee and Alborzi (2011). While the resistance to amphotericin B in this study is consistent with 3.1% resistance reported by Badiee and Alborzi (2011).

As observed in this study, *Penicillum species* exhibited varying degrees of resistance to all the antifungal agents employed in this study. The resistance of these species to antifungal agents may not be considered significant with the exception of *P. marneffei*. *A. fumigatus have* been shown to be associated with life threatening pulmonary infection most especially in immune deficient individuals. In General hospital, *A. fumigatus* exhibited resistance to amphotericin B and griseofulvin while *A. niger* recorded 100% resistance to nystatin and ketoconazole and 50% to amphotericin B and fluconazole. *A. flavus* on the other hand recorded resistance to azoles and polyenes. The finding of 0% resistance to azoles by *A. fumigatus* in this study is consistent with 0-26% reported by Chowdhary *et al* (2013). However, the resistance of *Aspergillus species* to polyenes observed in this study is consistent with reports of Hedayati *et al* (2007). Cannon *et al* (2009) further added that resistance of these strains (*A. flavus*) to polyenes is largely due to reduction in ergosterol content in the plasma membrane coupled with alterations that often occurs in the cell wall glucans. An important member of the *Mucor* family is the *Rhizopus species* exhibited 75% resistance to nystatin and griseofulvin, 50% to amphotericin B and ketoconazole and 25% to fluconazole.

CONCLUSION

This study revealed gross contamination of the environment of the hospitals studied with GH being more contaminated than IDH. The findings further confirm that the hospital environment plays a significant role in hospital associated infections. Cross contamination between patients, staff and surfaces was subsequently shown to have occurred. The effectiveness of environmental cleaning is an important factor in strategies to prevent hospital associated infections**.**

Conflict of Interest.

We hereby declare that there is no conflict of interest.

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