First Detection of Emergent Fungal Pathogen Candida auris in Khartoum State, Sudan

Ali M Badri*1 and Safa A Sherfi2
1Department of Microbiology, International University of Africa, Sudan
2Head Department of Basic Sciences, International University of Africa, Sudan

*Corresponding author: Ali M Badri, Department of Microbiology, Faculty of Medical Laboratory Sciences, International University of Africa, Khartoum, Sudan.


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Abstract

Background: Candida auris is an emerging multidrug-resistant yeast that can cause invasive infections and is associated with high mortality. Due to high mortality rates and multi-drug-resistant fungus this species is now attracting worldwide attention as Emergent fungal pathogen. The aim of this study to determine the prevalence of Candida auris Among Febrile Patients.

Methods: Blood samples from 100 Febrile Patients were collected and analyzed for the presence of C. auris DNA by PCR.

Results: PCR analysis revealed that 26 out of 100 patients (26%) were positive for C. auris infection.

Conclusion: High level of knowledge and alertness by physicians and healthcare workers, especially in critical care settings, would help to control the spread and improve diagnostic and therapeutic strategies.

Keywords: Candida auris; Candidemia; Febrile Patients; PCR; Khartoum; Sudan

Introduction

Fungal infections represent an important cause of human morbidity and mortality. C. auris is an emerging fungal pathogen that has attracted considerable attention because of its ability to cause infections that are difficult to diagnose and treat [1]. It was first described in 2009 after being isolated from the external ear discharge of a patient in Japan [2]. Outbreaks of C. auris infections have also been reported in patients from United States [3,4], United Kingdom [5,6], South Korea [7,8], South Africa [9,10], India [11,12], Pakistan [9], Israel [13], Venezuela [9,14], Spain [15], Colombia [16], and Kuwait [17]. C. auris is primarily detected in patients with a long period of hospitalization in intensive care units (ICU). C. auris has now been recognized as an important nosocomial pathogen in many countries causing chronic otitis media, bloodstream infections, ventriculitis, osteomyelitis, otomastoiditis, intra-abdominal infections, pleural effusion vulvovaginitis and pericarditis [18,19]. C. auris is resistant to fluconazole (FLU) and is also reported as MICs to all three major antifungal classes [7,20-23] so it may be difficult to start an adequate empirical therapy without accurate species identification. C. auris is usually misidentified by routinely used phenotypic methods in clinical microbiology laboratories. Biochemical assays as one of the most widely used phenotypic approaches, besides of being time consuming and expensive, cannot properly identify [24]. The development of specific PCR assays for C. auris and for C. auris-related species using cultured colonies seems promising for its rapid and accurate identification, particularly in outbreak settings [25,26]. Molecular identification of C. auris can be performed by sequencing various genetic loci, but it is not routinely used. This study was aimed to determine the prevalence of C. auris using PCR in Febrile Patients in Khartoum State, Sudan.

Materials and Methods

This was a health facility based descriptive Cross-sectional study: conducted in Khartoum State during the period from January to April 2019. A total of 100 blood samples were collected from febrile patients including 60 males and 40 females with ages ranged from 5 years to 65 years old. complained of fever and other symptoms like headache and general body pain, patient’s positive
for Malaria and Typhoid fever were excluded from the study. A structured questionnaire and referring to the patient clinical sheet were being used; demographic data and other data (Symptoms, using of drug, residence, etc.). From each patient, 3 ml of venous blood sample was collected in sterile EDTA blood containers and frozen at -20°C until used for DNA extraction. Ethical approval of the study in accordance with the guidelines of the ethical considerations and Patients consent was obtained before the commencement of the study.

**DNA Extraction**

DNA was extracted by using commercial DNA extraction kit (Intron, Korea) according to the manufacturers’ instructions and stored at -20°C until used.

**Polymerase Chain Reaction (PCR)**

PCR was performed and the test was carried out using following primers: forward: 5’- CGCACATTGCGCCTTGGGGTA-3’ and reverse: 5’-GTAGTCCTACCTGATTGGA GCAGC-3’. Each reaction was performed in total volume of 25 µl, containing 5 µl master mix (Solis Bio dyne master mix), 2 µl of primer, 5 µl of DNA and 13 µl of distilled water. Reactions were performed using PCR machine Techne (UK) under the following cycling conditions: 2 min at 95°C followed by 30 cycles of 30s at 95°C, 30s at 55°C and 1min at 72°C and final step of 7min at 72°C. 5 µl of the PCR product was analyzed using 1.5% Agarose gel electrophoresis and stained with 0.15% Ethidium bromide and the product was visualized using UV gel documentation. positive reaction was confirmed by the present of 163 bp (Figure 1).

**Results and Discussion**

The results of the current study showed prevalence of C. auris-DNA among 26 (26%) febrile patients and negative results among 74 (74%) of the total samples. The results were also clarified according to the gender and age group criteria as the following, according to the gender the C. auris-DNA among male show positive in 15 (25%) individuals and negative in 45 (75%) individuals, the female show positive in 11(27.5%) individuals and negative in 62 (72.5%) individuals, according to the age group the study individuals was classified into three groups the first group < 30 show positive in 5 (5%) individuals, the second group 30 - 50 show positive results in 12 (12%)individuals, the last group > 50 show positive results in 9 (9%) individuals which clarified in (Table 1).

**Table 1: Demographic characteristics of the study participants.**

<table>
<thead>
<tr>
<th></th>
<th>C. auris positive (N=26)</th>
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<tbody>
<tr>
<td>Gender</td>
<td></td>
<td></td>
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<tr>
<td>Male</td>
<td>15(25)</td>
<td>45(75)</td>
</tr>
<tr>
<td>Female</td>
<td>11(27.5)</td>
<td>62(72.5)</td>
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<tr>
<td>Age group</td>
<td></td>
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<tr>
<td>&lt;30</td>
<td>5 (5)</td>
<td>16(16)</td>
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<tr>
<td>30 – 50</td>
<td>12(12)</td>
<td>44(44)</td>
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<tr>
<td>&gt;50</td>
<td>9(9)</td>
<td>14(14)</td>
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</tbody>
</table>

Candida auris has emerged as a multidrug-resistant yeast causing substantial mortality in health care settings worldwide. Rapid and accurate diagnosis of C. auris infections is crucial to achieve timely and adequate medical treatment and to control fungal outbreaks. Rapid identification of C. auris directly from patient samples is of primary importance for the administration of empirical antifungal therapy. The lack of reliable clinical diagnostic tools potentially led to underestimation and neglect of C. auris cases [27-29]. The ability of C. auris isolates to grow at 40-42°C, inability to produce hyphae or pseudo hyphae in culture and resistance to fluconazole should prompt clinical microbiology laboratories to seek identification of the yeast isolate by molecular methods [30-33]. The molecular methods provided rapid results within hours of sample processing and highly sensitive compared to the much slower culture results, which may take anywhere from 4 to 14 days and usually misidentified. This is first report regarding C. auris in the Sudan, in the current study the qualitative PCR procedure was used to detect C. auris DNA in blood samples. Specimens were obtained from 100 febrile patients. The results showed positive among 26 febrile patients and negative results among 74 of the total samples. according to the gender the C. auris-DNA among male show positive in 15 (25%) individuals and negative in 45 (75%) individuals, the female show positive in 11(27.5%) individuals and negative in 29 (72.5%) individuals, according to the age group the study individuals was classified into three groups the first group < 30 show positive in 5 (5%) individuals, the second group 30 - 50 show positive results in 12 (12%)individuals, the last group > 50 show positive results in 9 (9%) individuals which clarified in (Table 1).
was classified into three groups: the first group < 30 show positive results in 5 (5%) individuals, the second group 30 – 50 show positive results in 12 (12%) individuals, while the last group > 50 show positive results in 9 (9%) individuals. In most cases, clinical presentation is non-specific and it is often difficult to differentiate between other types of systemic infections. Most of the reported cases in the last 5 years were isolated from blood and other deep-seated sites of infection (including invasive devices and catheters tips). Since the first isolation, C. auris infections have been reported from many countries, including India [11,12], Pakistan [9], South Korea [7,8], Malaysia [34], South Africa [9,10], Oman [35,36], Kenya [37], Kuwait [17], Israel [13], United Arab Emirates [38], Saudi Arabia [39], China [40], Colombia [16], Venezuela [9,14], the United States (US), Spain [15]. The real prevalence and the epidemiology of C. auris still remain uncertain. One of the causes may be the underestimation of its isolation due to the limited accuracy of available conventional diagnostic tools [41].

Conclusion

C. auris is a well-known nosocomial global pathogen that has recently emerged as a significant threat. According to reports of recent outbreaks, colonization is difficult to eradicate, and it tends to persist for months. High level of knowledge and alertness by physicians and healthcare workers, especially in critical care settings, would help to control the spread and improve diagnostic and therapeutic strategies.

Conflict of Interest

Authors declare that no conflict of interest exist in this paper.

References


