

Original Research

Received 29 May 2026

Revised 18 July 2026

Accepted 08 August 2026

Natural Resources for Human Health 2026, Vol. 6 (14s): 1591–1601

KEYWORDS:

Antiseptic agents, Antifungal drug, Candida, CDR1 & ERG11, Otomycosis.

Molecular Detection of (ERG11 and CDR1) Genes and Evaluation of Alternative Therapeutic Agents against Candida Species Isolated from Otomycosis Patients

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ABSTRACT:

Objectives: Antifungal resistance among *Candida* species has become a major clinical challenge worldwide, particularly in patients with recurrent or progressive otomycosis. This growing problem highlights the need to explore new therapeutic strategies and alternative treatment options.

Aim: Detection of resistance genes in *Candida* species isolated from otomycosis patients in Basra Governorate hospitals and evaluation of some alternative treatments for antifungal-resistant isolates.

Method: A total of 114 ear swab samples were collected from patients diagnosed with otomycosis. *Candida* isolates were identified using conventional phenotypic methods and confirmed by sequencing of the ITS1-5.8S-ITS2 region. Antifungal susceptibility testing was performed using the VITEK[®] 2 Compact System, while the antifungal activity of selected non-specific antiseptic agents was evaluated by the agar well diffusion method. The *CDR1* and *ERG11* resistance genes were detected by conventional PCR to determine their prevalence among the isolates.

Results: Females were more affected (54.38%) than males (45.61%), with the highest infection rate observed in the 31-40 years age group (20.2%). Four nonspecific topical antifungal agents for the ear, 7.5% povidone iodine, 3% hydrogen peroxide, 2% acetic acid and 70% isopropyl alcohol showed the strongest inhibitory activity against *Candida* species isolates, The VITEK[®] 2 system evaluated variable susceptibility profiles among the isolates. Molecular identification revealed that *Candida albicans* (36.8%) was the most prevalent species, followed by *Candida parapsilosis* (26.3%), *Candida glabrata* (14.0%), and *Candida tropicalis* (12.4%), while *Candida krusei* showed the lowest prevalence (10.5%). PCR amplification revealed that the prevalence of the *CDR1* and *ERG11* resistance genes among the *Candida* species was 67.5% and 75.4%, respectively.

Conclusions: A high prevalence of the *CDR1* and *ERG11* resistance genes was observed among *Candida* species isolated from otomycosis patients. The evaluated topical antiseptic agents exhibited promising antifungal activity and may serve as effective alternative therapies, contributing to yeast growth control and reducing the risk of antifungal resistance development.

INTRODUCTION

Otomycosis is an inflammatory lesion caused by the invasion or excessive proliferation of pathogenic fungi in the external auditory canal (Al-Obaedi *et al.*, 2022). It is a globally prevalent infection, with incidence varying across geographic regions (Al-mosawi, 2018). The spectrum of yeast species causing human disease and the variety of clinical manifestations associated with these infections have significantly expanded (Klimas *et al.*, 2025). Epidemiologic trends exhibit big changes with emerging novel multidrug-resistant pathogens and broadening geographic ranges (Fisher *et al.*, 2022).

The molecular and cellular mechanisms behind the yeast resistance may involve several virulence factors, including increased mutations in target protein genes (Gnat *et al.*, 2021), efflux that reduces the drug accumulation and the bioavailability within the cell (Lee *et al.*, 2023), and changes in metabolic pathways, such as alterations in ergosterol biosynthesis, which is crucial for the structure and function of the yeast cell wall (Song *et al.*, 2025). Moreover, the emergence of antifungal failure in clinical medicine can also be related to an altered drug uptake, drug target alteration, or drug inactivation (Guerra *et al.*, 2025).

There are several clinical treatment protocols for both noninvasive and invasive otomycosis. The effective dose of antifungal drugs is close to the toxic dose (Hetta *et al.*, 2025). Treatment methods for otomycosis include removing the causative agent, thoroughly cleaning the ear canal, and administering antifungal medications, including specific antifungal agents and nonspecific antifungal agents (Abed *et al.*, 2015; Aziz *et al.*, 2020). Currently, antifungal options are limited and associated with high toxicity, increasing resistance, and substantial costs, representing a significant burden on public health systems (de Souza *et al.*, 2025). The rise of antifungal resistance has necessitated careful monitoring of yeast susceptibility, creating new antifungal drugs, and the adoption of prudent use strategies to reduce resistance (Abeed *et al.*, 2022; Zeynel *et al.*, 2025). In addition, advances in diagnostic testing, such as molecular techniques, help to identify resistant yeast faster and more accurately, making treatment more targeted.

Accordingly, the present study aimed to investigate the prevalence of antifungal resistance genes (*ERG11* and *CDR1*) among *Candida* species isolated from patients with otomycosis, to evaluate their susceptibility to commonly used antifungal agents, and to assess the in vitro efficacy of selected topical antiseptic agents as alternative or adjunctive therapeutic options for isolates exhibiting antifungal resistance.

METHODOLOGY

Clinical examinations and samples collection

The study was conducted from October 2024 to September 2025. Ear swabs were obtained from patients attending the ENT Department at Al-Basrah Teaching Hospital. All patients presenting with clinical manifestations suggestive of otomycosis were examined by an otolaryngologist. Ear swab specimens were collected from 114 patients aged between 7 and 75 years. The isolates were classified according to patient sex and age groups. In addition, demographic and clinical data were recorded using a structured questionnaire, including the presence of symptoms such as otorrhea, pruritus, inflammation, and hearing impairment.

Culture and microscopic examination

Sterile ear swab samples obtained from patients with otomycosis were directly inoculated onto Sabouraud Dextrose Agar (SDA) plates. The cultures were incubated at 37 °C for 2-8 days. After incubation, colony morphology was evaluated, and representative yeast colonies were stained with lactophenol cotton blue solution and examined under a light microscope for preliminary identification based on their microscopic features (Rashak *et al.*, 2024).

Evaluation of the antifungal activity of antiseptic agents

A standardized yeast suspension (approximately 6×10^6 cells/mL, adjusted to the McFarland standard) was uniformly inoculated onto Sabouraud Dextrose Agar (SDA) plates (Muhsin, R. 2012). Wells measuring 6 mm in diameter were prepared using a sterile cork borer. Equal volumes of 1% clotrimazole, 3% hydrogen peroxide, 7.5% povidone-iodine, 70% isopropyl alcohol, and 2% acetic acid were dispensed into the wells. The plates were allowed to stand at room temperature for 2 h to facilitate diffusion, then incubated aerobically at 35-37 °C for 24-48 h. Antifungal activity was evaluated by measuring the diameter of the inhibition zones (mm).

Antifungal susceptibility testing (AST) using the VITEK® 2 compact system

Antifungal susceptibility testing of the *Candida* isolates was performed using the VITEK® 2 Compact System following the manufacturer's protocol. The isolates were tested against fluconazole, voriconazole, flucytosine, amphotericin B, and caspofungin. Results were interpreted as susceptible (S), intermediate (I), or resistant (R) according to the CLSI (2026) guidelines.

Molecular identification

Genomic DNA was extracted from all yeast isolates using the Presto™ Mini gDNA Yeast Kit according to the manufacturer's protocol. DNA quality was verified by agarose gel electrophoresis (Jordan et al., 2015). Molecular identification was performed by amplification of the ITS1-5.8S-ITS2 region using PCR with the universal primers ITS1-F (5'-TCC GTA GGT GAA CCT GCG G-3') and ITS4-R (5'-TCC TCC GCT TAT TGA TAT GC-3') (Mirhendi et al., 2006). PCR products were purified and subjected to Sanger sequencing (Macrogen, South Korea). The obtained sequences were analyzed and compared with reference sequences in the NCBI GenBank database using the BLAST algorithm and BioEdit software for species confirmation.

ERG11 and CDR1 gene detection

Conventional PCR was used to detect the *ERG11* and *CDR1* genes in the *Candida* isolates. PCR reactions were prepared in a final volume of 25 µL containing Master Mix, gene-specific primers, template DNA, and nuclease-free water. The primer sequences used for amplification are presented in Table 1.

Table 1: Primer sequence of *CDR1* and *ERG 11* gene of *Candida* species

species	Primers sequences (5'-3')	Product size bp	References
<i>Candida albicans</i>	<i>CDR1</i> -F 5'- TGGTGT'TTCCGGTGGTGAAA -3'	503bp	(Kadhim, et al., 2023)
	<i>CDR1</i> -R 5'- CCCTGGTGT'TGGATCGTTCA -3'		(El Said et al., 2022)
	<i>ERG11</i> -F 5'- GCAGCTTCATATGGTCAACAACC-3'	100bp	
	<i>ERG11</i> -R 5'- TAACATTGGCAACCCCATGAG-3'		
<i>Candida glabrata</i>	<i>CDR1</i> -F 5'- ACGGTACCAAGCCATACGAG -3'	188 bp	(Pushparaj et al., 2025)
	<i>CDR1</i> -R 5'- GAACACTGGGGTGGTCAAGT-3'		
	<i>ERG11</i> -F 5'- AGCTGCTTACTCCCACTTGACC-3'	412 bp	(dos Santos Silva et al. 2016)
	<i>ERG11</i> -R 5'-AGCTTGTTGGGCATGGTCTCTC-3'		
<i>Candida krusei</i>	<i>CDR1</i> -F 5'- ATTGGTGGTTCTATGGCTGGT-3'	205bp	(Vanden Bossche et al., 1998)
	<i>CDR1</i> -R 5'-TCTTCTTCTGGTGCGTTGGT -3'		
	<i>ERG11</i> -F 5'- ACTCCTGTTTTTCGGTAAGGGCG-3'	421bp	(Vandeputte et al., 2005)
	<i>ERG11</i> -R 5'-CACCGGCACGCTTTGTATTG-3'		
<i>Candida tropicalis</i>	<i>CDR1</i> -F 5'- GATCGGGAATTGCTCACACT -3'	163bp	(Pushparaj et al., 2025)
	<i>CDR1</i> -R 5'- AATTTGCAGCCGTCAAAAAC -3'		
	<i>ERG11</i> -F 5'-ATCCCACAGGCTTATTTGAAA-3'	614bp	(Vandeputte et al. 2005)
	<i>ERG11</i> -R 5'-GGTCTCTTTCCTTGGTTTTTG -3'		
<i>Candida parapsilosis</i>	<i>CDR1</i> -F 5'- CTTCCGGTCACTTGAATGGT -3'	221bp	(Pushparaj et al., 2025)
	<i>CDR1</i> -R 5'- TCCTCCATAATGGGCTTGTC-3'		
	<i>ERG11</i> -F 5'-CGAGATAATCATCA ACGAACATTC-3'	648bp	(Karthika et al.. 2024)

	ERG11-R 5'-AAAGACCGCATTTGACTACCGAT-3'		
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Statistical Analysis

Data were analyzed using IBM SPSS Statistics version 23 for Windows. Descriptive statistics were presented as frequencies and percentages. Statistical significance was considered at a p-value < 0.05.

RESULTS

Clinical and phenotypic findings

A total of 114 patients with otomycosis were included in this study. Females were more frequently affected (54.4%) than males (45.6%). The highest prevalence was observed in the (31-40)years age group (20.2%), followed by the (41-50) years (18.4%) and (51-60) years (16.7 %) age groups. Pruritus was the most common clinical manifestation (38.6%), followed by otalgia (28.9%) and otorrhea (20.2%), whereas ear fullness (8.8%) and tinnitus (3.5%) were less frequently reported.

Effectiveness of antiseptic agents against *Candida* species isolates

Among the evaluated agents, 7.5% povidone-iodine demonstrated the greatest antifungal activity, inhibiting 80% of the *Candida* isolates. Hydrogen peroxide (3%) also exhibited considerable inhibitory activity, affecting 70% of the isolates. In contrast, 70% isopropyl alcohol and 2% acetic acid inhibited 50% and 54% of the isolates, respectively. Clotrimazole (1%), included as the reference antifungal agent, was effective against 38.6% of the isolates Table 2.

Table 2: Number and percentage of susceptible *Candida* species isolates exhibiting inhibition zones against antiseptic agents

<i>Candida</i> species	Total isolates	povidone-iodine	Acetic acid	Hydrogen peroxide	Isopropyl alcohol	Clotrimazole
<i>C. albicans</i>	42	35 (83.3%)	23 (54.8%)	31 (73.8%)	22 (52.4%)	14 (33.3%)
<i>C. parapsilosis</i>	30	27 (90.0%)	12 (40.0%)	21 (70.0%)	14 (46.7%)	13 (43.3%)
<i>C. glabrata</i>	16	15 (93.8%)	7 (43.8%)	11 (68.8%)	9 (56.3%)	5 (31.3%)
<i>C. tropicalis</i>	14	12 (85.7%)	8 (57.1%)	9 (64.3%)	6 (42.9%)	7 (50.0%)
<i>C. krusei</i>	12	9 (75.0%)	4 (33.3%)	8 (66.7%)	6 (50.0%)	5 (41.7%)
Total	114	98 (86.0%)	54 (47.4%)	80 (70.2%)	57 (50.0%)	44 (38.6%)

C. albicans: $\chi^2 = 10.80$, df = 4, P = 0.029

C. parapsilosis: $\chi^2 = 9.49$, df = 4, P = 0.050

C. glabrata: $\chi^2 = 6.30$, df = 4, P = 0.178 (NS)

C. tropicalis: $\chi^2 = 2.52$, df = 4, P = 0.640 (NS)

C. krusei: $\chi^2 = 2.69$, df = 4, P = 0.611 (NS)

Total isolates: $\chi^2 = 28.94$, df = 4, P < 0.001

Antifungal susceptibility testing

Antifungal susceptibility testing revealed variable susceptibility patterns among the *Candida* isolates, with differences in the rates of susceptibility, intermediate response, and resistance to the tested antifungal agents Table 3.

Table 3: Antifungal susceptibility patterns of *Candida* species by the VITEK® 2 System

<i>Candida</i> species	Sensitive (S)	Intermediate / SDD	Resistant (R)
Fluconazole			
<i>C. albicans</i>	17 (40.5%)	4 (9.5%)	21 (50.0%)

<i>C. parapsilosis</i>	14 (46.7%)	5 (16.7%)	11 (36.7%)
<i>C. glabrata</i>	7 (43.8%)	2 (12.5%)	7 (43.8%)
<i>C. tropicalis</i>	8 (57.1%)	1 (7.1%)	5 (35.7%)
<i>C. krusei</i>	7 (58.3%)	1 (8.3%)	4 (33.3%)
Total	53 (46.5%)	13 (11.4%)	48 (42.1%)
Flucytosine			
<i>C. albicans</i>	22 (52.4%)	4 (9.5%)	16 (38.1%)
<i>C. parapsilosis</i>	19 (63.3%)	2 (6.7%)	9 (30.0%)
<i>C. glabrata</i>	10 (62.5%)	1 (6.3%)	5 (31.3%)
<i>C. tropicalis</i>	9 (64.3%)	1 (7.1%)	4 (28.6%)
<i>C. krusei</i>	8 (66.7%)	1 (8.3%)	3 (25.0%)
Total	68 (59.6%)	9 (7.9%)	37 (32.5%)
Voriconazole			
<i>C. albicans</i>	33 (78.5%)	2 (4.7%)	7 (16.6%)
<i>C. parapsilosis</i>	25 (83.3%)	1 (3.3%)	4 (13.3%)
<i>C. glabrata</i>	12 (75.0%)	1 (6.2%)	3 (18.7%)
<i>C. tropicalis</i>	11 (78.5%)	1 (7.1%)	2 (14.2%)
<i>C. krusei</i>	8 (66.6%)	1 (8.3%)	3 (25.0%)
Total	89 (78.0%)	6 (5.2%)	19 (16.6%)
Caspofungin (Echinocandin)			
<i>C. albicans</i>	26 (61.9%)	7 (16.7%)	9 (21.4%)
<i>C. parapsilosis</i>	24 (80.0%)	2 (6.7%)	4 (13.3%)
<i>C. glabrata</i>	11 (68.8%)	2 (12.5%)	3 (18.8%)
<i>C. tropicalis</i>	10 (71.4%)	2 (14.3%)	2 (14.3%)
<i>C. krusei</i>	9 (75.0%)	1 (8.3%)	2 (16.7%)
Total	80 (70.2%)	14 (12.3%)	20 (17.5%)
Amphotericin B			
<i>C. albicans</i>	38 (90.5%)	1 (2.4%)	3 (7.1%)
<i>C. parapsilosis</i>	26 (86.7%)	1 (3.3%)	3 (10.0%)
<i>C. glabrata</i>	14 (87.5%)	1 (6.3%)	1 (6.3%)
<i>C. tropicalis</i>	11 (78.6%)	1 (7.1%)	2 (14.3%)
<i>C. krusei</i>	10 (83.3%)	1 (8.3%)	1 (8.3%)
Total	99 (86.8%)	5 (4.4%)	10 (8.8%)

*Antifungal susceptible (S), susceptible dose-dependent (SDD), and resistant (R).

Fluconazole: (S) $\leq 2 \mu\text{g/mL}$, (I) $4 \mu\text{g/mL}$, and (R) $\geq 8 \mu\text{g/mL}$

Voriconazole: (S) $\leq 0.12 \mu\text{g/mL}$, (I) $0.25-0.5 \mu\text{g/mL}$, and (R) $\geq 1 \mu\text{g/mL}$

Amphotericin B: susceptible (S) $\leq 1 \mu\text{g/mL}$, (R) $\geq 2 \mu\text{g/mL}$

Flucytosine: (S) $\leq 4 \mu\text{g/mL}$, (I) $8-16 \mu\text{g/mL}$, (R) $\geq 32 \mu\text{g/mL}$

Caspofungin: (S) $\leq 0.25 \mu\text{g/mL}$, (I) $= 0.5 \mu\text{g/mL}$, (R) $\geq 1 \mu\text{g/mL}$

Molecular Identification

Molecular analysis based on sequencing of the ITS1-5.8S-ITS2 region revealed variations in the distribution of *Candida* species among the clinical isolates, as illustrated in Figure 1.

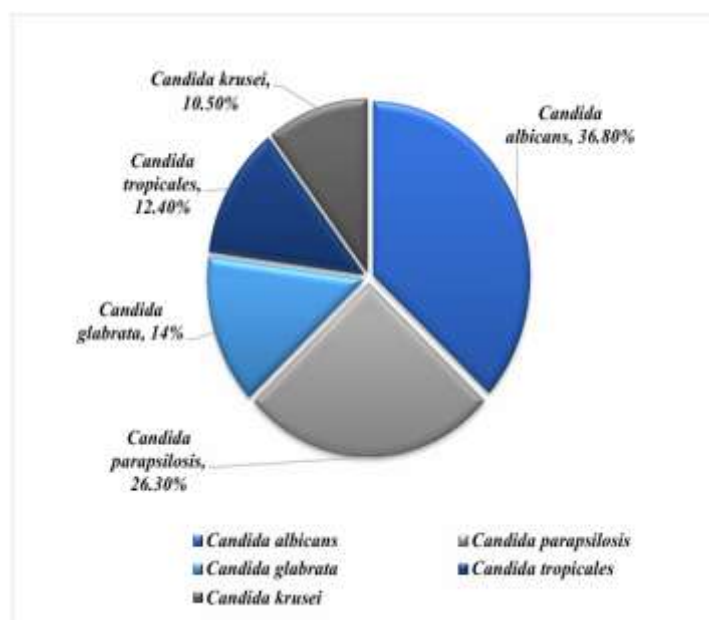


Figure 1: Percentage of *Candida* species identified by ITS1-5.8S-ITS2 rDNA sequencing among the yeast isolates.

Detection of the *CDR1* and *ERG11* gene

Conventional PCR detected the *ERG11* and *CDR1* resistance genes among the *Candida* isolates. The distribution of resistant isolates for both genes is presented in Table 4.

Table 4: Detection of *ERG11* and *CDR1* resistance genes among *Candida* species

<i>Candida</i> species	No. of isolates	<i>ERG11</i> resistant (%)	<i>CDR1</i> resistant (%)
<i>C. albicans</i>	42	36 (58.7%)	30 (71.4%)
<i>C. parapsilosis</i>	30	22 (73.3%)	18 (60%)
<i>C. glabrata</i>	16	10 (62.5%)	12 (75%)
<i>C. tropicalis</i>	14	11 (78.6%)	9 (64.3%)
<i>C. krusei</i>	12	7 (58.3%)	8 (66.7%)
Total	114	86 (75.4%)	77 (67.5%)

Comparison between antifungal susceptibility patterns and resistance-associated genes among *Candida* species

The comparison included resistance to fluconazole, voriconazole, caspofungin, and amphotericin B, together with the molecular detection of the *ERG11* and *CDR1* genes, as presented in Table 5.

Figure 5: Comparison between The VITEK® 2 system and resistance-associated genes (*ERG11* and *CDR1*) among *Candida* species.

<i>Candida</i> species	Fluconazole Resistance (%)	Voriconazole Resistance (%)	Caspofungin Resistance (%)	Amphotericin Resistance (%)	<i>ERG11</i> Resistance (%)	<i>CDR1</i> Resistance (%)
<i>C. albicans</i>	50.0	16.6	21.4	7.1	85.7	71.4
<i>C. parapsilosis</i>	36.7	13.3	13.3	10.0	73.3	60.0
<i>C. glabrata</i>	43.8	18.7	18.8	6.3	62.5	75.0
<i>C. tropicalis</i>	43.8	14.2	14.3	14.3	78.6	64.3
<i>C. krusei</i>	33.3	25.0	16.7	8.3	58.3	66.7
Total	42.1	16.6	17.5	8.8	75.4	67.5

DISCUSSION

Otomycosis is a frequently encountered condition in otolaryngology (ENT) practice. Its management remains challenging for both patients and otolaryngologists due to its recurrent nature and prolonged treatment (Al Husaini *et al.*, 2016). The present study demonstrated that otomycosis affected both females and males with a slightly higher prevalence among females. The higher prevalence among females may be related to prolonged use of head coverings, which can increase moisture and temperature within the external auditory canal, thereby creating a favorable environment for yeast growth (Kumar, 2005; Fasunla *et al.*, 2008). The findings of the present study are consistent with those reported by (Barati *et al.*, 2011) However, the present results are inconsistent with several other studies, including those by (Al-Abbasi *et al.*, 2011; Younes *et al.*, 2021) all of which reported a higher prevalence among males. In addition, the distribution of *Candida* species varied across different age groups, possibly reflecting differences in occupational exposure, environmental conditions, and personal ear care practices. Furthermore, the incidence of otomycosis remains higher in warm and humid climates, particularly during the hot season, where environmental conditions promote yeast proliferation (Naeimi *et al.*, 2024).

Morphological characteristics are still playing an important role in yeast taxonomy and the preliminary identification of *Candida* species (Mirhendi *et al.*, 2006). In the present study, the phenotypic characterization of the 114 yeast isolates represented the initial step in the identification process. This identification was based on the evaluation of macroscopic characteristics and colony morphology on primary isolation media. In the present study, 1% Clotrimazole, a specific azole antifungal agent, showed the largest mean inhibition zone diameter, indicating strong antifungal activity against the isolated *Candida* species. Despite this apparent activity, only 43.8% of the isolates were found to be susceptible, suggesting the emergence of resistance among certain *Candida* species. The findings of the current study are in agreement with those reported by Philip *et al.* (2013), who evaluated the therapeutic efficacy of clotrimazole in the treatment of Otomycosis and demonstrated its clinical effectiveness. However, increasing antifungal resistance has become an important concern worldwide. Gonçalves *et al.* (2016) reported an azole resistance prevalence of 3.2%, which was associated with worse clinical outcomes among patients. Similarly, Van der Linden *et al.* (2011), reported a significant increase in resistance to azole antifungal agents, which was attributed to the prolonged and widespread use of azoles. Automated systems, such as the VITEK® 2 system, have proven their ability to provide rapid and accurate data on the susceptibility of *Candida* isolates, facilitating early treatment decisions (Alrubayae *et al.*, 2020). The present examination revealed that all *Candida* species were susceptible to varying degrees of the antifungals used. Studies by AL-Maliki *et al.* (2011) indicated that yeasts are less susceptible to antifungals because the indiscriminate and frequent use of these drugs leads to the appearance of resistant *Candida* species.

The non-specific antifungal agents evaluated in the present study demonstrated variable inhibitory activities against *Candida* species isolates. Among the tested agents, 7.5% Povidone-iodine exhibited the highest overall antifungal activity. This high efficacy may be attributed to its broad-spectrum antifungal mechanism. Povidone-iodine gradually releases free iodine, which rapidly penetrates the yeast cell wall and membrane, causing oxidation and iodination of essential cellular proteins, enzymes, and nucleic acids, and ultimately result in yeast cell death. Unlike conventional antifungal agents that target a single metabolic pathway, povidone-iodine acts on multiple intracellular targets simultaneously, reducing the likelihood of resistance development (Meehan, 2025; Hadamu *et al.*, 2025). The present findings are consistent with previous studies by Dai *et al.* (2009), Mofatteh *et al.* (2018), and Aziz *et al.* (2020), which demonstrated that povidone-iodine is an effective therapeutic agent for otomycosis, providing rapid clinical improvement and, in some cases, superior efficacy compared with clotrimazole. Acetic acid exerts its antifungal effect by lowering the pH of the external auditory canal, thereby creating an unfavorable environment for fungal growth and interfering with essential cellular metabolic processes. In contrast, isopropyl alcohol promotes drying and cleansing of the ear canal, reducing moisture that supports yeast proliferation (Yassin *et al.*, 2023; Pereira *et al.*, 2024; Moghadam *et al.*, 2010). The antifungal activity of 3% hydrogen peroxide is mainly attributed to its oxidizing properties, as it generates free radicals that react with lipids, proteins, and nucleic acids, leading to nonspecific damage of essential cellular components (Maillard *et al.*, 2024). Besides its antifungal activity, hydrogen peroxide also exhibits cleansing and cerumenolytic properties, facilitating the removal of debris and improving ear canal hygiene. These additional effects may enhance its therapeutic value in the management of otomycosis and other otic infections (Demirtas *et al.*, 2025).

The high prevalence of resistance among *Candida* species was associated with both *ERG11* and *CDR1* genes, although the frequency of resistance varied between species. Overall, *ERG11*-associated resistance was higher than *CDR1*-associated resistance. *Candida albicans* demonstrated the highest rate of *ERG11* resistance, whereas *Candida glabrata* showed the highest rate of *CDR1* resistance. These variations indicate that the mechanisms underlying antifungal resistance may differ across *Candida* species. The findings also suggest that alterations or overexpression of the *ERG11* gene may play a major role in the development of azole resistance. At the same time, *CDR1*-mediated efflux pump activity may contribute differently among *Candida* species. Furthermore, no statistically significant association was observed between *Candida* species and the distribution of resistance genes ($P > 0.05$). The relationship between the *ERG11* and *CDR1* genes in resistance is based on two complementary mechanisms. *ERG11* encodes the target enzyme of azole drugs, lanosterol 14- α -demethylase, where alterations or overexpression of this gene can disrupt the normal ergosterol pathway, leading to reduced susceptibility and increased resistance to azole antifungal agents (Mohammed *et al.*, 2025). In contrast, the (ABC) transporter efflux pump contributes to resistance by actively reducing intracellular antifungal drug accumulation, thereby decreasing drug efficacy and promoting the survival of yeast cells under antifungal pressure (Kubica *et al.*, 2025). Although these genes act through distinct pathways, their co-expression can significantly enhance antifungal resistance in *Candida* species by simultaneously reducing drug-target susceptibility and intracellular drug concentration.

Conclusion: This study demonstrated that *Candida* species are important etiological agents of otomycosis, with *Candida albicans* being the predominant species. A high prevalence of the antifungal resistance genes *ERG11* and *CDR1* was detected among the isolates, which was associated with reduced susceptibility to antifungal agents. Furthermore, selected topical antiseptic agents exhibited promising inhibitory effects against resistant *Candida* isolates, suggesting their potential use as alternative or adjunctive therapeutic options.

Ethical Approval: The study was conducted in accordance with the ethical approval granted by the Basra Health Directorate.

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