Antifungi Pattern of Candida Species Isolated From the Children Diapers in Obafemi Awolowo University Teaching Hospital Complex, Nigeria

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ABSTRACT
Candidiasis is by far the most common type of yeast infection, these fungi live on all surfaces of our bodies, under certain conditions, they can become so numerous to cause infections, particularly in warm and moist areas, typical affected areas in babies include the mouth and diaper areas, an in vitro antifungal evaluation study was carried out on 48 strains of Candida species isolated from children diapers using disc diffusion method with five different antifungi drug such as Nystatin (50mcg), Amphotericin-B (50mcg), Fluconazole (10mcg), Miconazole (30mcg) and Voriconazole (1mcg), Candida albican 11 (22.9), Candida kruisei 23 (47.9), Candida tropicalis 1 (2.1), and Candida glabrata 13 (27.1). Miconazole had the highest overall sensitivity 100.0% and the least Fluconazole 12.5%. Fluconazole had the highest overall resistant 87.5 % and the least Miconazole 4.2 % in sensitivity within the drugs. Nystatin: C. kruisei had the highest 48.7 % and the least 2.6 %, Amphotericin B: C. kruisei had the highest 48.7 % and the least C. tropicalis 2.6 %, Miconazole: C. kruisei had the highest 45.7 % and the least C. tropicalis 2.2 %, Voriconazole: C. kruisei had the highest 47.4 % and the least C. albican 21.1% Fluconazole: C. glabrata had the highest 50.0 % and the least C. kruisei 16.7., in sensitivity within organisms: Candida albican: Miconazole had the highest sensitivity 100.0 % and the least Fluconazole 18.2%. C. kruisei: Nystatin and Amphotericin B had 82.6% each, voriconazole 78.3%, and the least Fluconazole 4.3 %. C. tropicalis: Nystatin, Amphotericin B and miconazole had 100.0 each. C. glabrata: miconazole had 100.0 % and the least Fluconazole 23.1 %, in resistance within drugs, Nystatin: C. kruisei had the highest resistance 44.4 %, the least C. glabrata 22.2 %, Amphotericin B: C. kruisei 44.4 %, the least C. albican 22.2 %. Miconazole: C. kruisei 100.0 %, voriconazole: C. kruisei 50.0 %, the least, 10.0% for both: C. tropicalis and C. glabrata. Fluconazole: C. kruisei 52.4 %, then the least C. tropicalis 2.4 %, in resistance within organisms, in C. albican: fluconazole had the highest 81.8 %, Nystatin and Voriconazole 27.3 % each, C. kruisei: Fluconazole had 95.7 % voriconazole 21.7 % while both Amphotericin B and Nystatin had 17.4 % each. C. tropicalis: Fluconazole and Voriconazole had 100.0 %, C. glabrata: Fluconazole had 76. % and the least 7.7 %. In conclusion, in this study, Miconazole which had the highest overall sensitivity to different Candida species is therefore recommended as the most suitable medication for the treatment of candidiasis.

Keywords: Antifungal, Candida species, Children, Diaper

Introduction
Disposable diapers were first produced in the 1940s, but were initially considered to be luxury items. It was not until the 1960s that they began to be used on a mass scale. By then, diapers were made with layers of cellulose, which made them more absorbent and resistant. (Ward et al, 2000, [1] Wolf R et al, 2000 [2]. However, they can also cause diaper dermatitis, also known as diaper rash, which can be associated with different infections, especially Candida infections.
thrive at skin pH, which is around 5.5, or 6.0 in newborns (owing to vernix caseosa and amniotic fluid), tending to normalize in a few days (Brook I, 1992 [8] Hoppe J.E 1997 Ferrazzini, et al. 2003) [9] C. albicans and other yeasts provide examples of perfect adaptation to pH changes, which is controlled by two genes: PHR2, which is activated in acidic environments and is deactivated when pH increases, and PHR1, which does the opposite, i.e. is activated at a high pH (neutral and basic levels) De Bernardis et al, 1998 [10] Another proven factor in developing fungi (yeasts and dermatophytes) is CO2 levels, which are higher in the occlusive environment of standard disposable diapers and barely detectable in breathable diapers (Odio and Friedlander 2000 [11] Allen and King 1978 [12] Akin et al, 2001) [13] The primary treatment for Candida diaper rash involves antifungal topical treatment and decreasing moisture in the diaper area. Nystatin (Mycostatin), Clotrimazole (Lotrimin), and Miconazole (Micatin, Monistat-Derm) are topical over-the-counter (nonprescription) treatments of equal strength for treating Candida diaper dermatitis. Occasionally, other prescription antifungal creams, such as ketoconazole (Nizoral cream) and econazole (Spectazole) may be necessary. This research is aimed at knowing the sensitivity and resistivity patterns of some selected antifungal drugs on the isolated Candida species from the diaper rash.

Materials and Methods

Study area: The study was conducted at the Obafemi Awolowo University Teaching Hospital Complex (OAUTHC), Ile-Ife, southwest of Nigeria. This is a 600 bedded hospital that serves as referral center to about 5 neighboring states of the country.

Study population: A total of forty-eight (48) isolated species of Candida from the children diapers attending Dermatology Clinic of the OAUTHC Ile-Ife were used in this study. The study was conducted between September and December 2018.

Identification of the Candida Species
The Identification of the Candida species were carried out by plating each Candida species on the prepared Hi Chrome Candida Differential Agar (M1297A) in a Petri dish and incubated for 12 hours. After incubation, the organisms come up with the colour specific for each species as indicated by the Manufacturer to give (11 Candida albicans, 23 Candida krusei, 1 Candida tropicalis and 13 Candida glabrata). There are no competing interests. The results were analyzed using SPSS package.

Anti-Fungi Sensitivity
An in vitro evaluation study was performed using Disc diffusion method and five different anti fungi drug such as Nystatin (SD 271-1VL, 50mcg), Amphotericin-B (SD 270-1VL, 50mcg). Fluconazole (SD 114-1VL, 10mcg), Miconazole (SD 273-1VL, 30mcg) and Voriconazole (SD 277-1VL, 1mcg) were used.

Results
In an in vitro evaluation study carried Out on 48 strains of Candida isolated from children diapers, Candida albicans 11 (22.9), Candida krusei 23 (47.9), Candida tropicalis 1 (2.1), and Candida glabrata 13 (27.1).

Miconazole had the highest overall sensitivity 46 (100.0) followed by Nystatin and amphotericin B 39 (81.3) each, Voriconazole 38 (79.2) and Fluconazole 6 (12.5). Table 1.

Fluconazole had the highest overall resistant 42 (87.5), Voriconazole 10 (20.8) Nystatin and Amphotericin B 9 (18.8) each and Miconazole 2 (4.2) figure 1.

Table 1: General antifungal sensitivity pattern of Candida species isolated from children diaper

<table>
<thead>
<tr>
<th>Organisms</th>
<th>Frequency (%)</th>
<th>Nystatin (Sens) %</th>
<th>Amphotericin B (Sens) %</th>
<th>Miconazole (Sens) %</th>
<th>Voriconazole (Sens) %</th>
<th>Fluconazole (Sens) %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Candida albicans</td>
<td>11.0 (22.9)</td>
<td>8.0 (16.7)</td>
<td>9.0 (18.8)</td>
<td>11.0 (22.9)</td>
<td>8.0 (21.1)</td>
<td>2.0 (4.2)</td>
</tr>
<tr>
<td>Candida krusei</td>
<td>23.0 (47.9)</td>
<td>19.0 (39.6)</td>
<td>19.0 (39.6)</td>
<td>21.0 (43.8)</td>
<td>18.0 (47.4)</td>
<td>1.0 (2.1)</td>
</tr>
<tr>
<td>Candida tropicalis</td>
<td>1.0 (2.1)</td>
<td>1.0 (2.1)</td>
<td>1.0 (2.1)</td>
<td>1.0 (2.1)</td>
<td>0.0 (0.0)</td>
<td>0.0 (0.0)</td>
</tr>
<tr>
<td>Candida glabrata</td>
<td>13.0 (27.1)</td>
<td>11.0 (22.9)</td>
<td>10.0 (20.8)</td>
<td>0.0 (0.0)</td>
<td>12.0 (35.0)</td>
<td>3.0 (6.3)</td>
</tr>
<tr>
<td>Total</td>
<td>48.0 (100)</td>
<td>39 (81.3)</td>
<td>39 (81.3)</td>
<td>46.0 (100)</td>
<td>38.0 (79.2)</td>
<td>6.0 (12.5)</td>
</tr>
</tbody>
</table>

Figure 1: General antifungal percentage resistant pattern of Candida species isolated from children diaper.

Sensitivity within the drugs:
Nystatin: C. krusei had the highest 48.7%, C. glabrata 28.2%, and the least 2.6% Amphotericin B: C. krusei had the highest 48.7%, C. glabrata 25.6% and the least C. tropicalis 2.6% Miconazole: C. krusei had the highest 45.7%, C. glabrata 28.3%, and the least C. tropicalis 2.2% Voriconazole: C. krusei had the highest 47.4%, C. glabrata 13.6% and the least C. albican 21.1% Fluconazole: C. glabrata had the highest (50.0%, C. albican 33.3%, and the least C. krusei 16.7%.

<table>
<thead>
<tr>
<th>Organisms</th>
<th>Frequency (%)</th>
<th>Nystatin (Sens) %</th>
<th>Amphotericin B (Sens) %</th>
<th>Miconazole (Sens) %</th>
<th>Voriconazole (Sens) %</th>
<th>Fluconazole (Sens) %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Candida albican</td>
<td>11 (22.9)</td>
<td>8.0 (20.5)</td>
<td>9.0 (23.1)</td>
<td>11.0 (23.9)</td>
<td>8.0 (21.1)</td>
<td>2.0 (33.3)</td>
</tr>
<tr>
<td>Candida Krusei</td>
<td>23 (47.9)</td>
<td>19.0 (48.7)</td>
<td>19.0 (48.7)</td>
<td>21.0 (45.7)</td>
<td>18.0 (47.4)</td>
<td>1.0 (16.7)</td>
</tr>
<tr>
<td>Candida tropicalis</td>
<td>1 (2.1)</td>
<td>1.0(2.6)</td>
<td>1.0 (2.6)</td>
<td>1.0 (2.2)</td>
<td>0.0 (0)</td>
<td>0.0 (0)</td>
</tr>
<tr>
<td>Candida glabrata</td>
<td>13 (27.1)</td>
<td>11.0 (28.2)</td>
<td>10.0 (25.6)</td>
<td>13.0 (28.3)</td>
<td>12.0 (31.6)</td>
<td>3.0 (50.0)</td>
</tr>
<tr>
<td>Total</td>
<td>48 (100)</td>
<td>39.0 (100)</td>
<td>39.0 (100)</td>
<td>46.0 (100)</td>
<td>38.0 (100)</td>
<td>6.0 (100)</td>
</tr>
</tbody>
</table>

Sensitivity within organisms:
Candida albican: Miconazole had the highest sensitivity 100.0%, Amphotericin B 81.8%, Nystatin and voriconazole 72.7% each and the least Fluconazole 18.2%. C. krusei: Nystatin and Amphotericin B had 82.6% each, voriconazole 78.3%, and the least Fluconazole 4.3%. C. tropicalis: Nystatin, Amphotericin B and miconazole had 100.0% each. C. glabrata: miconazole had 100.0% voriconazole 92.3%, Nystatin 84.6% and the least Fluconazole 23.1%.

<table>
<thead>
<tr>
<th>Organisms</th>
<th>Frequency (%)</th>
<th>Nystatin (Sens) %</th>
<th>Amphotericin B (Sens) %</th>
<th>Miconazole (Sens) %</th>
<th>Voriconazole (Sens) %</th>
<th>Fluconazole (Sens) %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Candida albican</td>
<td>11 (22.9)</td>
<td>8.0 (72.7)</td>
<td>9.0 (81.8)</td>
<td>11.0 (100.0)</td>
<td>8.0 (72.7)</td>
<td>2.0 (18.2)</td>
</tr>
<tr>
<td>Candida Krusei</td>
<td>23 (47.9)</td>
<td>19.0 (82.6)</td>
<td>19.0 (82.6)</td>
<td>21.0 (91.3)</td>
<td>18.0 (78.3)</td>
<td>1.0 (4.3)</td>
</tr>
<tr>
<td>Candida tropicalis</td>
<td>1 (2.1)</td>
<td>1.0(100.0)</td>
<td>1.0 (100.0)</td>
<td>1.0 (100.0)</td>
<td>0.0 (0)</td>
<td>0.0 (0)</td>
</tr>
<tr>
<td>Candida glabrata</td>
<td>13 (27.1)</td>
<td>11.0 (84.6)</td>
<td>10.0 (76.9)</td>
<td>13.0 (100.0)</td>
<td>12.0 (92.3)</td>
<td>3.0 (23.1)</td>
</tr>
<tr>
<td>Total</td>
<td>48 (100)</td>
<td>39.0 (81.3)</td>
<td>39.0 (81.3)</td>
<td>46.0 (95.8)</td>
<td>38.0 (79.2)</td>
<td>6.0 (12.5)</td>
</tr>
</tbody>
</table>

Resistance within Drugs
Nystatin: C. krusei had the highest resistance 44.4%, C. albican 33.3% the least C. glabrata 22.2%, Amphotericin B: C. krusei 44.4%, C. glabrata 33.3%, then the least C. albican 22.2%, Miconazole: C. krusei 100.0%, voriconazole: C. krusei 50.0% C. albican 30.0%, the least, 10.0% for both C. tropicalis and C. glabrata. Fluconazole: C. krusei 52.4%, C. glabrata 23.8% then the least C. tropicalis 2.4%.

Resistance within Organisms
The highest resistance was seen in C. albican; fluconazole 81.8%, Nystatin and Voriconazole 27.3% each, C. krusei: Fluconazole had 95.7% voriconazole 21.7% while both Amphotericin B and Nystatin had 17.4% each. C. tropicalis: Fluconazole and Voriconazole had 100.0%, C. glabrata: Fluconazole had 76.3% Amphotericin B 23.1% and the least 7.7%.

![Figure 2](image2.png)  
Figure 2: Resistant pattern of Candida species isolated from children diaper in percentage (%) within the Antifungal drugs used

![Figure 3](image3.png)  
Figure 3: Resistant pattern of Candida species isolated from children diaper in percentage (%) within the Organisms

Discussion

An invitro evaluation study was carried out on 48 strains of Candida spp isolated from children diapers in ile iife, In this study miconazole had the highest overall sensitivity 100.0% and the least was fluconazole 12.5%, this was in conjunctions with the report from world health organization that fluconazole resistance was indeed more common in non Candida albican species and this may pose negative signal since fluconazole was not that expensive and well tolerated medication that is given orally (WHO 2014). C. krusei is more sensitive to Nystatin 48.7% Amphotericin B 48.7%, Miconazole 45.7%, Voriconazole 47.4% while the C. glabrata is more sensitive to fluconazole 50% within the drugs used in the study. Within the organism used, C. albican had the highest sensitivity 100% Amphotericin B 81.8%, Nystatin and Voriconazole 72% and Fluconazole 18.2 %, this was in line with the findings of Jasem et al, 2014 in which the sensitivity to Voriconazole was higher with 94.3% but on the contrary, the sensitivity to Fluconazole 67.9% was more than what we got.

The highest resistance in the study was seen with Fluconazole 87.5 % whereas In the study done by Jasem et al, 2014 it was 35.8% and also, according to the work done by the CDC on the resistivity of the Candida isolates, where 70.0 % resistance was seen with C. glabrata and C. krusei but in the study it was seen with C. tropicalis and C. krusei. To all drugs used in the study, C. krusei had the highest resistance of 44.4 % on Nystatin and Amphotericin B, 100 % Miconazole, 50.0 % C. albican, 52.4 % fluconazole and 50.0 % Voriconazole. The resistance among the Candida species used was seen with Fluconazole, where C. albican had 81 %, C. krusei had 95.7 %, C. tropicalis had 100%, and C. glabrata had 76 %. The most common cause of candida infection C. albican in the study done by Mohamadi et al, 2014 had Nystatin to be 100% sensitive to C. albican whereas in the study it was found to be 72.7 % showing that there is increase in the resistivity to the drug.

In conclusion, in this study, over all resistance to different Candida species, Miconazole (100.0), Nystatin and amphotericin B (81.3) each, Voriconazole (79.2) and Fluconazole (12.5) which implies that Fluconazole that is commonly used had 87.5 resistance, it is therefore recommended according to the study that Miconazole is the most suitable medication for the treatment of especially diaper rash induce candidiasis. [14-19].

References


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