REVIEW

Small molecules for combating multidrug-resistant superbug Candida auris infections

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Abstract Candida auris is emerging as a major global threat to human health. C. auris infections are associated with high mortality due to intrinsic multi-drug resistance. Currently, therapeutic options for the treatment of C. auris infections are rather limited. We aim to provide a comprehensive review of current strategies, drug candidates, and lead compounds in the discovery and development of novel therapeutic agents against C. auris. The drug resistance profiles and mechanisms are briefly summarized. The structures and activities of clinical candidates, drug combinations, antifungal chemosensitizers, repurposed drugs, new targets, and new types of compounds will be illustrated in detail, and perspectives for guiding future research will be provided. We hope that this review will be helpful to prompting the drug development process to combat this fungal pathogen.

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1. Introduction

There are approximately 200 species in the genus Candida, and these are the main causal agents (e.g., Candida albicans) of worldwide invasive fungal infections (IFIs)†. Candida auris was first isolated in 2009 and since then has rapidly spread globally‡. C. auris is characterized by a high level of multi-drug resistance and has emerged as a major and urgent healthcare threat§. C. auris infections have been reported in more than 45 countries and have caused serious hospital outbreaks, with crude mortality rates as high as 72%∥. C. auris can be transmitted by direct or indirect contact¶. Persistent skin colonization, environmental adaptation and contamination, and nosocomial transmission have contributed to the global pandemic of C. auris¶.

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C. auris is a member of the Candida haemulonii clade and is distantly related to common fungal pathogens such as C. albicans and Candida glabrata. To date, the origins of C. auris are still largely unknown. Based on genetic studies using whole-genome sequencing (WGS), C. auris strains are classified into four major geographic clades, namely, clade I (the South Asian clade), clade II (the East Asian clade), clade III (the South African clade), and clade IV (the South American clade). Recently, a potential fifth clade of C. auris was isolated from Iran. Considerable differences in genetics and phenotypes have been observed among C. auris strains from different clades.

The characteristics and mechanisms of C. auris infections slightly differ from those of other Candida species. The body sites of C. auris colonization mainly include skins, mucosa and gastrointestinal tract, overlapping with those of other Candida species, such as C. albicans, C. glabrata, Candida parapsilosis, and Candida tropicalis. However, C. auris exhibits a stronger capacity for skin colonization than other Candida species. The number of colonized patients was 2–3 folds more than that of infected patients, which causes clonal inter- and intra-hospital transmission and healthcare-associated infections. The colonization of C. auris on skin or other sites may not cause infections, which could possibly lead to the contamination of the nosocomial and healthcare environment and pose a risk on immunocompromised individuals. Thus, guidelines on the prevention of spread of C. auris are much stricter than those of other Candida species. Routine screening on colonization sites of patients and medical staff and improved environmental decontamination may interrupt healthcare-associated transmission. Chlorine-containing disinfectants and 2% chlorhexidine are currently used in clinical practice for environmental decontamination and skin decolonization, respectively. Moreover, recent studies showed that C. auris may behave differently as other Candida species to induce innate immune responses, facilitating its colonizations and infections in hosts.

Unfortunately, therapeutic options for the treatment of C. auris infections are rather limited. Only three major classes of antifungal agents (Fig. 1), namely, azoles (1–5), polyenes (6), and echinocandins (7–9), are clinically available for the treatment of IFIs. Additionally, the nucleoside analogue 5-flucytosine (10) is generally used in adjunctive therapy. However, most C. auris strains were reported to be resistant to fluconazole (1), and multi-drug resistance has also observed against two, three, or even four classes of antifungal agents. C. auris is the only Candida species in which several isolates have been identified to be resistant to all four classes of antifungal drugs. The order of drug resistance is fluconazole > amphotericin B (6) > echinocandins. Thus, echinocandins (e.g., caspofungin, 7) are commonly recommended as the first-line therapy for the treatment of C. auris infections. Even so, several cases of deaths were reported for patients after the administration of echinocandins. In addition to intrinsic resistance, rapid development of multidrug resistance has also been documented during antifungal treatments. Thus, there is an urgent need to develop effective therapeutics to treat life-threatening and multidrug-resistant C. auris infections.

The biology, pathogenicity, epidemiology, resistance mechanisms and active compounds of C. auris have been reviewed previously. Continuing our efforts in the discovery of novel antifungal agents against resistant fungal pathogens, this review focuses on the small molecules and potential drug targets with which to tackle C. auris infections. After a brief introduction of resistance profiles and mechanisms, the activity of clinical candidates and drug combinations is discussed. Then, we provide a detailed illustration of drug discovery strategies and active lead compounds for combating C. auris infections, focusing on antifungal chemosensitizers, drug repurposing, new targets, and new chemotypes. Finally, perspectives for future research on drug development for this superbug fungal pathogen are provided.

2. Susceptibility of C. auris to antifungal agents

Several studies have investigated the susceptibility of C. auris to antifungal agents using different sets of isolates. On the basis of a susceptibility test against 350 isolates collected in India, 90% of the isolates were resistant to fluconazole (MIC: 32–64 μg/mL); 8% were resistant to amphotericin B (MIC: 2 μg/mL), and 2% were resistant to echinocandins (MIC: 8 μg/mL). In another test of 296 C. auris isolates, a similar resistance trend was observed in which 80% of the strains were resistant to fluconazole, 23% to amphotericin B, and 7% to micafungin. Notably, 24% of the tested strains were resistant to at least two classes of antifungal agents, and 1% were resistant to all three of the classes. The resistance profiles appeared to be clade-specific. For example, C. auris isolates in clade III were reported to be more resistant to fluconazole and voriconazole than isolates in clade I. Newer azoles such as posaconazole (5, MIC range: 0.06–1 μg/mL) and isavuconazole (3, MIC range: 0.008–4 μg/mL) showed improved in vitro activity against C. auris. Elevated MIC values were observed for the new azoles (e.g., voriconazole) compared with those against other Candida species.

3. Resistance mechanisms of C. auris

The antifungal drug resistance mechanisms of C. auris are similar to those observed in other Candida species, including overexpression or mutation of the drug target, overexpression of efflux pumps, reductions of drug intake, and biofilm formation. Azole antifungal agents act by inhibiting lanosterol 14α-demethylase (CYP51, encoded by the ERG11 gene), a key enzyme in the biosynthesis of ergosterol of the fungal cell membrane. In C. auris strains resistant to azoles, no significant overexpression of the ERG11 gene was observed, and substitution mutations in CYP51 were generally clade-dependent: F126T (clade III), Y132F (clade IV), and Y132F or K143R (clade I). Higher expression of multidrug efflux pumps was also involved in decreased susceptibility of C. auris to azoles. The ATP-binding cassette (ABC) family and the major facilitator superfamily (MFS) are two major transporters associated with antifungal resistance that are conserved in C. auris. Increased expression of the CDRI gene of the ABC transporter and the MDRI gene of the MFS transporter contributed to the azole resistance of C. auris.

Amphotericin B exerts fungicidal activity by binding to ergosterol in fungal cell membranes and thereby altering the membrane permeability, resulting in the leakage of vital cytoplasmic components. Overexpression of genes involved in ergosterol biosynthesis,
such as ERG1, ERG2, ERG6, and ERG13, was reported to be related to amphotericin B resistance in *C. auris* strains. Although significant mutation for amphotericin B resistance is rare, a point mutation in transcription factor FLO8 has been observed in a resistant *C. auris* isolate.

Echinocandins act on the fungal cell wall via inhibition of 1,3-β-glucan synthase (encoded by the *FKS1* gene). In *C. auris*, *FKS1* substitution mutations S639F, S639P, S639Y, and S652Y were responsible for echinocandins resistance. The compound 5-flucytosine inhibits fungal DNA and RNA synthesis and is activated in fungal cells by Fur1. In *C. auris*, a substitution mutation F211I in the *FUR1* gene was detected in an isolate resistant to 5-flucytosine. The increased expression of ABC and MFS transporters also contributes to the formation of biofilms that are highly resistant to antifungal agents. Most antifungal agents, such as fluconazole, voriconazole, and amphotericin B, showed higher MIC values against *C. auris* biofilms than against planktonic cells. Although planktonic cells are susceptible to echinocandins, these compounds are ineffective against biofilms. Similar to other *Candida* species, *C. auris* is able to form biofilms that are largely composed of mannans and glucans. *C. auris* formed significantly less biofilm than *C. albicans* with a limited amount of extracellular matrix. *C. auris* seems to be unable to form true hyphae, and its biofilms consist largely of yeast cells. The phenotypic, biochemical, and functional features of *C. auris* are discussed in the paper.
biofilms seem to be clade- or strain-specific. Differences in the extent of biofilm formation were observed among various C. auris isolates. Compared with C. albicans, C. auris formed more consistent biofilms in colonization models, suggesting higher virulence and resistance.

4. Investigational antifungal agents for the treatment of C. auris infections

Currently, several new antifungals have entered into the clinical research, including VT-1598 (11), PC945 (12), rezafungin (13), ibrexafungerp (14), SCY-247 (15), fosmanogepix (16), manogepix (17) and T-2307 (18), which have demonstrated promising results against C. auris (Fig. 3). Herein the in vitro and in vivo anti-C. auris activities of these investigated antifungal agents are discussed. The antifungal assays and expression levels of activity are summarized in Table 1.

4.1. New CYP51 inhibitors: Triazole antifungal agents VT-1598 and PC945

VT-1598, a tetrazole-based fungal CYP51 inhibitor, has entered the clinical evaluation. Currently, phase I clinical trial of VT-1598 has been completed and no more clinical trial is ongoing. Compared with traditional triazole antifungal agents, VT-1598 showed better selectivity between fungal CYP51 and mammalian cytochrome P450 enzymes, resulting in reduced drug–drug interactions. VT-1598 demonstrated potent in vitro activity against a collection of 100 C. auris isolates (MIC range: 0.03 μg/mL; MIC50 = 0.25 μg/mL; MIC90 = 1 μg/mL). VT-1598 also showed dose-dependent in vivo efficacy in a neutropenic murine model of C. auris infections. At the doses of 15 and 50 mg/kg (once daily), oral VT-1598 treatment achieved significant improvement in survival, with median survival of 15 days and >21 days, respectively. Moreover, VT-1598 also significantly reduced kidney and brain fungal burdens, suggesting that VT-1598 deserved further evaluation as a potential option for treating C. auris infections.

PC945 is a novel triazole antifungal derivative designed for inhaled administration of Aspergillus fumigatus infections. PC945 also showed excellent antifungal activity against a collection of 50 C. auris clinical isolates, with GM MIC, MIC50 and MIC90 values of 0.058, 0.063, and 0.25 μg/mL, respectively. PC945 also completely inhibited C. auris growth, with GM MIC and MIC90 values of 0.16 and 0.5 μg/mL, respectively. Notably, PC945 showed better anti-C. auris activity than fluconazole, voriconazole, and posaconazole.

4.2. New glucan synthase inhibitors: Rezafungin and ibrexafungerp

Rezafungin (CD101), an optimized echinocandin derivative, is currently under clinical development. Compared with marketed echinocandin-like antifungal agents (e.g., caspofungin and micafungin), rezafungin possessed a better safety profile and improved pharmacokinetic properties such as an longer half time ($t_{1/2} > 130$ h) and higher plasma drug exposure, enabling once-weekly intravenous therapy. Several studies have confirmed that rezafungin had excellent in vitro and in vivo activities against C. auris infections. In a susceptibility assay of a collection of 100 C. auris isolates, the MIC values of rezafungin ranged from 0.03 to 8 μg/mL. The MIC50 and MIC90 values were 0.125 and 0.5 μg/mL, respectively. Similar in vitro activity was observed in a test of rezafungin against 122 Indian C. auris isolates (MIC range: 0.016–16 μg/mL; MIC50 = 0.25 μg/mL; MIC90 = 1 μg/mL). In a mouse model of disseminated C. auris infections, rezafungin (20 mg/kg ip) showed potent in vivo efficacy and effectively reduced the fungal burden. In particular, rezafungin showed superior activity compared to amphotericin B and micafungin, even with less frequent dosing. The PK/PD advantage of rezafungin was further validated in a C. auris neutropenic mouse model. The PK/PD index of rezafungin suggested that the clinically evaluated dose (400 mg, iv, once a week) may be a useful option to treat patients infected with C. auris infections, although further clinical trials are warranted.

Ibrexafungerp (SCY-078) is an orally active inhibitor of glucan synthase that exhibited in vitro and in vivo inhibitory activity against
Candida species, including echinocandin-resistant isolates. Ibrexafungerp differs from echinocandin-like glucan synthase inhibitors in that it can be administered both orally and intravenously, and it is active against the most common mutations of the target gene FKS. A susceptibility assay indicated that the MIC values of ibrexafungerp ranged from 0.0625 to 2 μg/mL against a collection of 100 C. auris isolates, with MIC50 and MIC90 values of 0.5 and 1 μg/mL, respectively. Furthermore, ibrexafungerp showed similar MIC values against C. auris isolates resistant to echinocandin antifungal agents. Larkin et al. and Arendrup et al. reported similar MIC results for ibrexafungerp. Ibrexafungerp was able to completely inhibit the growth of C. auris, with an MIC90 value of 1 μg/mL. Moreover, ibrexafungerp interrupted cell division of C. auris and inhibited biofilm formation (0.5–4 μg/mL) by reducing metabolic activity and biofilm thickness. In a neutropenic murine model of C. auris infections, oral treatment with ibrexafungerp (20, 30, and 40 mg/kg, twice daily) resulted in dose-dependent improvements of survival and reductions in fungal burden, while caspofungin showed similar potency, and fluconazole was ineffective. In an in vivo guinea pig cutaneous model of C. auris infections, oral dosing with ibrexafungerp (10 mg/kg) was effective in controlling skin infections and significantly reduced the fungal burden and the severity of lesions. Ibrexafungerp is currently in phase II open-label clinical trials to evaluate efficacy and safety in patients infected with C. auris (identifier: NCT03363841). In an emergency-use phase III clinical trial, ibrexafungerp therapy successfully cured two patients without drug-related adverse events, highlighting its potential for further clinical evaluation.

SCY-247 is an analogue of ibrexafungerp that showed broad-spectrum antifungal activity and an excellent safety profile, and it is suitable for both intravenous and oral administration. Ghanoun’s group compared in vitro anti-C. auris activity between SCY-247 and ibrexafungerp. In a panel of 44 C. auris isolates, SCY-247 (MIC range: 0.06–1 μg/mL, MIC90 = 0.5 μg/mL, MIC90 = 0.5 μg/mL) showed similar MIC values to ibrexafungerp (MIC range: 0.06–2 μg/mL, MIC50 = 0.5 μg/mL, MIC90 = 0.5 μg/mL). The fungicidal activity of SCY-247 (MFC90 = 8 μg/mL) was slightly better than that of ibrexafungerp (MFC90 = 8 μg/mL). The in vivo potency of SCY-247 against C. auris infections has not been reported. However, SCY-247 (40 mg/kg) exhibited a 100% survival rate in a murine model of disseminated infections of C. albicans, suggesting that the efficacy of SCY-247 to treat C. auris deserves further evaluation.

4.3. Fungal cell wall Gwt1 inhibitor: Manogepix

Manogepix (APX001A) is an inhibitor of fungal Gwt1 (glycosylphosphatidylinositol-anchored wall transfer protein 1) that showed broad-spectrum antifungal activity. Fosmanogepix (APX001), the prodrug of manogepix, is currently being evaluated...
in clinical trials to treat various fungal infections\textsuperscript{35}. The efficacy of APX001A to treat \textit{C. auris} infections has been well characterized\textsuperscript{35,36–39}. Hager et al.\textsuperscript{38} determined the inhibitory activity of APX001A against 16 \textit{C. auris} clinical strains. APX001A had MIC values in the range of 0.002–0.063 \textmu g/mL (MIC\textsubscript{90} = 0.004 \textmu g/mL; MIC\textsubscript{90} = 0.031 \textmu g/mL), values that demonstrated greater potency than 10 tested antifungal agents\textsuperscript{38}. The excellent activity of APX001A was further confirmed in a large collection of \textit{C. auris} containing 100 geographically distinct isolates\textsuperscript{37}. The MIC values ranged from <0.005 to 0.015 \textmu g/mL, and the MIC\textsubscript{50} and MIC\textsubscript{90} values were 0.002 and 0.008 \textmu g/mL, respectively\textsuperscript{37}. Zhu et al.\textsuperscript{39} evaluated the in vitro inhibitory activity of APX001A against 200 New York \textit{C. auris} isolates. APX001A demonstrated lower MIC values (MIC range: 0.004–0.06 \textmu g/mL; MIC\textsubscript{90} = 0.03 \textmu g/mL; MIC\textsubscript{90} = 0.03 \textmu g/mL) than 10 clinical antifungal agents\textsuperscript{38}. APX001 also showed potent in vivo efficacy to treat \textit{C. auris} infections and was more effective than caspofungin and amidulafungin\textsuperscript{9}. In a murine model of disseminated \textit{C. auris} infections, APX001 effectively prolonged the survival time of the treated mice (100% survival, 78 mg/kg TID) and significantly reduced the fungal burden of kidney, lung, and brain\textsuperscript{37}. In a pharmacokinetics (PK) and pharmacodynamics (PD) study of APX001, the ED\textsubscript{50} (50% of the maximum effect) to treat \textit{C. auris} infections was 77 mg/kg\textsuperscript{61}. Even delayed therapy with fosmanogepix showed good potency, significantly reducing the kidney fungal burden at the dose of 260 mg/kg (BID)\textsuperscript{89}. These in vitro and in vivo data supported further clinical evaluation of fosmanogepix as an anti-\textit{C. auris} agent. An open-label clinical study of APX001 for the treatment of patients with candidemia caused by \textit{C. auris} was started in 2019 (identifier: NC-T04148287). Although the trial was terminated due to the impact of COVID-19, the objectives of the study were successfully met. However, the clinical data have not been disclosed to date.

5.1. Combinations of antifungal agents

Among the combinations of antifungal agents, voriconazole with micafungin, fluconazole with amphotericin B, and fluconazole with micafungin have shown synergistic effects regarding inhibition of the growth of \textit{C. auris} (Table 2). Fakhim et al.\textsuperscript{46} evaluated the synergism between echinocandins and azoles against 10 multidrug-resistant \textit{C. auris} clinical isolates. Synergistic effects were observed for the combination of micafungin and voriconazole against all of the tested isolates (FICI range: 0.15–0.5)\textsuperscript{65}. Another study systematically evaluated 864 antifungal drug combinations against 15 \textit{C. auris} isolates\textsuperscript{30}. Fluconazole (1.0 \textmu g/mL) was able to potentiate the activity of other antifungal agents, including azoles, echinocandins, and amphotericin B\textsuperscript{12}. However, in another study by Bidaud et al.\textsuperscript{13}, indifferent interactions between fluconazole and other antifungal agents were observed. Thus, the therapeutic effects of drug combinations remain to be further evaluated by in vivo studies.

More recently, the synergistic activity of isavuconazole and voriconazole in combination with anidulafungin was evaluated

### Table 1: Assays and expression of the activity for the research and development of novel antifungal agents against \textit{C. auris.}

| Activity                        | Assay                                         | Ref. |
|--------------------------------|-----------------------------------------------|------|
| **In vitro susceptibility**    | Clinical and Laboratory Standards Institute (CLSI) | 55   |
| **Synergistic activity**       | European Committee on Antimicrobial Susceptibility Testing (EUCAST) | 55   |
| **Biofilm inhibition**         | Fractional inhibitory concentration index (FICI) | 56   |
| **In vivo potency**            | Bliss independence model                       | 57   |
| **Candida auris**              | Inhibition of biofilm formation: XTT reduction assay | 50   |
|                                | Caenorhabditis elegans infections model (preliminary screen) | 58   |
|                                | Gallera mellonella infections model (preliminary screen) | 59   |
|                                | C. auris candidemia mouse model: survival curve, reduction of fungal burden (log\textsubscript{10} CFU/g) and ED\textsubscript{50} | 60   |
|                                | Pharmacokinetic/pharmacodynamic (PK/PD) index | 61   |
|                                | Guinea pig cutaneous infections model         | 62   |

ia: Minimum inhibitory concentration (MIC); MIC\textsubscript{50}, MIC\textsubscript{90}, MIC\textsubscript{90}: the lowest concentration inhibiting fungal growth by 50%, 80%, and 90%, respectively; geometric mean (GM) MIC; mode MIC; MFC: minimum fungicidal concentration.

b: FICI < 0.5: synergism; 0.5 < FICI < 4: FICI > 4: antagonism.

c: Sessile MIC (SMIC\textsubscript{50}): the concentration inhibiting 50% of biofilm formation; XTT: 2,3-bis(2-methoxy-4-nitro-5-sulfophenyl)-2H-tetrazolium-5-carboxanilide.

d: ED\textsubscript{50}: doses required to produce 50% of the maximal effect; PK/PD index: AUC (concentration–time curve)/MIC value.
against a collection of 36 *C. auris* isolates. The isavuconazole–anidulafungin combination (active in 11/36 isolates) showed stronger synergistic effects than the voriconazole–anidulafungin combination (active in 5/36 isolates). Similar synergistic interactions between isavuconazole and echinocandin-like antifungal agents were also observed in an assay with six clinical *C. auris* isolates. In addition to the synergistic effects against planktonic isolates, isavuconazole was also able to potentiate the activity of caspofungin in inhibiting the biofilm formation of *C. auris* (FICI range: 0.023–0.5, 12/14 sessile isolates). In a mouse infections model of *C. auris*, single uses of caspofungin (1 mg/kg, daily) or isavuconazole (20 mg/kg, daily) were statistically ineffective. When used in combination, the kidney fungal burden was significantly decreased by more than three log volumes. Thus, isavuconazole had direct and synergistic activity against *C. auris*, providing a promising option for further evaluation.

In a time–kill curve assay against six *C. auris* isolates, monotherapy with echinocandins (anidulafungin and caspofungin) was ineffective, while high concentrations of amphotericin B (≥1 μg/mL) only showed fungistatic activity. When used in combination, higher fungal killing activity was observed. Lower doses of amphotericin B (0.5 mg/L) and anidulafungin or caspofungin (2 mg/L) achieved rapid synergism with potent fungicidal activity.
Table 2. Synergistic combinations of antifungal drugs against C. auris.

| Drug                        | Antifungal drug | FICI  | Isolate | Ref. |
|-----------------------------|-----------------|-------|---------|------|
| **Antifungal drug**         |                 |       |         |      |
| Miconafungin                | Voriconazole    | 0.15−0.5 | 10/10  | 56   |
| Anidulafungin               | Isavuconazole   | 0.25−0.38 | 11/36  | 38   |
|                             | Voriconazole    | 0.25−0.38 | 5/36   | 38   |
|                              | **Amphotericin B** | Caspofungin | ND b | 6/6 | 100  |
|                              |                 | Anidulafungin | ND   | 6/6 | 100  |
| **Non-antifungal drug**     |                 |       |         |      |
| Sulfamethoxazole            | Fluconazole     | 0.156 | 1/1     | 101  |
|                             | Voriconazole    | 0.09−0.5 | 3/8   | 101  |
|                             | Itraconazole    | 0.186−0.373 | 3/4   | 101  |
|                             | Voriconazole    | 0.04−0.09 | 10/10 | 58   |
|                             | Itraconazole    | 0.19−0.31 | 6/10  | 58   |
|                             | Fluconazole     | 0.13−0.31 | 3/10  | 58   |
|                             | Itraconazole    | 0.14−0.31 | 8/10  | 102  |
|                             | Flucanazole     | 0.25−0.5 | 5/5   | 103  |
|                             | Itraconazole    | 0.14−0.27 | 5/5   | 104  |
| Chlorhexidine acetate       | Fluconazole     | <0.1875 | 2/2   | 105  |
| Miltefosine                 | Amphotericin B  | 0.5   | 3/12   | 106  |
| Colistin                    | Caspofungin     | 0.08−0.14 | 15/15 | 107  |
|                            | Isavuconazole   | 0.3125−0.5 | 15/15 | 107  |
| Chemosensitizer             | Azolithin       | Fluconazole | 0.25  | 1/1  | 108  |
|                            | Carvacrol       | Amphotericin B | 0.28−1.5 | 7/25 | 109  |
|                            | Farnesol        | Caspofungin | 0.156−0.5 | 3/4  | 110  |
|                            |                  | Miconafungin | 0.133−0.281 | 4/4  | 110  |
|                            |                  | Anidulafungin | 0.14−0.375 | 4/4 | 110  |
|                            |                  | Itraconazole | 0.188  | 1/3  | 111  |
|                            |                  | Voriconazole | 0.311  | 1/3  | 111  |

*Number of isolates: active isolates/tested isolates.
1ND: not determined.
2Synergistic effect against biofilm inhibition.

5.2. Combinations of antifungal agents and non-antifungal agents

Seleem’s group performed systemic screening of known drugs and identified several synergists that were able to potentiate the activity of antifungal agents against C. auris. Inspired by the effects of sulfon antibacterial drugs to reverseazole resistance against C. albicans, the sulfon drugs were also confirmed to possess synergistic activity withazole antifungals to inhibit the growth of C. auris isolates. Among them, sulfamethoxazole exhibited the best synergistic activity with voriconazole (FICI range: 0.09−0.5) and itraconazole (FICI range: 0.186−0.373). Sulfamethoxazole alone was inactive against C. auris (MIC ≥256 μg/mL). When sulfamethoxazole (128 μg/mL) was used in combination with voriconazole (0.5 μg/mL), the survival of the infected nematodes was prolonged by about 70% in an in vivo model using C. elegans. The underlying mechanisms of the synergism were possibly associated with interference with the expression of the target protein CYP51 and the fungal folate pathway. After screening 1547 compounds, Seleem’s group found that the antiviral agent lopinavir was a potent chemosensitizer that could potentiate the activity of fluconazole against resistant C. auris isolates. At a therapeutically acceptable concentration (10 μg/mL), lopinavir showed good synergistic effects with fluconazole, voriconazole, and itraconazole (Table 2). The strongest synergism was observed when lopinavir and itraconazole (FICI range: 0.04−0.09). The drug combination also showed good in vivo efficacy in a C. auris-infected model using C. elegans, improving the survival rate by 90% and reducing the fungal burden by 88.5%. The mechanism of the synergism was investigated by comparative transcriptomic analysis. The drug combination may act by interfering with the expression of several transporters that are related to glucose permeation and drug efflux.

The same group identified aprepitant, an antiemetic agent, as a potent synergist of itraconazole by assaying the azole chemosensitizing activity of a compound library containing about 1600 FDA-approved drugs. Aprepitant was able to reduce the MIC of itraconazole by up to eight-fold against C. auris (FICI range: 0.14−0.31). The drug combination was fungicidal and significantly inhibited biofilm formation (95%) and mature biofilms (52%). The combination of aprepitant and itraconazole also showed in vivo activity in a C. elegans infections model, significantly prolonging the survival rate by ~90% and reducing the fungal burden by ~92%. The mechanism of synergistic effects was associated with interfering with metal ion homeostasis and the ROS detoxification ability of C. auris.

In a screen of synergists against azole-resistant C. albicans, the pitavastatin (22)-fluconazole combination was identified to have broad-spectrum synergistic activity. In particular, pitavastatin displayed potent fluconazole chemosensitizing activity against 5 C. auris isolates (FICI range: 0.25−0.5). The combination of pitavastatin-fluconazole effectively inhibited the biofilm-forming abilities and reduced the CFU burden by 14%−92% in an in vivo C. elegans model with C. auris. The mechanism of synergism was associated with interference with the efflux machinery. Eldesouky et al. assayed nine stilbene compounds for their synergistic activity with azole drugs against azole-resistant fungal isolates. The ospemifene (23)-itraconazole combination displayed the most potent chemosensitizing activity against a variety of fungal pathogens, including C. auris (FICI range: 0.14−0.27). The drug combination reduced C. auris CFU burden by 96% in a C. elegans infections model. Ospemifene exerted synergistic activity by directly interfering with fungal efflux systems such as ABC and MFS transporters and facilitating the entry of azoles into fungal cells.

Chlorhexidine acetate is a broad-spectrum antibacterial agent. When used alone, its MIC values against C. auris isolates CBS12373 and CBS10913 were 8 and 2 μg/mL, respectively. When chlorhexidine acetate was used in combination with fluconazole, significant synergism was observed in the growth curve assay. In particular, the drug combination showed strong synergism against the biofilm formation of C. auris strains (FICI <0.1875).

The antileishmanial drug miltefosine possessed both in vitro and in vivo antifungal activities, with an MIC value of 2 μg/mL against 12 C. auris isolates. When used in combination with amphotericin B, miltefosine showed marginal synergistic effects against 3 out of 12 isolates (FICI = 0.5). In contrast, indifferent interaction was observed for the miltefosine and fluconazole combination against all of the tested isolates. Colistin, an antibiotic, had synergistic activity with caspofungin against several azole-resistant Candida spp. For C. auris, colistin used alone was totally ineffective (MIC >64 μg/mL). Synergistic activities were observed for the...
5.3. Chemosensitizers potentiating the activity of antifungal agents

Cowan’s group\textsuperscript{108} screened a diverse chemical library and identified azofloxin (28, a bis-benzoxidoxyindolinolindinone derivative) as an effective synergist with fluconazole against \textit{C. auris}. Azofloxin exerted species-selective synergistic activity against \textit{C. auris} that reduced the MIC value of fluconazole more than eightfold (FICI = 0.25). The synergistic activity was also observed in \textit{C. auris} clades I, II, and IV, whereas azofloxin was ineffective against clade III. In a mouse infections model of systemic \textit{C. auris}, azofloxin (10 mg/kg, subcutaneously, four times daily) significantly enhanced fluconazole (32 mg/kg, intraperitoneally, twice daily) activity in reducing the fungal burden. Unexpectedly, azofloxin alone also reduced the fungal burden despite it showing no \textit{in vitro} inhibitory activity against \textit{C. auris} growth. Further mechanistic studies revealed that the inhibition of the efflux pump Cdr1 was associated with the potency of azofloxin. Thus, Cdr1 may be an effective target for development of novel therapeutics.

Shaban et al. evaluated the anti-\textit{C. auris} activity of four phenolic natural products, and carvacrol (27) was found to be the most potent compound\textsuperscript{109}. Carvacrol had direct activity against the highest concentration (125 \textmu g/mL) and exerted synergistic and additive effects in combination with fluconazole, caspofungin, amphotericin B, and nystatin. Carvacrol also inhibited virulence factors of \textit{C. auris}, including proteinase production and adherence ability. Although echinocandins were used as the first-line therapy for the treatment of \textit{C. auris} infections, their activity against \textit{C. auris} biofilms was significantly lower than that against \textit{C. albicans}\textsuperscript{50}. Farnesol (29) is a quorum-sensing antibacterial molecule that has been demonstrated to enhance the activity of echinocandins against \textit{C. auris} biofilms\textsuperscript{109}. The synergism was observed for caspofungin (FICI range: 0.156–0.5), micafungin (FICI range: 0.133–0.281), and anidulafungin (FICI range: 0.14–0.375).

The antipsychotic drug haloperidol exhibited direct inhibitory effects against \textit{C. albicans}\textsuperscript{116}. Our group designed a series of haloperidol derivatives that showed improved antifungal activities\textsuperscript{111,117}. The compound B2 (30) exhibited potent synergistic activity against \textit{C. auris} when used in combination with itracanazole (FICI = 0.188) or voriconazole (FICI = 0.313)\textsuperscript{111}.

6. Drug repurposing

Drug repurposing has become an effective approach to rapidly identifying new therapeutics for emerging infectious disease\textsuperscript{118–120}. Three independent HTS studies have been performed to identify potential agents against \textit{C. auris} from among marketed drugs (Fig. 5)\textsuperscript{121–123}. Several hits were shown to possess potent anti-\textit{C. auris} activity when used alone or in combination with antifungal agents (Table 3).

Among such drugs, the effects of ebselen (31) have been well characterized in two screens\textsuperscript{122,123}. Ebselen is an antioxidant agent with diverse biological activities; it is currently undergoing clinical trials for various applications\textsuperscript{124,125}. Ebselen had IC\textsubscript{50} values in the range 0.2345–1.47 \textmu g/mL against 10 \textit{C. auris} clinical isolates\textsuperscript{123}. Moreover, ebselen effectively inhibited biofilm formation of \textit{C. auris} (IC\textsubscript{50} range: 5.864–9.781 \textmu g/mL)\textsuperscript{123}. Ebselen was unable to synergize with fluconazole, amphotericin B, or caspofungin\textsuperscript{123}, while it showed moderate synergism with anidulafungin\textsuperscript{124}. However, the \textit{in vivo} potency of ebselen against \textit{C. auris} has not been reported. In addition to \textit{C. auris}, ebselen also showed broad-spectrum antifungal activity\textsuperscript{123,126,127}. The antifungal target and mechanism of ebselen have not been fully characterized. The diverse activity of ebselen may be related to its electrophilic nature, meaning that it could interact with cysteine-rich proteins. In fungal cells, the antifungal activity of ebselen was associated with the inhibition of plasma membrane \textit{H}\textsuperscript{+}-ATPase, regulation of glutathione (GSH), and reactive oxygen species (ROS) production\textsuperscript{120–128}.

Suloctidil (32), an antiplatelet drug, has been reported to be active against \textit{C. albicans} and \textit{C. neoformans}\textsuperscript{129,130}. Suloctidil also showed significant inhibitory activity against \textit{C. auris}, with MIC values ranging from 4 to 8 \textmu g/mL\textsuperscript{122}. In addition, suloctidil was able to synergize with voriconazole against \textit{C. auris}, with FICI values ranging from 0.11 to 0.5. The synergistic antifungal activity of suloctidil may be due to vacuolar biogenesis and membrane trafficking\textsuperscript{131}.

Myriocin (33), a serine palmitoyltransferase inhibitor, showed IC\textsubscript{50} values of 0.94 and 0.47 \textmu mol/L against \textit{C. auris} 0384 and \textit{C. auris} 0385, respectively. Moreover, myriocin demonstrated a synergistic effect with fluconazole against 13 clinical isolates of \textit{C. auris} (FICI range: 0.49–0.53)\textsuperscript{121}.

Sertraline (34), an antidepressant agent, was reported to possess broad spectrum antifungal activity, including against \textit{C. auris}\textsuperscript{32–135}. Sertraline significantly inhibited the growth of \textit{C. auris}, with MIC values ranging from 20 to 40 \textmu g/mL\textsuperscript{132}. Sertraline displayed fungicidal activity against \textit{C. auris} and effectively inhibited virulence factors such as the yeast to hyphae formation and biofilm formation\textsuperscript{132}. The possible mechanism of action of sertraline was associated with cell membrane damage in \textit{C. auris}\textsuperscript{132}. CYP51, the target of azole antifungal agents, was suggested to be the target of sertraline by molecular docking. However, there is still a lack of experimental evidence to support this hypothesis. Recently, our group designed a series of sertraline derivatives by scaffold hopping\textsuperscript{136}. Compound D16 (35) showed potent activity against three \textit{C. auris} isolates (MIC range: 4–16 \textmu g/mL). Antifungal mechanistic studies revealed that compound D16 blocked the biosynthesis of ergosterol through the inhibition of \textit{\Delta}^{5,6}\textsuperscript{13}desaturase, a potential target for the development of anti-\textit{C. auris} therapeutics\textsuperscript{136}.

Mefloquine (36) is an antimalarial agent that was reported to possess moderate antifungal activity\textsuperscript{137}. Montoya et al. further evaluated the activity of mefloquine derivatives, and they identified several compounds with improved potency. Among these, compound 4377 (37) showed the best activity against five \textit{C. auris} isolates (MIC range: 2–4 \textmu g/mL)\textsuperscript{138}. However, this compound was still less active than caspofungin and amphotericin B. Mefloquine derivatives acted by multi-targeting mechanisms in which interference with the functions of mitochondria and vacuoles was preliminarily confirmed\textsuperscript{138}.

Disulfiram (38), an aldehyde dehydrogenase enzyme inhibitor for the treatment of alcohol dependence, was identified as an antifungal agent against \textit{C. auris}\textsuperscript{139}. Disulfiram exhibited superior activity against \textit{C. auris} over fluconazole, with MIC values ranging from 1 to 8 \textmu g/mL. In addition, disulfiram showed inhibitory activity against biofilm formation of \textit{C. auris} by increasing fungal cell aggregation, with SMIC\textsubscript{50} values ranging from 32 to 128 \textmu g/mL\textsuperscript{139}. Preliminary
mechanism studies indicated that disulfiram could combat drug resistance by inhibiting the ABC transporter proteins140. Alexidine dihydrochloride (39), an anticancer drug act by inhibiting mitochondrial tyrosine phosphatase, has been reported to possess antifungal and anti-biofilm activity against C. auris141. Alexidine dihydrochloride had MIC values in the range of 0.73–1.5 mg/mL against C. auris, and displayed low toxicity on lung epithelial cells and HUVECs (IC50 > 7.37 mg/mL). Moreover, alexidine dihydrochloride effectively inhibited biofilm formation and mature biofilm of C. auris, with SMIC50 values of 6 and 3 mg/mL, respectively141.

7. New targets for the development of anti-C. auris agents

7.1. Phosphatidylinositol—phosphatidylcholine transfer protein: Sec14p

Bugni’s group performed anti-C. albicans high-throughput screening of the microbiomes of marine animals through an integrated platform of metabolomic and genomic tools142. Turbinmicin (40), a highly oxidized polyketide, was identified to possess broad-spectrum inhibitory activity against Candida spp., Fusarium spp., Scedosporium spp., and Rhizopus spp. (MIC range: 0.03–0.5 μg/mL). In particular, turbinmicin was effective against C. auris (strain number: B11211) with an MIC value of 0.25 μg/mL. Further evaluation indicated that turbinmicin was fungicidal with low toxicity, and the maximum tolerated dose (MTD) in a mouse model was above 256 mg/kg. Turbinmicin also showed dose-dependent in vivo potency for the treatment C. auris infections. At the dose of 4 mg/kg, turbinmicin treatment led to a 3.6 log10 reduction in fungal burden compared with a blank control. The mode of action of turbinmicin was preliminary clarified by screening knockdown and knockout gene libraries of Saccharomyces cerevisiae. Sec14p, a phosphatidylinositol—phosphatidylcholine transfer protein, was validated as the molecular target of turbinmicin (Fig. 6A). Turbinmicin binds to the phospholipid binding pocket of Sec14p through hydrophobic and hydrogen bonding interactions (Fig. 6B).

The promising in vitro and in vivo antifungal activity and favorable mammalian safety profile have made turbinmicin a valuable lead compound. However, turbinmicin was administrated by intraperitoneal injection; this limited its further clinical development. After removal of the side chain by ester hydrolysis, the antifungal activity was reduced. Thus, structure optimization of turbinmicin into an orally active antifungal agent is required. To facilitate extensive SAR investigation, the difficulty involved in total synthesis should be solved. However, Sec14p may be further exploited as a drug target to design drug-like inhibitors against C. auris infections. Ergoline143, benzamide144, and picolinamide144 derivatives have been reported to be fungal Sec14p inhibitors. However, the antifungal activities of these Sec14p inhibitors were rather weak143,144. Fortunately, the crystal structure of Sec14p has been solved144; this could improve the efficiency of designing potent Sec14p inhibitors.

7.2. Casein kinase: Yck2

By screening a library of 736 protein kinase inhibitors, the arylpyrazolopyridine derivative GW461484A (41) was identified as a
profiling Yck2 belongs to the protein family of CK1 (casein molecular target of GW461484A by chemogenomic and biochemical of fluconazole. Furthermore, Yck2 was identified to be the mo-

C. auris pofungin against a multidrug-resistant

Interestingly, GW461484A also potentiated the activity of cas-

FIC80 (fractional inhibitory concentration 80 index) value lower

than 0.156, whereas it had little effect on the anti-

sensitizer to reverse caspofungin resistance against

Figure 6 Crystal structure of Sec14p (A, PDB code: 6F0E) and a proposed binding model of turbinmicin with Sec14p (B). The magenta mesh indicates turbiminicin, and the dashed green lines represent hydrogen bonding interactions.

Table 3 Natural peptides and synthetic derivatives with inhibitory activity against C. auris.

| Peptides         | Description                                      | Antifungal activity                                                                 | Ref. |
|------------------|--------------------------------------------------|--------------------------------------------------------------------------------------|------|
| Crotamine        | Natural peptide                                  | MIC range: 40–80 µmol/L                                                              | 164  |
| Myr-B            | Myristoylated lipopeptide                         | MIC: 16 µg/mL; MIC range: 16–32 µg/mL; In vivo potency in a Galleria mellonella infection model. | 165  |
| Peptide 3        | Cyclic temporin L peptide analogue                | MIC: 50 µmol/L; MFC: 50 µmol/L; 50% biofilm inhibition at 6.25 µmol/L; In vivo potency on the infected G. mellonella larvae without significant toxicity. | 59   |
| Pom-1            | A fragment of Clostricin 574                     | Planktonic cells IC50: 13.8 µg/mL                                                   | 166  |
| Pom-2            | A fragment of cecropin D-like peptide             | Biofilm IC50: 4.2 µg/mL                                                              | 166  |
| NCR169C17-38     | A derivative of specific cysteine-rich (NCR) peptide | MIC: 6.25 µmol/L; Additive effect with fluconazole                                  | 167  |
| NCR335C17-33     | A derivative of specific cysteine-rich (NCR) peptide | No direct activity; Synergic effect with fluconazole                                | 167  |
| Cm-p5            | Natural peptide                                  | MIC: 11 µg/mL                                                                        | 168  |
| Dimer 1 and 2    | Cyclic and helical-stabilized analogues of the antifungal peptide Cm-p5 | Inactive against planktonic cells                                                   | 169  |
| CR-184           | Cathelicidin-inspired AMPs                       | Biofilm IC50: 10–21 µg/mL                                                           | 170  |
| θ-Defensins      | 18-Amino-acid macrocyclic peptides               | MIC range: 3.125–6.25 µg/mL                                                         | 171  |
| AF4              | Lipopeptide homologues                           | MIC: 3.48 µg/mL; MFC: 3.48 µg/mL; Inhibition of biofilm formation: SMIC50 = 6.96 µg/mL | 172  |

7.3. Acetohydroxyacid synthase

Acetohydroxyacid synthase (AHAS), an enzyme in the biosynthesis pathway of branched-chain amino acid, has been demonstrated as a promising target for the development of antifungal agents against C. auris. Guddat et al. expressed and obtained the AHAS from C. auris (CauAHAS), and identified several sulfonylurea and triazolopyrimidine herbicides as potent antifungal inhibitors against C. auris (MIC<sub>50</sub> < 5 µmol/L), with the Kᵢ values of < 2 µmol/L for CauAHAS. Among them, bensulfuron methyl (BSM, 42), a sulfonylurea inhibitor, exhibited the best fungicidal potency with the MIC<sub>50</sub> values of 0.09 µmol/L. BSM was also an excellent inhibitor for preventing the biofilms formation of C. auris (SMIC<sub>50</sub> = 0.6 µmol/L). Cell viability assays revealed that BSM was non-cytotoxic to human embryonic kidney (HEK)-293 cells at the concentrations of < 100 µmol/L. The possible binding model of these inhibitors with CauAHAS was identified by homology modelling based on the crystal complex of C. albicans AHAS with chlorimuron ethyl (CE, 43), an analogue of compound 42. CE interacted with the binding sites of CauAHAS by hydrophobic, hydrogen bonding and π–π stacking interactions (Fig. 8). These data indicated that CauAHAS was a viable target for treating C. auris infections.

7.4. New chemical scaffolds against C. auris

7.4.1. Rocaglates

Cowan’s group screened a library containing 2454 compounds to identify anti-C. auris compounds, and the hits shared a common rocaglate scaffold (44). Representative compound CMLD010515 (Fig. 9) displayed inhibitory activity against C. auris (active concentration: < 12.5 µmol/L) and was demonstrated to be fungicidal. Interestingly, the anti-C. auris activity was species-specific, because the rocaglates were inactive against pathogenic related Candida species such as C. albicans. The antifungal mechanisms of rocaglates were preliminary elucidated; these involved inhibition of translation
initiation in C. auris, triggering an apoptosis-like cell death program and blocking vacuolar homeostasis.\textsuperscript{148}

7.4.2. Hydroxyquinolines: Nitroxoline
The hydroxyquinoline derivate nitroxoline (45) is an antibacterial agent used for urinary tract infections. It also has inhibitory activity against C. auris, with MICs ranging from 0.125 to 1 $\mu$g/mL (35 isolates: MIC$_{50}$ = 0.25 $\mu$g/mL, MIC$_{90}$ = 0.5 $\mu$g/mL).\textsuperscript{149} It was more potent than amphotericin B (MIC > 1 $\mu$g/mL in 4/35 isolates) and fluconazole (MIC > 4 $\mu$g/mL in 31/35 isolates). Nitroxoline was proposed as a potential treatment option for C. auris candiduria. However, its in vivo efficacy remains to be confirmed.

7.4.3. Halogenated salicylanilides
In an antivirulence phenotypic screen, halogenated salicylanilides 1 and its analog niclosamide (46, 47) exhibited potent inhibitory activities against C. albicans filamentation and biofilm formation.\textsuperscript{150} Both were also active against the biofilms of C. auris in a dose-dependent manner.\textsuperscript{150} Mechanistic studies revealed that the mitochondrial protein import machinery may be involved in the activity of halogenated salicylanilides.

7.4.4. Pyrimidinedione: MYC-053
The pyrimidinedione derivative MYC-053 (48) showed broad-spectrum effects against Candida spp., Cryptococcus spp., and Pneumocystis spp. It had an MIC value of 4 $\mu$g/mL against 5 C. auris isolates, and it was also active against several strains resistant to fluconazole and caspofungin.\textsuperscript{151}

7.4.5. Macroyclic amidinoureas: BM1
BM1 (49) is a derivative of macrocyclic amidinoureas whose chemical structure features an amphiphilic macrocycle, a methylene linker, and a terminal alkenyl guanidine.\textsuperscript{152} BM1 showed potent inhibitory activity against various fluconazole-sensitive and fluconazole-resistant Candida spp., including C. auris isolates. The MIC value of BM1 against 18 C. auris isolates was in the range of 8 $\mu$g/mL to 64 $\mu$g/mL.\textsuperscript{153} However, the antifungal activity of BM1 against C. auris was significantly lower than that against C. albicans (MIC range: 0.125–2 $\mu$g/mL). The activity of BM1 against resistant fungi was associated with the overexpression of ABC transporters.\textsuperscript{153} BM1 showed in vivo efficacy for treating infections by drug-resistant C. albicans, whereas the in vivo potency against C. auris is still unknown.

7.4.6. Oxadiazolylthiazoles
Hagras et al. synthesized a series of oxadiazolylthiazole derivatives and identified selective antifungal agents.\textsuperscript{154} Diaminocyclohexyl derivative 50 showed broad-spectrum in vitro antifungal activity, including against C. auris. It had MIC values of 4, 2, and 2 $\mu$g/mL against C. auris 381, C. auris 383, and C. auris 384, respectively. Moreover, compound 50 showed low toxicity against human colorectal adenocarcinoma (Caco-2) and monkey fibroblast-like kidney epithelial (Vero) cells, with CC$_{50}$ values larger than 64 $\mu$g/mL.

7.4.7. Phenylthiazoles
Mohammad et al. assayed 85 synthetic phenylthiazole derivatives for inhibitory activity against drug-resistant C. albicans.\textsuperscript{155} Thiazole-
aminoguanidine derivative 51 showed the most potent antifungal activity, with a broad spectrum. Compound 51 had an MIC value of 2 μg/mL against eight C. auris isolates, a value that was more potent than fluconazole (MIC > 64 μg/mL) and comparable to amphotericin B (MIC range: 0.50–2 μg/mL). Compound 51 showed rapid fungicidal activity against C. auris within 30 min. At the concentration of 2 μg/mL, compound 51 effectively inhibited biofilm formation of C. auris (91.2% reduction), and it was equally effective as amphotericin B (92.4% reduction). In contrast, the cytotoxicity of compound 51 against mammalian cells was significantly lower than that of amphotericin B. In a C. elegans model with C. auris, compound 51 prolonged the survival of infected nematodes by about 70% at the concentration of 10 μg/mL.

### 7.4.8 2-Aryloxazolines

Stefani’s group synthesized a series of 2-aryloxazoline derivatives and assayed these for inhibitory activity against C. albicans. Most compounds showed comparable or superior antifungal activity to fluconazole. The compounds 4i (52) and 9i (53) were also effective against C. auris isolates CBS 10913 (MIC = 0.06 μg/mL) and CBS 12766 (MIC = 2 μg/mL).

### 8. Antimicrobial peptides

Antimicrobial peptides (AMPs) are emerging an attractive area in antifungal therapy due to the important roles in human innate immunity and host defense and the low risk of inducing MDR. Several antifungal AMPs have also shown potent activity against C. auris (Table 3).

Histatin 5 (Hst 5) was reported to possess good antifungal activity against C. albicans. In a susceptibility assay of 10 C. auris clinical isolates, Hst 5 showed fungicidal activity against the majority of tested isolates, killing 55%–90% of C. auris cells at the concentration of 7.5 μmol/L. The high tolerance of C. auris strains to oxidative stress was possibly associated with the killing effect of Hst 5.

Human cathelicidin peptides LL-37 showed both direct and synergistic activities against C. auris. The growth inhibitory activity of LL-37 was moderate in a collection of 10 clinical strains (MIC range: 25–100 μg/mL; MFC range: 50–200 μg/mL). LL-37 also effectively synergized with antifungal agents such as fluconazole (80% of strains, FICI range: 0.27–0.5), amphotericin B (100% of strains, FICI range: 0.13–0.31), and caspofungin (100% of strains, FICI range: 0.13–0.26). The antifungal mechanistic studies revealed that LL-37 acted by disrupting the cell membrane, causing oxidative stress, and arresting the S phase of cell cycle of C. auris.

AMPs are generally the substrates for proteases and are prone to be degraded in vivo. Thus, non-peptide AMP mimics were designed to overcome the limitations of peptide molecules. Ceragenins feature a bile acid scaffold and a lipid chain that mimics the common amphiphilic secondary structure of AMPs and that has shown broad-spectrum antifungal activity. The compound ceragenin CSA-131 (54) had potent fungistatic and fungicidal activity against a set of 100 C. auris clinical isolates (MIC range: 0.5–8 μg/mL; MFC range: 5–40 μg/mL).

![Chemical structures of new chemotypes with inhibitory activity against C. auris.](image-url)
range: 2–64 μg/mL) and was generally more potent than fluconazole, caspofungin, and amphotericin B. The antifungal activity of CSA-131 was clade independent without variation between the four clades (overall mode: 1 μg/mL; MIC\text{90} = 1 μg/mL). Notably, no loss of inhibitory activity was observed for those isolates resistant to fluconazole and/or echinocandin. CSA-131 also effectively inhibited the activity of C. auris biofilm formation (SMIC\text{90} range: 2–4 μg/mL). In an \textit{ex vivo} infections model of mucosal tissues, topical use of CSA-131 (2% gel and cream formulations) resulted in significant reductions of fungal burdens.

9. \textbf{Perspectives and conclusions}

Although some progress has been made, we still do not clearly know where \textit{C. auris} originated or why \textit{C. auris} has independently and simultaneously spread worldwide. Our understanding of the virulence, risk factors, and mechanisms of drug resistance remains in its infancy. In some cases, the results obtained from different clades (strains) of \textit{C. auris} are controversial or contradictory. Thus, there is still a long way to go before we can fully understand this novel fungal pathogen. To tackle the threat of \textit{C. auris}, improvement in early diagnosis, control measures, and education of healthcare providers will help to reduce the incidence of infections. More importantly, the development of effective therapeutics is urgently needed to improve clinical outcomes and decrease mortality.

Although echinocandins have been recommended as the first-line therapy, the options for effective treatment of \textit{C. auris} infections are rather limited. Several antifungal agents in clinical development have been shown to be effective against \textit{C. auris} through \textit{in vitro} and \textit{in vivo} evaluations. These compounds have also shown favorable pharmacokinetic and safety profiles with a low risk of drug–drug interactions in clinical trials. However, only two clinical trials have been started for \textit{C. auris} infections (ibrexafungerp and APX001) with as yet undisclosed data. Therefore, more clinical studies are required to validate the potential usefulness of these candidates in clinical practice.

Synergistic drug combinations have been suggested as a potential option for the treatment of \textit{C. auris} infections. Although a number of active combinations have been reported, it is premature to predict their clinical utility, because most have only been evaluated \textit{in vitro}. Additional evaluation in animal models or eventually in clinical trials is required to identify useful combinations for pan-resistant \textit{C. auris}.

Drug repurposing has been demonstrated to be a useful approach to accelerate the drug development process, particularly for emerging infectious diseases. Several “non-antifungal” drugs have been shown to be active in inhibiting the growth of \textit{C. auris} when used alone or in combination with the antifungal agents. However, such known drugs could hardly be used directly in clinical application due to limited potency and side effects. An important value of drug repurposing is to offer drug-like lead compounds for the optimization of therapeutic efficacy and safety profiles. For example, our group has designed a series of new derivatives of sertraline and piperidol as antifungal agents, and it has also identified new leads with improved antifungal activity and reduced original activity of approved agents. With better understanding of the virulence and resistant mechanisms of \textit{C. auris}, the discovery and identification of new targets is highly important for developing effective therapeutics with new modes of action. Sec14p and Yck2 have been preliminarily identified as potential targets for \textit{C. auris} infections. These targets were identified through chemogenomic profiling of active compounds. Biology-inspired discovery of new targets is still rare, possibly due to limited knowledge concerning \textit{C. auris}. Another problem for the new antifungal targets is the inconsistency between molecular and antifungal activity. After proof-of-concept validation, extensional medicinal chemistry exploration of the inhibitors would contribute to identifying selective and drug-like inhibitors. Recently, a large number of anti-\textit{C. auris} compounds were identified by phenotypic antifungal screening, and their molecular targets are mostly unknown. These bioactive compounds could be used as chemical probes to look for new targets by chemogenomic profiling; this would provide an alternative for target discovery in \textit{C. auris}. With a better understanding of \textit{C. auris}, increased medicinal chemistry effort, and more preclinical and clinical trials, highly effective antifungal drugs will become a reality for the treatment of patients with severe \textit{C. auris} infections.

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\textbf{Author contributions}

Yahui Huang and Wanzhen Yang performed the literature search and data collection. Chuanguan Sheng conceived the concept of the study. Jie Tu designed and regenerated the conceptual pictures. Jie Tu, Na Liu and Chunquan Sheng prepared and revised the manuscript. All authors have read and approved the final manuscript.

\textbf{Conflicts of interest}

The authors declare no conflicts of interest.

\textbf{References}

1. Lamoth F, Lockhart SR, Berkow EL, Calandra T. Changes in the epidemiological landscape of invasive candidiasis. \textit{J Antimicrob Chemother} 2018;73:14–13.

2. Satoh K, Makimura K, Hasumi Y, Nishiyama Y, Uchida K, Yamaguchi H. \textit{Candida auris} sp. nov., a novel ascomycetous yeast isolated from the external ear canal of an inpatient in a Japanese hospital. \textit{Microbiol Immunol} 2009;53:41–4.

3. Du H, Bing J, Hu T, Ennis CL, Noble CJ, Huang G. \textit{Candida auris}: epidemiology, biology, antifungal resistance, and virulence. \textit{PLoS Pathog} 2020;16:e1008921.

4. Cortegiani A, Missier G, Fasciana T, Giammanco A, Giarratano A, Chowdhary A. Epidemiology, clinical characteristics, resistance, and treatment of infections by \textit{Candida auris}. \textit{Intensive Care Med} 2018;44:671–82.

5. Welsh RM, Bentz ML, Shams A, Houston H, Lyons A, Rose L, et al. Survival, persistence, and isolation of the emerging multidrug-resistant pathogenic yeast \textit{Candida auris} on a plastic health care surface. \textit{J Clin Microbiol} 2017;55:2996–3005.

6. Proctor DM, Dangana T, Sexton DJ, Fukuda C, Yelin RD, Stanley M, et al. Integrated genomic, epidemiologic investigation of \textit{Candida auris} infections.
auris skin colonization in a skilled nursing facility. Nat Med 2021;27:1401–9.
7. Ben-Ami R, Berman J, Novikov A, Bash E, Shachor-Meyouhas Y, Zakin S, et al. Multidrug-resistant Candida haemulonii and C. auris, tel Aviv, Israel. Emerg Infect Dis 2017;23:192–203.
8. Chatterjee S, Alamplali SV, Nageshan RK, Chettiar ST, Joshi S, Tatu US. Draft genome of a commonly misdiagnosed multidrug resistant pathogen. Candida auris. BMC Genomics 2015;16:686–97.
9. Munoz JF, Gade L, Chow NA, Loparev VN, Juieng P, Berkow EL, et al. Potential fifth clade of Candida auris, Iran, 2018. Emerg Infect Dis 2019;25:1780–1.
11. Chowdhary A, Sharma C, Meis JF. Candida auris: a rapidly emerging cause of hospital-acquired multidrug-resistant fungal infections globally. PLoS Pathog 2017;13:e1006290.
12. Chowdhary A, Sharma C, Duggal S, Agarwal K, Prakash A, Singh PK, et al. New clonal strain of Candida auris, Delhi, India. Emerg Infect Dis 2013;19:1670–3.
13. Calvo B, Meo AS, Perozo-Mena A, Hernandez M, Francisco EC, Hagen F, et al. First report of Candida auris in America: clinical and microbiological aspects of 18 episodes of candidemia. J Infect 2016;73:369–74.
14. Chow NA, de Groot T, Badali H, Abastabar M, Chiller TM, Meis JF, et al. Multidrug-resistant Candida auris infections globally. Emerg Infect Dis 2017;73:1780–9.
15. Osei Sekyere J. New clonal strain of Candida auris. Clinical and epidemiological situation, laboratory capacity and preparedness in Europe. Euro Surveill 2018;23:18–136.
16. Schelenz S, Hagen F, Rhodes JL, Abdolrasouli A, Chowdhary A, Hall A, et al. First hospital outbreak of the globally emerging Candida auris in a European hospital. Antimicrob Resist Infect Control 2016;5:35–9.
17. Cadnum JL, Shaikh AA, Piedrahita CT, Sankar T, Jenson AL, Larkin EL, et al. Effectiveness of disinfectants against Candida auris and other Candida species. Infect Control Hosp Epidemiol 2017;38:1240–3.
18. Abdolrasouli A, Armstrong-James D, Ryan L, Schelenz S. In vitro efficacy of disinfectants utilised for skin decontamination and environmental decontamination during a hospital outbreak with Candida auris. Mycoses 2017;60:758–63.
19. Bruno M, Kersten S, Bain JM, Jaeger M, Rosati D, Kruppa MD, et al. Transcriptional and functional insights into the host immune response against the emerging fungal pathogen. Candida auris. Nat Microbiol 2020;5:1516–31.
20. Wang Y, Zou Y, Chen X, Li H, Yin Z, Zhang B, et al. Innate immune responses against the fungal pathogen. Candida auris. Nat Commun 2022;13:3533–73.
21. Yadav B, Mora-Montes HM, Wagener J, Cunningham I, West L, Haynes K, et al. Differences in fungal immune recognition by monocytes and macrophages. N-mannan can be a shield or activator of immune recognition. Cell surf 2020;6:100042–54.
22. Chowdhary A, Prakash A, Sharma C, Kordalewska M, Kumar A, Sarma S, et al. A multicentre study of antifungal susceptibility patterns among 350 Candida auris isolates (2009–17) in India: role of the ERG11 and FKS1 genes in azole and echinocandin resistance. J Antimicrob Chemother 2018;73:891–9.
23. Pappas PG, Kaufman CA, Andes DR, Clancy CJ, Murr KA, Ostrosky-Zeichner L, et al. Clinical practice guideline for the management of Candidiasis: 2016 update by the infectious diseases society of America. Clin Infect Dis 2016;62:e1–50.
Infections and therapeutic strategies: mechanisms of action for traditional and alternative agents. *Front Microbiol* 2018;9:1351—9.
44. Rybak JM, Doorley LA, Nishimoto AT, Barker KS, Palmer GE, Rogers PD. Abrogation of triazole resistance upon deletion of CDR1 in a clinical isolate of. *Candida auris*. *Antimicrob Agents Chemother* 2019;63:e00057-19.
45. Wasi M, Khandelwal NK, Moorhouse AJ, Nair R, Vishwakarma P, Bravo Ruiz G, et al. ABC transporter genes show upregulated expression in drug-resistant clinical isolates of *Candida auris*: a genome-wide characterization of ATP-binding cassette (ABC) transporter genes. *Front Microbiol* 2019;10:1445—61.
46. Escandon P, Chow NA, Caceres DH, Gade L, Berkow EL, Richardson MD, et al. Transcriptome analysis and profiling of *Candida auris* reveals novel insights into biofilm-mediated resistance. *mSphere* 2018;3:e00334-18.
47. Sherry L, Ramage G, Kean R, Borman A, Johnson EM, Richardson MD, et al. Biofilm-forming capability of highly virulent, multidrug-resistant. *Candida auris*. *Emerg Infect Dis* 2017;23:328—31.
48. Dominguez EG, Zarnowski R, Choy HL, Zhao M, Sanchez H, Nett JE, et al. Conserved role for biofilm matrix polysaccharides in *Candida auris* drug resistance. *mSphere* 2019;4:e00680-18.
49. Larkin E, Delaney C, Sherry L, Borman A, Johnson EM, Richardson MD, et al. Biofilm-forming capability of highly virulent, multidrug-resistant. *Candida auris*. *Emerg Infect Dis* 2017;23:328—31.
50. Romero D, Aguilera-Correia JI, Gadea I, Vinuela-Sandoval L, Garcia-Rodriguez J, Esteban J. *Candida auris*: a comparison between planktonic and biofilm susceptibility to antifungal drugs. *J Med Microbiol* 2018;68:e00485-17.
51. Horton MV, Johnson CJ, Kernien JF, Patel TD, Lam BC, Cheong JZA, et al. *Candida auris* forms high-burden biofilms in skin niche conditions and on porcine skin. *mSphere* 2020;5:e00910—9.
52. Arendrup MC, Prakash A, Miletiajs I, Sharma C, Chowdhry A. Comparison of EUCAST and CLSI reference microdilution MICs of eight antifungal compounds for *Candida auris* and associated tentative epidemiological cutoff values. *Antimicrob Agents Chemother* 2017;61:e00485-17.
53. Fakhim H, Chowdhry A, Prakash A, Vaeti A, Dannaoui E, Meis JF, et al. *In vitro* interactions of echinocandins with triazoles against multidrug-resistant. *Candida auris*. *Antimicrob Agents Chemother* 2017;61:e01056-17.
54. Nagy F, Toth Z, Nyikos F, Forgacs L, Jakab A, Borman AM, et al. *In vitro* and *in vivo* interaction of caspofungin with isavuconazole against *Candida auris* planktonic cells and biofilms. *Med Mycol* 2021;59:1015—23.
55. Eldesokey HE, Salama EA, Lanman NA, Hazzun TR, Selemee MN. Potent Synergic interactions between lipopavir and azole anti-fungal drugs against emerging multidrug-resistant. *Candida auris*. *Antimicrob Agents Chemother* 2020;65:e00684-20.
56. Bellavita R, Maione A, Merlino F, Siciliano A, Dardano P, De Stefano L, et al. Antifungal and antibiofilm activity of cyclic temporin L peptide analogues against albicans and non-albicans *Candida* species. *Pharmaceutics* 2022;14:454—79.
84. Shaw KJ, Ibrahim AS. Fosmanogepix: a review of the first-in-class triterpenoid antifungal in development for the treatment of Candida auris infections. Antimicrob Agents Chemother 2021;65:e02694-20.

85. Ghannoun M, Arendrup MC, Chaturvedi S, Chaturvedi V. Efficacy of T-2307, a novel arylamidine, against fluconazole-resistant Candida auris and in vivo efficacy with delayed initiation of therapy in an experimental model of invasive candidiasis. Antimicrob Agents Chemother 2021;65:e01988-20.

86. Chu S, Long L, McCormick TS, Chaturvedi S, et al. Ibrexafungerp: a novel oral tripterpenoid antifungal in development for the treatment of Candida auris. Antimicrob Agents Chemother 2021;65:e01988-20.

87. Schwab KL, El-Sharkawy A. Development of new antifungal drugs against emerging multidrug-resistant Candida auris. J Antimicrob Chemother 2021;76:1395-6.

88. Wu Y, Totten M, Memon W, Ying C, Zhang SX. Activity of APX001A against Candida auris. Antimicrob Agents Chemother 2021;65:e00656-20.

89. Berkow EL, Lockhart SR. Activity of novel antifungal compound APX001A against a large collection of Candida auris. J Antimicrob Chemother 2018;73:3060-2.

90. Hager CL, Larkin EL, Long L, Zohra Abidi F, Shaw KJ, Ghannoun MA. In vitro and in vitro evaluation of the antifungal activity of APX001A/APX001 against Candida auris. Antimicrob Agents Chemother 2018;62:e02319-17.

91. Wiederhold NP, Najvar LK, Jaramillo R, Olivo M, Patterson H, Connell A, et al. The novel arylamidine T-2307 demonstrates in vitro and in vivo activity against Candida auris. Antimicrob Agents Chemother 2020;64:e02195-19.

92. Shibata T, Takahashi T, Yamada E, Kimura A, Nishikawa H, Hayakawa H, et al. T-2307 causes collapse of mitochondrial membrane potential in yeast. Antimicrob Agents Chemother 2012;56:5892-7.

93. Abe M, Nakamura S, Kinjo Y, Masuyama Y, Mitsuya M, Kaku M, et al. Efficacy of T-2307, a novel arylamidine, against ocular complications of disseminated candidiasis in mice. J Antimicrob Chemother 2019;74:1327-32.

94. Shibata T, Takahashi T, Yamada E, Kimura A, Nishikawa H, Hayakawa H, et al. T-2307 causes collapse of mitochondrial membrane potential in yeast. Antimicrob Agents Chemother 2012;56:5892-7.
113. Zeidler U, Bougnoux ME, Lupan A, Helynck O, Doyen A, Garcia Z, et al. Synergy of the antibiotic colistin with echinocandin antifungals in Candida species. *J Antimicrob Chemother* 2013;68:1285–96.

114. Schwarz P, Nikolskiy I, Bidaud AL, Sommer F, Bange G, Dannaoui E. *In vitro* activity of amphotericin B in combination with colistin against fungi responsible for invasive infections. *J Fungi* (Basel) 2022;8:115–31.

115. Schwarz P, Bidaud AL, Dannaoui E. *In vitro* synergy of isavuconazole in combination with colistin against *Candida auris*. *Sci Rep* 2020;10:21448–56.

116. Stylianou M, Kuleskiiy E, Lopes JP, Granlund M, Wennerberg K, Urban CF. Antifungal application of nonantifungal drugs. *Antimicrob Agents Chemother* 2014;58:1055–62.

117. Ji C, Liu N, Tu J, Li Z, Han G, Li J, et al. Drug Repurposing of Haloperidol: discovery of new benzocyclene derivatives as potent antifungal agents against cryptococcosis and candidiasis. *ACS Infect Dis* 2020;6:768–86.

118. Yi D, Li Q, Wang H, Lv K, Ma L, Wang Y, et al. Repurposing of berbamidine hydrochloride to inhibit Ebola virus by targeting viral glycoprotein. *Acta Pharmacol Sin B* 2022. Available from: http://doi.org/10.1016/j.apsb.2022.05.023.

119. Yan H, Sun J, Wang K, Wang H, Wu S, Bao L, et al. Repurposing carrimycin as an antiviral agent against human coronaviruses, including the currently pandemic SARS-CoV-2. *Acta Pharmacol Sin B* 2021;11:2850–8.

120. Wang G, Li L, Wang X, Li X, Zhang Y, Ju J, et al. *Hypericum* enhances β-lactam antibiotics activity by inhibiting sarA expression in meticillin-resistant. *Staphylococcus aureus*. *Acta Pharmacol Sin B* 2019;9:174–82.

121. Cheng YS, Roma JS, Shen M, Mota Fernandes C, Tsang PS, Forbes HE, et al. Identification of antifungal compounds against multidrug-resistant *Candida auris* utilizing a high-throughput drug-repurposing screen. *Antimicrob Agents Chemother* 2021;65:e01305–20.

122. de Oliveira HC, Monteiro MC, Rossi SA, Pemen J, Ruiz-Gaitán A, Mendes-Giannini MJS, et al. Identification of off-patent compounds that present antifungal activity against the emerging fungal pathogen. *Candida auris*. *Front Cell Infect Microbiol* 2019;9:83–93.

123. Wall G, Chaturvedi AK, Wormley Jr FL, Wiederhold NP, Patterson HP, Patterson TF, et al. Screening a repurposing library for inhibitors of multidrug-resistant *Candida auris* identifies ebselen as a repositionable candidate for antifungal drug development. *Antimicrob Agents Chemother* 2018;62:e01084–18.

124. Sant C, Scirmici C, Sancineto L, Ebselen and analogues: pharmacological properties and synthetic strategies for their preparation. *Molecules* 2021;26:4230–55.

125. Wang J, Wang P, Dong C, Zhao Y, Zhou J, Yuan C, et al. Mechanisms of ebselen as a therapeutic and its pharmacology applications. *Future Med Chem* 2020;12:2141–60.

126. Billack B, Santoro M, Lau-Cam C. Growth inhibitory action of ebselen on flavocazole-resistant *Candida albicans*: role of the plasma membrane H+–ATPase. *Microb Drug Resist* 2009;15:77–83.

127. Thangamani S, Eldesouky HE, Mohammad H, Pasuzzi PE, Avranova L, Hazbun TR, et al. Ebselen exerts antifungal activity by regulating glutathione (GSH) and reactive oxygen species (ROS) production in fungal cells. *Biochim Biophys Acta Gen Subj* 2017;1861:3002–11.

128. Azad GK, Singh V, Mandal P, Singh P, Golla U, Baranwal S, et al. Ebselen induces reactive oxygen species (ROS)-mediated cytotoxicity in *Saccharomyces cerevisiae* with inhibition of glutamate dehydrogenase being a target. *FEBS Open Bio* 2014;4:77–89.

129. Butts A, DiDone L, Koselny K, Baxter BK, Chabrier-Rosello Y, Wellington M, et al. A repurposing approach identifies off-patent drugs with fungidical cryptococcal activity, a common structural chemotype, and pharmacological properties relevant to the treatment of cryptococcosis. *Eukaryot Cell* 2013;12:278–87.

130. Zeng B, Li J, Wang Y, Chen P, Wang X, Cui J, et al. *In vitro* and *in vivo* effects of solucitid on growth and biofilm formation of the opportunistic fungus. *Candida albicans*. Oncotarget 2017;8:69972–82.

131. Spitzer M, Griffiths E, Blakely KM, Wildenhan J, Eijm L, Rossi L, et al. Cross-species discovery of synergistic drug combinations that potentiate the antifungal fluconazole. *Mol Syst Biol* 2011;7:499–513.

132. Gowri M, Jayashree B, Jeyakanthan J, Girjka ER. Sertraline as a promising antifungal agent: inhibition of growth and biofilm of *Candida auris* with special focus on the mechanism of action. *In vitro*. *J Appl Microbiol* 2020;128:426–37.

133. Trevino-Rangel RJ, Villanueva-Lozano H, Mendez-Galommo KS, Solis-Villegas EM, Becerril-Garcia MA, Montoya AM, et al. *In vivo* evaluation of the antifungal activity of sertraline against *Aspergillus fumigatus*. *J Antimicrob Chemother* 2019;74:663–6.

134. Lass-Florl C, Dierich MP, Fuchs D, Semenitz E, Ledochowski M. Antifungal activity against *Candida species* of the selective serotonin-reuptake inhibitor, sertraline. *Clin Infect Dis* 2001;33:E135–6.

135. Rhein J, Hupper Hullsieck K, Tugume L, Nuwagura E, Mpoza E, Evans EE, et al. Adjunctive sertraline for HIV-associated cryptococcal meningitis: a randomised, placebo-controlled, double-blind phase 3 trial. *Lancet Infect Dis* 2019;19:843–51.

136. Li W, Yun Z, Ji C, Tu J, Yang W, Li J, et al. Discovery of novel sertraline derivatives as potent anti-cryptococcus agents. *J Med Chem* 2022;65:6541–54.

137. Kunin CM, Ellis WY. Antimicrobial activities of mellofane and a series of related compounds. *Antimicrob Agents Chemother* 2000;44:848–52.

138. Montoya MC, Beattie S, Alden KM, Krysan DJ. Derivatives of the antimarial drug mellofane are broad-spectrum antifungal molecules with activity against drug-resistant clinical isolates. *Antimicrob Agents Chemother* 2020;64:e02331-19.

139. Hao W, Qiao D, Han Y, Du N, Li X, Fan Y, et al. Identification of disulfiram as a potential antifungal drug by screening small molecular libraries. *J Infect Chemother* 2021;27:696–701.

140. Khan S, Singhal S, Mathur T, Upadhyay DJ, Rattan A. Antifungal potential of disulfiram. *Nippon Ishinkin Gakkai Zasshi* 2007;48:109–13.

141. Mamouei Z, Alqarihi A, Singh S, Xu S, Mansour MK, Ibrahim AS, et al. Alden KM, Pakalnis PM, Ibrahim AS, et al. Alexidine dihydrochloride has broad-spectrum activities against diverse fungal pathogens. *mSphere* 2018;3.e00539-18.

142. Zhang F, Zhao M, Braun DR, Erickson SS, Piotrowski JS, Nelson J, et al. A marine microbiome antifungal targets urgent-threat drug-resistant fungi. *Science* 2020;370:974–8.

143. Filipuzzi I, Cotesia S, Perruccio F, Knapp B, Fu Y, Studer C, et al. High-resolution genetics identifies the lipid transfer protein Sec14p as target for antifungal ergolines. *PLoS Genet* 2016;12:e1006374.

144. Pries V, Nocker C, Khan D, Johnen P, Hong Z, Tripathi A, et al. Target identification and mechanism of action of plicolimamide and benzamide chemotypes with antifungal properties. *Cell Chem Biol* 2018;25:279–90, e7.

145. Caplan T, Lorente-Macias A, Stogijs PJ, Evdokimova E, Hyde S, Wellington MA, et al. Overcoming fungal echinocandin resistance through inhibition of the non-essential stress kinase Yck2. *Cell Chem Biol* 2020;27:269–82, e5.

146. Garcia MD, Chua SMH, Low YS, Lee YT, Agnew-Francis K, Wang JG, et al. Commercial AHAS-inhibiting herbicides are promising drug leads for the treatment of human fungal pathogenic infections. *Proc Natl Acad Sci U S A* 2018;115:E9649-e58.

147. Agnew-Francis KA, Tang Y, Lin X, Low YS, Sun SJ, Kuo A, et al. Herbicides that target acetohydroxyacid synthase are potent inhibitors of the growth of drug-resistant. *Candida auris*. *ACS Infect Dis* 2020;6:2901–12.

148. Iyer KR, Whitesell L, Porco Jr JA, Henkel T, Brown LE, Robbins N, et al. Translation inhibition by rogalates activates a species-specific cell death program in the emerging fungal pathogen. *Candida auris*. *mBio* 2020;11:e03529-19.
149. Fuchs F, Hof H, Hofmann S, Kurzai O, Meis JF, Hamprecht A. Antifungal activity of nitrooxoline against Candida auris isolates. Clin Microbiol Infect 2021;27:1697, e7-e10.

150. Garcia C, Burgain A, Chailloit J, Pic E, Khemiri I, Sellam A. A phenotypic small-molecule screen identifies halogenated salicylanilides as inhibitors of fungal morphogenesis, biofilm formation and host cell invasion. Sci Rep 2018;8:11559–64.

151. Tetz G, Collins M, Vikina D, Tetz V. In vitro activity of a novel antifungal compound, MYC-053, against clinically significant antifungal-resistant strains of Candida glabrata, Candida auris, Cryptococcus neoformans, and Pneumocystis spp. Antimicrob Agents Chemother 2019;63:e01975-18.

152. Deodato D, Maccari G, De Luca F, Sanfilippo S, Casian A, Martini R, et al. Biological characterization and in vivo assessment of the activity of a new synthetic macrocyclic antifungal compound. J Med Chem 2016;59:3854–66.

153. Orofino F, Truglio GI, Fiorucci D, D’Agostino I, Borgini M, Poggialini F, et al. In vitro characterization, ADME analysis, and histological and toxicological evaluation of BM1, a macrocyclic amidinourea active against azole-resistant Candida strains. Int J Antimicrob Agents 2020;55:105865–72.

154. Hagras M, Salama EA, Sayed AM, Abutaleb NS, Kotb A, Seleem MN, et al. Oxadiazolothiazoles as novel selective antifungal agents. Eur J Med Chem 2020;189:112046–56.

155. Mohammad H, Eldeosouky HE, Hazbun T, Mayhoub AS, Seleem MN. Identification of a phenylthiazole small molecule with dual antifungal and antibiofilm activity against Candida albicans and Candida auris. Sci Rep 2019;9:18941–53.

156. Argomedo LMZ, Barroso VM, Barreiro CS, Darbem MP, Ishida K, Stefani HA. Novel 2-aryloxazoline compounds exhibit an inhibitory effect on Candida spp., including antifungal-resistant isolates. ACS Med Chem Lett 2020;11:2470–5.

157. Buda De Cesare G, Cristy SA, Garsin DA, Lorenz MC. Antimicrobial peptides: a new frontier in antifungal therapy. mBio 2020;11:e02123-20.

158. Pathirana RU, Friedman J, Norris HL, Salvatori O, McCall AD, Kay J, et al. Fluconazole-resistant Candida auris is susceptible to salivary histatin 5 killing and to intrinsic host defenses. Antimicrob Agents Chemother 2018;62:e01872-17.

159. Rather IA, Sabir JSM, Asseri AH, Ali S. Antifungal activity of human cathelicidin LL-37, a membrane disrupting peptide, by triggering oxidative stress and cell cycle arrest in Candida auris. J Appl Microbiol 2022;8:204–21.

160. Olekson MA, You T, Savage PB, Leung KP. Antimicrobial ceragenins inhibit biofilms and affect mammalian cell viability and migration. in vitro. FEMS Open Bio 2017;7:953–67.

161. Durnas B, Wnorowska U, Pogoda K, Depta P, Watek M, Piktel E, et al. Candidacidal activity of selected ceragenins and human cathelicidin LL-37 in experimental settings mimicking infection sites. PLoS One 2016;11:e0157242.

162. Hashemi MM, Rovig J, Holden BS, Taylor MF, Weber S, Wilson J, et al. Ceragenins are active against drug-resistant Candida auris clinical isolates in planktomic and biofilm forms. J Antimicrob Chemother 2018;73:1537–45.