Ibrexafungerp is a novel triterpenoid antifungal that inhibits glucan synthase and thus fungal cell wall synthesis. We examined the in vitro activity against contemporary clinical yeast, investigated inter-laboratory and intra-laboratory variability, suggested wild-type upper-limit values (WT-UL), and compared in vitro activity of ibrexafungerp to five licensed antifungals. Susceptibility to ibrexafungerp and comparators was investigated prospectively for 1965 isolates (11,790 MICs) and repetitively for three QC strains (1764 MICs) following the EUCAST E.Def 7.3.2 method. Elevated ibrexafungerp/echinocandin MICs prompted FKS sequencing. Published ibrexafungerp EUCAST MIC-distributions were retrieved and aggregated for WT-UL determinations following EUCAST principles. Ibrexafungerp MICs were ≤2 mg/L except against C. parapsilosis, Cryptococcus and some rare yeasts. Modal MICs (mg/L) were 0.06/0.125/0.25/0.5/0.5/0.5/1/1/2 for C. albicans/C. dubliniensis/C. glabrata/C. krusei/C. parapsilosis/C. tropicalis/S. cerevisiae/S. guilliermondii/L. lusitaniae and aligned within ±1 dilution with published values. The MIC ranges for QC strains were: 0.06–0.25/0.5–1/0.125–0.5 for CNM-CL-F8555/ATCC6258/ATCC22019. The WT-UL (mg/L) were: 0.25/0.5/1/1/2 for C. albicans/C. glabrata/C. krusei/C. parapsilosis/C. tropicalis. Adopting these, non-wild-type rates were 0.3%/0.6%/0%/8%/3% for C. albicans/C. glabrata/C. krusei/C. parapsilosis/C. tropicalis and overall lower than for comparators except amphotericin B. Five/six non-wild-type C. albicans/C. glabrata were echinocandin and Fks non-wild-type (F641S, F659del or F659L). Eight C. parapsilosis and three C. tropicalis non-wild-type isolates were echinocandin and Fks wild-type. Partial inhibition near 50% in the supra-MIC range may explain variable MICs. Ibrexafungerp EUCAST MIC testing is robust, although the significance of paradoxical growth for some species requires further investigation. The spectrum is broad and will provide an oral option for the growing population with azole refractory infection.

Keywords: Candida; echinocandins; ECOFF; WT-UL

1. Introduction

Ibrexafungerp (formerly SCY-078) is a novel triterpenoid antifungal that interferes with the fungal cell wall synthesis through inhibition of glucan synthase [1]. It has fungicidal activity against Candida, including the often multidrug-resistant species Candida auris and fungistatic activity against Aspergillus [2–5]. Moreover, ibrexafungerp has activity against the ascus form of Pneumocystis jirovecii and has displayed activity in a murine prophylaxis model of pneumocystosis [6]. Ibrexafungerp is available in oral formulations and was approved in 2021 by the FDA for treatment of Candida vaginitis. A liposomal intravenous formulation is under development and ibrexafungerp is currently in clinical studies for treatment of refractory or intolerant fungal diseases, for C. auris infections, for treatment of invasive pulmonary aspergillosis in combination therapy with voriconazole, and for recurrent vulvovaginal candidiasis. The mode of action is comparable to that of the echinocandins. The binding site is different from, but overlapping with that of the
echinocandins leading to cross-resistance to some but not all FKS mutations in Candida spp. [3,7].

Clinical breakpoints have not yet been set for ibrexafungerp against Candida spp. An obligatory ingredient in breakpoint setting is analysis of MIC distributions from multiple sources and determination of epidemiological cut off values (ECOFF in EUCAST terminology and ECV in CLSI terminology). These are defined as the highest MIC value for isolates devoid of phenotypically detectable acquired resistance mechanisms, also called wild-type isolates [8]. The ECOFFs do not inform on clinical susceptibility because they only reflect the inherent susceptibility of the species, but they inform on the likelihood of presence of acquired resistance mechanisms that may or may not affect outcome depending on the drug exposure during therapy. Microbroth susceptibility testing is associated with technical variation related to differences between products from different manufacturers (such as brand of microtitre plates, medium and characteristics of the antifungal agent), method used for serial dilution, differences in how materials are made or handled, differences between how different individuals perform the same test, differences in cell density in the inoculum, differences in temperature stability of incubators, differences in atmospheres in incubators, etc. [8–11]. To encompass this variation, EUCAST requires at least 100 MICs from at least five independent sources, each consisting of at least 15 isolates and for which the modal MIC may not deviate more than one two-fold dilution from the most common mode for dataset used for EUCAST ECOFF setting [8]. So far, three studies have reported single or multicentre MICs [12–14]. The objectives of this study were to examine the in vitro activity of ibrexafungerp against a nationwide and representative sample of contemporary clinical yeast isolates of Candida, to compare the MICs obtained with those recently reported and set preliminary wild-type upper limits (WT-UL), and to compare the activity of ibrexafungerp to that of other agents licensed for the therapy of candidiasis.

2. Materials and Methods

Isolates. All unique yeast isolates received at the Statens Serum Institut (SSI) as pure cultures or cultured from clinical samples during 2020 and 2021 were included. Same patient same species isolates were regarded as unique if obtained >21 days apart or with a different susceptibility pattern.

Susceptibility testing and target gene sequencing. EUCAST E.Def 7.3.2 MIC determination was performed for ibrexafungerp and five comparators with Candida krusei ATCC 6258, Candida parapsilosis ATCC 22019 and Candida albicans CNM CL-F8555 as QC strains [15]. The MIC determination was performed prospectively during 2020–2021 using multiple batches of in-house prepared trays. Cell culture treated (Nunc™ MicroWell™ 96-Well Microplates, ThermoFisher Scientific cat. no. 167008/161093) were used throughout. Microtitre plates with 2-fold dilutions were prepared using serial dilution and two pipette tip changes and frozen at −80 °C for at least 24 h prior to use [10]. Ibrexafungerp (SCY-078, Scynexis Inc., Jersey City, NJ, USA) pure substance was stored in aliquots at −80 °C and stock solutions prepared in DMSO (Sigma-Aldrich, Brøndby, Denmark, 5000 mg/L). The final drug concentration range studied was 0.008–8 mg/L. The following comparator compounds were also investigated (source of compound and final concentration range in parentheses). Anidulafungin (Pfizer A/S, Ballerup, Denmark, 0.004–4 mg/L), micafungin (Astellas Pharma Inc., Tokyo, Japan until August 2021, then from Molcan corporation, Toronto, Canada, 0.004–4 mg/L), amphotericin B (Sigma-Aldrich, 0.004–4 mg/L), fluconazole (Sigma-Aldrich, either 0.06–64 mg/L), and voriconazole (Pfizer A/S, Ballerup, Denmark, 0.004–4 mg/L). The following quality control (QC) strains (number of repetitions) were included: C. albicans CNM-CL-F8555 (n = 79), C. krusei ATCC 6258 (n = 116) and C. parapsilosis ATCC 22019 (n = 99) as quality controls for the comparators and to generate QC MIC data for Ibrexafungerp (1764 MICs for QC strains in total).

FKS sequencing was performed for echinocandin-resistant isolates as previously described [16].
Data management. The MIC ranges, modal MIC (the most common MIC), and MIC$_{50}$ and MIC$_{90}$ (the MIC value that includes 50% and 90% of the isolates, respectively) values were determined for ibrexafungerp and comparators (anidulafungin, micafungin, amphotericin B, fluconazole and voriconazole). Published EUCAST MIC distributions were retrieved and inspected for agreement with data from this study according to the EUCAST SOP 10.2 [8]. Ibrexafungerp WT-UL values, defined as the upper MIC value where the wild-type distribution ends, were determined visually and statistically using the EUCAST ECOFFinder programme and inclusion of 97.5% to 99.9% of the predicted wild-type population for species-specific MIC distributions with ≥15 isolates [17]. However, as the values reported here are not formally accepted EUCAST ibrexafungerp ECOFFs, we used the term “WT-UL” to avoid confusion.

3. Results

Ibrexafungerp activity against clinical isolates. In total, 1965 yeast isolates (hereof 1893 Candida spp. isolates) were included and tested prospectively in parallel with repetitive testing of three quality control strains during 2020 and 2021 using multiple batches of in-house-prepared microdilution EUCAST plates. The number of clinical isolates per year was comparable with 967 isolates, including 540 from blood cultures, from 794 patients in 2020, and 1001 isolates, including 561 blood isolates, from 824 patients in 2021, respectively. The modal MICs were identical comparing MIC distributions from 2020 and 2021 for all species represented with at least 10 isolates (C. albicans, C. dubliniensis, C. glabrata, C. krusei, C. parapsilosis, C. tropicalis, C. guilliermondii, C. lusitaniae and S. cerevisiae) and hence the data from the two years were compiled (Table 1). As expected for Gaussian distributions, the modal MIC and MIC$_{50}$ values were identical for each species: C. albicans: 0.06 mg/L, C. dubliniensis 0.125 mg/L, C. glabrata 0.25 mg/L and for C. krusei, C. parapsilosis, C. tropicalis and S. cerevisiae all 0.5 mg/L. C. guilliermondii, C. lusitaniae and C. pararugosa were less susceptible (modal MIC/MIC$_{50}$ values of 1, 2 and 8 mg/L, respectively). Among the rare yeast, ibrexafungerp MICs ≤2 mg/L were found for Arxula adeninivorans (n = 1), Lodderomyces elongisporus (n = 1), Pichia manshurica (n = 1), Wickerhamomyces anomalus (Candida pelliculosa) (n = 2), Wickerhamomyces onychis (n = 1), Yarrowia lipolytica (Candida lusitaniae) (n = 1), Kodamaea ohmeri (n = 1) and Wickerhamomyces species (n = 1) whereas the majority of the MICs against the remaining rare yeast species were ≥4 mg/L (Supplementary Table S1).

Activity against quality control strains. The MIC ranges for ibrexafungerp against all three QC strains spanned two to three two-fold dilutions both years (Table 2). The ibrexafungerp modal MICs (mg/L) in 2020, 2021 and combined were: C. albicans CNM-CL F8555: 0.125; 0.125 and 0.125, C. krusei ATCC 6258: 0.5; 1 and 0.5 and C. parapsilosis ATCC 22019: 0.25; 0.25 and 0.25. In comparison, the MIC ranges for the comparators spanned two to four two-fold dilutions for amphotericin B, three to four dilutions for anidulafungin, two to four dilutions for micafungin, three dilutions for fluconazole and three to four dilutions for voriconazole. At least 95% of the MICs for comparators fell within the established MIC ranges for the three QC strains for amphotericin B, anidulafungin and voriconazole but fewer 79% for micafungin against C. krusei ATCC 6258 in 2020 and 31–46% for fluconazole against C. albicans CNM-CL F8555 in 2020 and 2021, specifically.

Inter-laboratory agreement for ibrexafungerp MIC testing. Next, the ibrexafungerp MIC distributions were compared to those from three recent publications (Table 3) [12–14]. All MIC distributions were unimodal with the exception of the bimodal MIC distribution for C. albicans (modes 0.03 and 0.125 mg/L) reported in the multicentre study by Quindos et al. [14], and which therefore was excluded from the WT-UL determination for the aggregated data. The species-specific modal MICs all fell within ±1 two-fold dilution from the most common species-specific modal MIC. The modal MICs for the aggregated dataset were for all species identical to the ones obtained in this study: C. albicans: 0.06 mg/L, C. glabrata: 0.25 mg/L, C. krusei: 0.5 mg/L, C. parapsilosis: 0.5 mg/L and C. tropicalis 0.5 mg/L.
Table 1. Ibrexafungerp MICs (mg/L) for 1965 yeast isolates (incl. 1893 Candida spp. isolates) and three other species isolates from Denmark collected in 2020 and 2021 combined.

| Species          | N  | MIC (mg/L)         | MIC<sub>50</sub> | MIC<sub>90</sub> | MIC Range | % NWT |
|------------------|----|--------------------|------------------|------------------|-----------|-------|
|                  |    | 0.016  | 0.03  | 0.06  | 0.125 | 0.25  | 0.5   | 1     | 2     | 4     | 8     | >8    |       |
| Candida          |    |         |       |       |       |       |       |       |       |       |       |       |       |
| C. albicans      | 896| 5       | 91    | 574   | 213   | 10    | 2<sup>+</sup> | 1     | 0.06   | 0.125 | 0.016–2 | 0.3   |
| C. dubliniensis  | 117| 15      | 64    | 33    | 4     | 1     | 0.06–2 | 0.9   |
| C. glabrata      | 475| 5       | 319   | 148   | 2<sup>+</sup> | 1<sup>+</sup> | 0.25 | 0.5   | 0.125–2 | 0.6   |
| C. kruzei        | 110| 14      | 74    | 22    | 1     | 0.5   | 1     | 0.25–1 | 0     |
| C. parapsilosis  | 78 | 1       | 23    | 44    | 2     | 1     | 6     | 1     | 0.5   | 2     | 0.125–8 | 10.3  |
| C. tropicalis    | 146| 8       | 44    | 75    | 16    | 2     | 1     | 0.5   | 1     | 0.125–8 | 2.1   |
| C. guilliermondii| 12 | 1       | 10    | 1     | 1     | 0.25  | NA    | 0.5–2 | NA    |
| C. kefyr         | 8  | 1       | 2     | 1     | 1     | 0.25  | NA    | 0.125–0.5 | NA    |
| C. lusitaniae    | 19 | 7       | 11    | 1     | 1     | NA    | 2     | 2     | 1–4   | 0     |
| Other Candida #  | 33 | 2       | 16    | 4     | 10    | 1     | NA    | NA    | 0.125–8 | NA    |
| Other yeast      |    |         |       |       |       |       |       |       |       |       |       |       |       |
| S. cerevisiae    | 30 | 1       | 10    | 19    | 5     | 2     | 2     | NA    | 2–4   | NA    |
| Cryptococcus spp.| 7 | 1       | 3     | 3     | 5     | 2     | 2     | NA    | NA    | 0.03–8 | NA    |
| Rare yeast *     | 34 | 1       | 3     | 3     | 2     | 3     | 13    | 6     | NA    | NA    | 0.125–2 | 11.2  |
| In total         | 1965 | 5       | 92    | 589   | 295   | 477   | 374   | 74    | 23    | 14    | 16    | 6     | NA    |

Modal MICs are underlined; non-wild-type (NWT) MIC defined as MICs above the consensus WT-UL (see WT-UL determination section below) are highlighted using bold font. * indicates ibrexafungerp non-wild-type isolates that harbour Fks amino acid (AA) alterations: C. albicans: both isolates F641S, C. glabrata: two isolates with F659del and one with F659L. # Other Candida included the following species with ibrexafungerp MIC in the 0.125–0.5 range: C. metaspilosis (one), C. orthopsilosis (eight), C. lampaica (one), C. norvegensis (six), C. pelliculosa (two), C. utilis (one); the following species with ibrexafungerp MIC in the 0.5–2 range: C. intermedia (one), C. stellimalicola (one), C. auris (two), C. fermentati (four), C. inconspicua (one), C. lipolytica (one), C. metbiosa (one), C. pulcherrina (one), C. sorbosorans (one); and finally one C. pararugosa with an MIC of 8 mg/L. * Rare yeast included the following species with ibrexafungerp MIC ≤ 0.5 range: Arxula adeninivorans (one), Lodderomyces elongisporus (one), Pichia manshurica (one), Wickerhamomyces anomalus (two), Wickerhamomyces onychis (one) and Yarrowia lipolytica (one); the following species with ibrexafungerp MIC in the 0.5–2 range: Wickerhamomyces species (one); and finally the following species that included isolates with the majority of MICs being 4 mg/L or greater: Magnusomyces capitatus (five), Trichosporon asahii (four), Geotrichum candidum (five), Rhodotorula mucilaginosa (three), Exophiala dermatitidis (one), Lachancea thermotolerans (one), Trichosporon dermatitis (two), Geotrichum capitatum (two), Geotrichum species (two) and Geotrichum silvicola (one). NA: Not applicable, MIC<sub>50</sub> values for species with ≥15 isolates. MIC<sub>90</sub> values for species with ≥15 isolates.
Table 2. MIC values for ibrexafungerp and comparators against three QC strains that were tested repetitively throughout the 2-year study period in parallel with the clinical isolates.

| Compound           | QC Strain, Year (N. of Repetitions) | MIC (mg/L) | % within Range |
|--------------------|------------------------------------|------------|----------------|
|                    |                                    | ≤0.004     | 0.008          | 0.016 | 0.03 | 0.06 | 0.125 | 0.25 | 0.5 | 1 | 2 | 4 | 8 | 16 | 32 | 64 |
| Ibexafungerp       | C. albicans CNM-CL F8555, 2020 (37) | 16         | 20             | 1     |      |      |      |      |      |    |    |    |    |    |    |    |
|                    | C. albicans CNM-CL F8555, 2021 (42)| 16         | 25             | 1     |      |      |      |      |      |    |    |    |    |    |    |    |
|                    | C. krusei ATCC 6258, 2020 (68)     |            |                | 43    | 25   |      |      |      |      |    |    |    |    |    |    |    |
|                    | C. krusei ATCC 6258, 2021 (48)     |            |                | 23    | 25   |      |      |      |      |    |    |    |    |    |    |    |
|                    | C. parapsilosis ATCC 22019, 2020 (55)| 4           | 32             | 19    |      |      |      |      |    |    |    |    |    |    |    |
|                    | C. parapsilosis ATCC 22019, 2021 (44)| 31         | 13             |      |      |      |      |      |    |    |    |    |    |    |    |
| Amphotericin B     | C. albicans CNM-CL F8555, 2020 (37)| 10         | 23             | 4     |      |      |      |      |    |    |    |    |    |    |    |
|                    | C. albicans CNM-CL F8555, 2021 (42)| 12         | 30             | 4     |      |      |      |      |    |    |    |    |    |    |    |
|                    | C. krusei ATCC 6258, 2020 (68)     | 0          | 36             | 32    |      |      |      |      |    |    |    |    |    |    |    |
|                    | C. krusei ATCC 6258, 2021 (48)     | 31         | 13             | 5     |      |      |      |      |    |    |    |    |    |    |    |
|                    | C. parapsilosis ATCC 22019, 2020 (55)| 3           | 16             | 33    | 3    |      |      |      |    |    |    |    |    |    |    |
|                    | C. parapsilosis ATCC 22019, 2021 (44)| 1           | 23             | 19    | 1    |      |      |      |    |    |    |    |    |    |    |
| Anidulafungin      | C. albicans CNM-CL F8555, 2020 (37)| 21         | 16             |      |      |      |      |      |    |    |    |    |    |    |    |
|                    | C. albicans CNM-CL F8555, 2021 (42)| 37         | 5              |      |      |      |      |      |    |    |    |    |    |    |    |
|                    | C. krusei ATCC 6258, 2020 (68)     | 6          | 57             | 5     |      |      |      |      |    |    |    |    |    |    |    |
|                    | C. krusei ATCC 6258, 2021 (48)     | 9          | 39             |      |      |      |      |      |    |    |    |    |    |    |    |
|                    | C. parapsilosis ATCC 22019, 2020 (55)| 1           | 5              | 42    | 7    |      |      |      |    |    |    |    |    |    |    |
|                    | C. parapsilosis ATCC 22019, 2021 (44)|            | 7              | 35    | 2    |      |      |      |    |    |    |    |    |    |    |
| Micafungin         | C. albicans CNM-CL F8555, 2020 (37)| 15         | 22             |      |      |      |      |      |    |    |    |    |    |    |    |
|                    | C. albicans CNM-CL F8555, 2021 (42)| 31         | 11             |      |      |      |      |      |    |    |    |    |    |    |    |
|                    | C. krusei ATCC 6258, 2020 (68)     | 0          | 54             | 14    |      |      |      |      |    |    |    |    |    |    |    |
|                    | C. krusei ATCC 6258, 2021 (48)     | 0          | 47             | 1     |      |      |      |      |    |    |    |    |    |    |    |
|                    | C. parapsilosis ATCC 22019, 2020 (55)|            | 1              | 6     | 43   | 5    |      |      |    |    |    |    |    |    |    |
|                    | C. parapsilosis ATCC 22019, 2021 (44)|            | 2              | 36    | 6    |      |      |      |    |    |    |    |    |    |    |
Table 2. Cont.

| Compound      | QC Strain, Year (N. of Repetitions) | MIC (mg/L) | % within Range |
|---------------|-------------------------------------|------------|----------------|
|               | ≤0.004 0.008 0.016 0.03 0.06 0.125 0.25 0.5 1 2 4 8 16 32 64 |            |                |
| **Fluconazole** | C. albicans CNM-CL F8555, 2020 (37) | 20 17 0 46%   |                |
|               | C. albicans CNM-CL F8555, 2021 (42) | 29 12 1 31%   |                |
|               | C. krusei ATCC 6258, 2020 (68)       | 5 39 4 100%  |                |
|               | C. krusei ATCC 6258, 2021 (48)       | 1 42 5 100%  |                |
|               | C. parapsilosis ATCC 22019, 2020 (55) | 14 38 3 95%  |                |
|               | C. parapsilosis ATCC 22019, 2021 (44) | 14 28 2 95%  |                |
| **Voriconazole** | C. albicans CNM-CL F8555, 2020 (37) | 2 33 2 95%   |                |
|               | C. albicans CNM-CL F8555, 2021 (42) | 4 36 1 90%   |                |
|               | C. krusei ATCC 6258, 2020 (68)       | 1 10 57 100% |                |
|               | C. krusei ATCC 6258, 2021 (48)       | 0 4 42 2 96% |                |
|               | C. parapsilosis ATCC 22019, 2020 (55) | 8 43 4 100% |                |
|               | C. parapsilosis ATCC 22019, 2021 (44) | 1 29 14 100% |                |

The concentration range tested varied by agent. Grey colour indicates concentrations not tested. The modal MICs for the MICs obtained in this study are highlighted in bold font. The recommended EUCAST MIC range and target MIC for the QC strains are indicated in green shading and underlined font, respectively, as summarised in “The European Committee on Antimicrobial Susceptibility Testing. Routine and extended internal quality control for MIC determination and agar dilution for yeasts, moulds and dermatophytes as recommended by EUCAST. Version 6.0, 2022. http://www.eucast.org”. 

Table 3. Ibrexafungerp MICs for the five most common Candida species isolates (3146 in total) from this study (n = 1705) and from three additional studies (two single-centre studies [12,13] and a multicentre study [14]). The Pfaller et al. study [13] included isolates selected to include wild-type and fluconazole and echinocandin-resistant strains, whereas the two other studies included unselected clinical isolates. The modal MIC is highlighted in underlined font. The consensus WT-UL is indicated by a dashed vertical line (Table 4).

| Species      | N   | 0.016 | 0.03 | 0.06 | 0.125 | 0.25 | 1   | 2   | 4   | 8   | >8  | nwt | %   | Reference |
|--------------|-----|-------|------|------|-------|------|-----|-----|-----|-----|-----|-----|-----|----------|
| C. albicans  | 896 | 5     | 91   | 574  | 213   | 10   | 2   | 1   |     |     |     | 3   | 0.3 | This study |
| C. albicans  | 462 | 5     | 101  | 245  | 107   | 3    | 1   |     |     |     |     | 1   | 0.2 | [12] |
| C. albicans  | 29  | 3     | 12   | 7    | 1     | 2    | 3   | 1   |     |     |     | 6   | 20.7 | [13] |
| C. albicans  | 163 | 7     | 50   | 50   | 6     | 2    |     |     |     |     |     | 2   | 1.2 | [14] |
| Aggregated excl. [14] | 1387 | 10 | 195 | 831 | 327 | 14 | 3 | 5 | 2 | 10 | 0.7 | |
| C. glabrata  | 475 |      | 5    | 319  | 148   | 2    | 1   |     |     |     |     | 3   | 0.6 | This study |
| C. glabrata  | 120 | 3     | 54   | 59   | 3     | 1    |     |     |     |     |     | 4   | 3.3 | [12] |
| C. glabrata  | 29  | 3     | 15   | 12   | 3     | 1    |     |     |     |     |     | 4   | 13.8 | [13] |
| C. glabrata  | 60  | 2     | 10   | 22   | 16    | 5    | 1   | 2   | 1   |     |     | 9   | 15.0 | [14] |
| Aggregated   | 684 | 2     | 18   | 410  | 235   | 13   | 4   | 2   | 1   |     |     | 20  | 2.9 | |
| C. krusei    | 110 |       | 14   | 74   | 22    |     |     |     |     |     |     | 0   | 0.0 | This study |
| C. krusei    | 24  | 2     | 10   | 10   | 2     |     |     |     |     |     |     | 2   | 8.3 | [12] |
| C. krusei    | 19  | 3     | 121  | 4    |     |     |     |     |     |     |     | 4   | 0.0 | [13] |
| C. krusei    | 29  | 1     | 2    | 10   | 16    |     |     |     |     |     |     | 0   | 0.0 | [14] |
| Aggregated   | 182 | 1     | 21   | 213  | 52    | 2    |     |     |     |     |     | 2   | 1.1 | |
| C. parapsilosis| 78   | 1   | 23   | 44   | 2     | 1    | 6   | 1   |     |     |     | 8   | 10.3 | This study |
| C. parapsilosis| 249 | 1  | 22   | 127  | 92    | 7    |     |     |     |     |     | 0   | 0.0  | [12] |
| C. parapsilosis| 15  | 1   | 11   | 3    |     |     |     |     |     |     |     | 0   | 0.0  | [13] |
| C. parapsilosis| 108 | 6 | 1   | 10   | 38    | 24   | 15  | 6   | 5   |     |     | 26  | 24.1 | [14] |
| Aggregated   | 450 | 6    | 2    | 27   | 171   | 177  | 33  | 16  | 12  | 6   |     | 34  | 7.6 | |
| C. tropicalis| 146 |     | 8    | 44   | 75    | 16   | 2   | 1   |     |     |     | 3   | 2.1 | This study |
| C. tropicalis| 73  | 1    | 3    | 36   | 24    | 8    | 1   |     |     |     |     | 1   | 1.4 | [12] |
| C. tropicalis| 21  | 1    | 3    | 10   | 3     | 2    | 2   |     |     |     |     | 0   | 0.0  | [13] |
| C. tropicalis| 40  | 2    | 3    | 9    | 12    | 8    | 5   |     |     |     |     | 1   | 1   | [14] |
| Aggregated   | 280 | 1   | 3    | 17   | 99    | 114  | 34  | 7   | 3   | 1   | 1   | 5   | 1.8 | |
| C. dubliniensis | 117 | 15 | 64   | 33   | 4     | 1    |     |     |     |     |     | 1   | 0.9 | This study |

nwt: non-wild-type.
Table 4. Statistical and visual wild-type upper limit values (WT-UL, mg/L) for the five most common Candida spp. determined using the aggregated distributions presented in Table 3, single-centre data for C. dubliniensis and 97.5–99.9% of the modelled MIC distributions.

| Species            | Statistical WT-UL Values | Visual WT-UL | Consensus WT-UL |
|--------------------|--------------------------|--------------|-----------------|
|                    | 97.5% | 99.0% | 99.5% | 99.9% |                 |              |                 |
| C. albicans        | 0.125 | 0.125 | 0.25  | 0.25  | 0.25            | 0.25         |
| C. glabrata        | 0.5   | 0.5   | 0.5   | 1     | 1               | 0.5          |
| C. krusei          | 1     | 1     | 1     | 1     | 1               | 1            |
| C. parapsilosis    | 1     | 2     | 2     | 2     | 2               | 2            |
| C. tropicalis      | 1     | 0.25  | 0.5   | 0.5   | 0.5             | 0.5          |
| C. dubliniensis    | 0.25  | 0.5   | 0.5   | 0.5   | 0.5             | 0.5          |

WT-UL determination and wild-type non-wild-type classification. WT-ULs were determined statistically including 97.5%, 99%, 99.5% and 99.9% of the modelled aggregated wild-type distributions, and visually. The best agreement between the statistical and visual values was found for the WT-UL including 99.5%, which was chosen for wild-type versus non-wild-type classification. A single-centre WT-UL was also determined for C. dubliniensis MICs from this study following the same criteria (Table 4).

Applying the consensus WT-UL to the Danish isolates, three C. albicans, one C. dubliniensis, three C. glabrata, eight C. parapsilosis and three C. tropicalis were classified as non-wild-type isolates (Tables 1 and 3). The MIC and Fks information for these isolates are detailed in Table 5, together with isolates that were found wild-type for ibrexafungerp but anidulafungin- and micafungin-resistant. Among C. albicans isolates with ibrexafungerp MIC 1–2 mg/L, two of three were resistant to anidulafungin and micafungin, and both harboured an F641S alteration in Fks1. The third isolate was anidulafungin, micafungin and FKS wild-type. Among C. albicans isolates with ibrexafungerp MIC 0.25 mg/L and wild-type to ibrexafungerp, two of ten were resistant to anidulafungin and micafungin. Both isolates were FKS-sequenced and harboured alterations known to confer echinocandin resistance (S645P and R1361H, respectively), whereas the remaining eight were anidulafungin and micafungin wild-type and not FKS-sequenced. For C. glabrata with ibrexafungerp MICs 1-2 mg/L, three of three isolates were resistant to anidulafungin or micafungin, and all three harboured alterations involving F659 in Fks2. Eleven C. glabrata isolates were ibrexafungerp wild-type (MIC 0.25–0.5 mg/L) but were resistant to anidulafungin or micafungin. Among these, two harboured an F659Y alteration (ibrexafungerp MIC 0.25 and 0.5 mg/L, respectively), eight isolates harboured alterations involving codons further downstream in Fks1 or Fks2 hot spot regions (Table 5) and one isolate had alteration (K1323E)17 amino acids prior to Fks1 hot spot two. For the remaining species with MICs above the WT-ULs, one of one C. dubliniensis, eight of eight C. parapsilosis and three of three C. tropicalis isolates were anidulafungin- and micafungin-susceptible. Seven of these were FKS-sequenced and confirmed without mutations in the echinocandin hotspots or regions upstream thereof (Table 5).
Table 5. MIC and Fks protein details for isolates with elevated MICs to either ibrexafungerp or echinocandin.

| Species          | MIC (mg/L) * | Fks Amino Acid Alterations |
|------------------|--------------|----------------------------|
|                  | IBX | ANF | MFG | AMB | FLZ | VRZ |                  |                  |
| C. albicans      | 2   | 0.008 | 0.03 | 0.125 | 0.125 | 0.004 | WT |                  |                  |
| C. albicans      | 1   | 0.06 | 0.25 | 0.25 | 0.5 | 0.016 | F641S |                  |                  |
| C. albicans      | 1   | 0.125 | 0.5 | 0.25 | 0.125 | 0.004 | F641S |                  |                  |
| C. albicans      | 0.25 | 0.25 | 1 | 0.125 | 0.125 | 0.008 | R1361H |                  |                  |
| C. albicans      | 0.25 | 0.008 | 0.03 | 0.25 | 0.25 | 0.008 |                  |                  |
| C. albicans      | 0.25 | 0.008 | 0.03 | 0.25 | 0.125 | ≤0.004 |                  |                  |
| C. albicans      | 0.25 | 0.004 | 0.016 | 0.25 | 0.25 | 0.008 |                  |                  |
| C. albicans      | 0.25 | 0.008 | 0.016 | 0.25 | 0.5 | 0.008 |                  |                  |
| C. albicans      | 0.25 | 0.25 | 2 | 0.25 | 0.25 | 0.008 |                  |                  |
| C. albicans      | 0.25 | 0.004 | 0.016 | 0.25 | 0.25 | 0.008 |                  |                  |
| C. albicans      | 0.25 | 0.008 | 0.125 | 0.125 | ≤0.004 |                  |                  |
| C. albicans      | 0.25 | ≤0.004 | 0.016 | 0.5 | 0.5 | 0.008 |                  |                  |
| C. albicans      | 0.125 | 0.03 | 0.06 | 0.25 | 0.25 | 0.008 | R1361H |                  |                  |
| C. albicans      | 0.06 | 0.06 | 0.125 | 0.25 | 0.25 | ≤0.004 | R1361S |                  |                  |
| C. albicans      | 0.06 | 0.125 | 0.25 | 0.25 | 0.25 | ≤0.004 | R1361S |                  |                  |
| C. albicans      | 0.06 | 0.016 | 0.125 | 0.25 | 0.125 | 0.004 | S645P |                  |                  |
| C. albicans      | 2   | 0.016 | 0.03 | 0.06 | 0.5 | 0.016 | WT |                  |                  |
| C. albicans      | 0.5 | 0.016 | 0.03 | 0.06 | 0.5 | 0.016 |                  |                  |
| C. albicans      | 0.5 | 0.03 | 0.06 | 0.25 | 0.5 | 0.016 |                  |                  |
| C. albicans      | 0.5 | 0.016 | 0.03 | 0.25 | 0.5 | 0.016 |                  |                  |
| C. albicans      | 0.5 | 0.008 | 0.016 | 0.06 | 0.25 | 0.016 |                  |                  |
| C. albicans      | 2   | 0.5 | 0.5 | 0.25 | 2 | 0.03 | F659del (Fks2) |                  |                  |
| C. albicans      | 1   | 0.03 | 0.06 | 0.25 | 2 | 0.06 | F659L (Fks2) |                  |                  |
| C. albicans      | 0.5 | 0.06 | 0.06 | 0.25 | 16 | 0.5 | S629P (Fks1) |                  |                  |
| C. albicans      | 0.5 | 0.06 | 0.06 | 0.125 | 4 | 0.06 | L662F (Fks2) |                  |                  |
| C. albicans      | 0.5 | 0.125 | 0.06 | 0.25 | 8 | 0.125 | F659Y (Fks2) |                  |                  |
| C. albicans      | 0.5 | 0.125 | 0.125 | 0.25 | 32 | 1 | P667I (Fks2) |                  |                  |
| C. albicans      | 0.5 | 1 | 1 | 0.25 | 4 | 0.125 | S663P (Fks2) |                  |                  |
| C. albicans      | 0.5 | 1 | 1 | 0.5 | >64 | 4 | S663P (Fks2) |                  |                  |
| C. albicans      | 0.5 | 0.125 | 0.125 | 0.5 | 64 | 2 | L662W (Fks2) |                  |                  |
| C. albicans      | 0.5 | 2 | 2 | 0.25 | 4 | 0.06 | S629P (Fks1)/L640X (Fks2) |                  |                  |
| C. albicans      | 0.25 | 0.06 | 0.25 | 0.25 | 4 | 0.125 | R665G (Fks2) |                  |                  |
| C. albicans      | 0.25 | 0.125 | 0.06 | 0.25 | 2 | 0.06 | F659Y (Fks2) |                  |                  |
| C. albicans      | 0.25 | 0.03 | 0.06 | 0.25 | 2 | 0.06 | K1323E 17 AA prior to Fks1 HS-2 |                  |                  |
| C. parapsilosis  | 8   | 2 | 4 | 1 | >16 | >4 | WT |                  |                  |
| C. parapsilosis  | 4   | 1 | 2 | 1 | 64 | 1 |                  |                  |
| C. parapsilosis  | 4   | 2 | 2 | 0.5 | 64 | 2 |                  |                  |
| C. parapsilosis  | 4   | 0.5 | 1 | 0.5 | 1 | 0.016 |                  |                  |
| C. parapsilosis  | 4   | 1 | 2 | 0.5 | 0.5 | 0.008 |                  |                  |
| C. parapsilosis  | 4   | 0.5 | 2 | 0.5 | 0.25 | 0.008 |                  |                  |
| C. parapsilosis  | 4   | 0.5 | 1 | 0.5 | 0.25 | 0.016 |                  |                  |
| C. parapsilosis  | 2   | 2 | 4 | 1 | 1 | 0.016 | WT |                  |                  |
| C. tropicalis    | 8   | 0.016 | 0.03 | 0.5 | 0.5 | 0.03 | WT |                  |                  |
| C. tropicalis    | 4   | 0.016 | 0.03 | 0.5 | 2 | 0.125 | WT |                  |                  |
| C. tropicalis    | 4   | 0.016 | 0.016 | 0.125 | 0.5 | 0.03 | WT |                  |                  |
| C. tropicalis    | 1   | 0.03 | 0.06 | 0.5 | 4 | 0.125 | WT |                  |                  |
| C. auris         | 1   | 2 | >4 | 1 | >64 | 1 | F635Y |                  |                  |

* MICs that are categorised as non-wild-type for ibrexafungerp with all four WT-UL values and for the echinocandins adopting the EUCAST ECOFFs are indicated in red. Blue is used for Fks alterations at the first codon in hot spot one of the target genes, which has been associated with ibrexafungerp cross-resistance. Empty fields indicate that FKS sequencing has not been done.
Two additional isolates were found non-wild-type to echinocandins. One was a C. krusei wild-type for ibrexafungerp but with a micafungin MIC of 2 mg/L and an S659P alteration. The other was a C. auris isolate with anidulafungin and micafungin resistance and an Fks1 F635Y alteration [18]. The ibrexafungerp MIC of this isolate was 1 mg/L, and thus only one two-fold dilution higher than the modal MIC determined in our study of ibrexafungerp EUCAST susceptibility of C. auris [3].

**Growth patterns for ibrexafungerp non-wild-type but echinocandin-susceptible isolates.** A total of 13 isolates (one C. albicans, one C. dubliniensis, eight C. parapsilosis and three C. tropicalis) were deemed ibrexafungerp non-wild-type but anidulafungin- and micafungin-susceptible and FKS wild-type if sequenced. The growth curves of these isolates were inspected and compared to those for selected wild-type isolates (Supplementary Figure S1a–h). The growth curves for repeated testing of the C. albicans isolate were characterised by complete inhibition in the presence of 8 and 4 mg/L ibrexafungerp, by partial inhibition close to 50% in the 2–0.25 mg/L concentration range, and by decreasing inhibition with decreasing concentrations thereafter (Figure S1a). This led to inconsistent MIC determination in three runs: 0.25 mg/L, 2 mg/L and 4 mg/L, respectively, depending on where the curve intersected the 50% endpoint in the range of partial inhibition. For comparison, the growth curves for five C. albicans isolates without partial inhibition but with differential susceptibility are shown in Figure S1b. The C. dubliniensis isolate displayed a steadily rising growth curve with decreasing concentrations on repeat testing (Figure S1c). Three randomly selected C. dubliniensis with MIC 0.125 mg/L are shown in Figure S1d and show a characteristic paradoxical growth in supra MIC concentrations 1–4 mg/L but that does not exceed the 50% cut-off line. Growth curves for the eight C. parapsilosis isolates with MICs 4–8 mg/L when applying a 50% endpoint are shown in Figure S1e and compared to repetitive testing of the C. parapsilosis QC strain ATCC 22019 in Figure S1f. Complete inhibition is seen in the presence of 8 mg/L, partial inhibition in the 0.5–4 mg/L range and loss of activity in the following decreasing concentrations. The level of growth in the partial inhibition area was variable on repeated testing and again led to random MICs depending on if the level exceeded the 50% cut-off line or not. Finally, the three C. tropicalis isolates were compared to three selected isolates where the growth curve intersected the 50% growth line several times complicating endpoint reading, and three randomly chosen isolates displaying paradoxical growth at supra MIC concentrations but not above the 50% (Figure S1g,h).

**Susceptibility to comparators.** Amphotericin B was the agent with the broadest activity, as resistance was only found against four Trichosporon spp. isolates out of the thirty-four rare yeast (Table 6). Fluconazole resistance was found among all Candida species except C. kefyr and C. lusitaniae and with acquired resistance ranging from 2.6% in C. albicans, C. dubliniensis and C. tropicalis to over 6.4% in C. parapsilosis to 10.1% in C. glabrata. A notable number of Candida isolates were resistant to voriconazole with rates varying from 0.9% in C. dubliniensis to 6.4 and 6.6% in C. parapsilosis and C. glabrata, respectively. Finally, acquired resistance to anidulafungin/micafungin was found in C. albicans 1.0/1.8%, C. glabrata 2.3/3.6%, C. krusei 0.9/0.9% and C. parapsilosis 0/2.6%.
Table 6. Susceptibility characteristics given as MIC$_{50}$ and MIC range (both mg/L) and percentage of isolates classified as I (susceptible, increased exposure) $^a$ and R (resistant) for 1965 Danish yeast isolates collected 2020–2021 to five licensed antifungal agents.

| Species (N) | Anidulafungin | Micafungin | Amphotericin B | Fluconazole | Voriconazole |
|-------------|---------------|------------|----------------|-------------|--------------|
|             | MIC$_{50}$    | MIC Range | %R/nwt         | MIC$_{50}$  | MIC Range    | %I | %R |
| Candida spp. |               |            |                |             |              |    |    |
| C. albicans (896) | $\leq0.004$ | $\leq0.004$–0.5 | 1.0 | 0.016 | $\leq0.004$–2 | 1.8 | 0.25 | 0.06–1 | 0 | 0.25 | $\leq0.06$–64 | 0.3 | 2.7 | 0.008 | $\leq0.004$–4 | 3.0 |
| C. dubliniensis (117) | 0.016 | $\leq0.004$–0.03 | 0 | 0.03 | 0.008–0.06 | 0 | 0.06 | 0.016–0.25 | 0 | 0.25 | 0.125–64 | 0.0 | 2.6 | 0.008 | $\leq0.004$–4 | 0.9 |
| C. glabrata (475) | 0.016 | 0.008–2 | 2.3 | 0.03 | $\leq0.004$–2 | 3.6 | 0.25 | 0.03–1 | 0 | 4 | 2–>64 | 89.9 | 10.1 | 0.06 | 0.016–4 | 6.7 |
| C. krusei (110) | 0.03 | 0.016–0.125 | 0.9 | 0.125 | 0.06–2 | 0.9 | 0.5 | 0.125–1 | 0 | 32 | 2–>64 | 100.0 | 0.5 | 0.125–4 | 4.5 |
| C. parapsilosis (78) | 1 | 0.25–2 | 0 | 1 | 0.5–4 | 2.6 | 0.5 | 0.125–1 | 0 | 1 | 0.25–4 | 6.4 | 6.4 | 0.016 | 0.008–4 | 6.4 |
| C. tropicalis (146) | 0.016 | $\leq0.004$–0.03 | 0 | 0.03 | 0.008–6 | 0 | 0.5 | 0.125–1 | 0 | 0.5 | 0.125–32 | 5.5 | 2.7 | 0.03 | 0.008–1 | 5.5 |
| C. guilliermondii (12) | 0.5 | 0.25–1 | NA | 0.25 | 0.25–0.5 | NA | 0.125 | 0.125–0.25 | 0 | 8 | 2–64 | 18.2 | 72.7 | 0.25 | 0.06–4 | NA |
| C. kefyr (8) | 0.03 | 0.016–0.125 | NA | 0.06 | 0.06–0.25 | NA | 0.125 | 0.06–0.25 | 0 | 0.5 | 0.125–1 | 0.0 | 0.0 | 0.008 | 0.004–0.016 | NA |
| C. lusitaniae (19) | 0.03 | 0.016–0.06 | NA | 0.06 | 0.06–0.125 | NA | 0.125 | 0.06–0.25 | 0 | 0.5 | 0.125–1 | 0 | 0 | 0.008 | 0.004–0.016 | NA |
| Other spp. (34) | 0.125 | 0.008–4 | NA | 0.125 | 0.03–0.5 | NA | 0.25 | 0.06–0.5 | 0 | 8 | 2–32 | 76.7 | 125 | 125 | 0.06–1 | NA |
| Other yeast | S. cerevisiae (30) | 0.06 | 0.03–0.125 | NA | 0.25 | 0.125–0.25 | NA | 0.25 | 0.06–0.5 | 0 | 8 | 2–32 | 76.7 | 125 | 0.06–1 | NA |
| Cryptococcus spp. (7) | >4 | >4 | NA | >4 | >4 | NA | 0.25 | 0.25–1 | 0 | 4 | 2–16 | NA | NA | 0.125 | 0.03–0.25 | 0 |
| Rare yeast (34) | >4 | 0.008–>4 | NA | >4 | 0.03–>4 | NA | 0.5 | 0.125–4 | 11.8 | 16 | 0.25–64 | NA | NA | 0.25 | $\leq0.004$–4 | NA |

$^a$ No isolates are classified as I to anidulafungin, micafungin, amphotericin B or voriconazole; hence, columns for the I-category are omitted for these agents. $^b$ NA: not applicable (EUCAST breakpoints or ECOFFs are not established). nwt: non-wild-type.
4. Discussion

The main findings in this study were (1) that ibrexafungerp susceptibility testing during 2 years of routine testing conditions was robust, and that inter-laboratory agreement was high when compared to published data, (2) that WT-ULs could be set and adopted for classification into wild-type and non-wild-type phenotypes until formal EUCAST ECOFFs and clinical breakpoints have been established, (3) that ibrexafungerp displays broad activity against all the included Candida spp. except C. pararugosa, (4) that ibrexafungerp retains activity against most FKS mutant isolates found in our laboratory during 2020–2021, and (5) that only amphotericin B displayed a lower resistance rate in agreement with the almost universal activity of this agent. However, the study also identified some technical challenges that may lead to random classification of isolates, particularly for C. dubliniensis, C. parapsilosis and C. tropicalis due to partial growth inhibition in a supra-MIC range for EUCAST testing.

The ibrexafungerp MIC testing was robust as documented by narrow and comparable MIC distributions for QC strains and identical modal MICs for clinical isolates across the two years. This suggests a stable and robust intra-laboratory performance of the EUCAST method. The modal MICs obtained in this study were identical to or one dilution from the modal MIC both for the two QC strains also repeatedly tested by Mesquida et al. [12], and for the common Candida species included in recent single and multicentre studies of ibrexafungerp EUCAST susceptibility [12–14]. These observations suggest a good inter-laboratory agreement. Of note however, a bimodal MIC distribution was noted in the multicentre study for C. albicans specifically, with a first peak at 0.03 mg/L and another at 0.125 mg/L suggesting that some optimisation may be warranted, which will be facilitated once QC MIC target and ranges have been established [14].

The activity of ibrexafungerp was broad and more uniform than for echinocandins against Candida spp. with C. pararugosa being the only clearly ibrexafungerp-resistant Candida species in our study. The activity included the rarer species S. cerevisiae, A. adeninivorans, L. elongisporus, P. manshurica, W. anomalus (C. pelliculosa), W. onychis and Y. lipolytica (C. lipolytica), which were susceptible at concentrations up to 2 mg/L. The inherent echinocandin susceptibility of C. parapsilosis is notably different compared to that for other Candida species (e.g., eight two-fold dilutions for anidulafungin MICs in this study). In contrast, the ibrexafungerp MICs are more discretely elevated for C. parapsilosis compared to C. albicans (three dilutions) and comparable to those against C. tropicalis and C. krusei. C. parapsilosis harbours an intrinsic amino acid substitution, alanine A660, at the last position of the hot spot region in Fks1 or Fks2 (C. albicans F641S (n = 2), C. glabrata F659del (n = 2) and F659L (n = 1)), previously associated with ibrexafungerp echinocandin cross-resistance [7,20]. However, we also found two C. glabrata and one C. auris with F659Y and F635Y alterations, respectively, which were echinocandin-resistant but ibrexafungerp wild-type. Pfaller et al. found elevated ibrexafungerp MICs for C. albicans isolates harbouring F641I, F641Y and F641S in Fks1, C. glabrata F659del and F659S in Fks2 and in 2/3 C. tropicalis isolates harbouring the F641S alteration in Fks1 [13]. Mesquida et al. found three isolates with non-wild-type MICs of which the C. glabrata isolates had a F659S alteration but the C. albicans and C. tropicalis isolates did not harbour Fks alterations among their blood stream isolates [12,20]. These data support that alterations at the phenylalanine (F) codon can cause echinocandin and/or ibrexafungerp resistance but not universally and that some resistant isolates do not harbour Fks alterations. Obviously, this will complicate a translation of molecular data to phenotypic susceptibility pattern. None of our 12 C. dublin-
iensis, C. parapsilosis or C. tropicalis isolates with elevated ibrexafungerp MICs harboured Fks alterations or displayed resistance to the echinocandins. For a number of these, we believe that a paradoxical growth in the 0.5 to 4 mg/L range led to a random wild-type versus non-wild-type classification depending on if the growth curve intersected the 50% inhibition line or not. Of note, this was not observed for the highest concentration tested (8 mg/L), which is somewhat in contrast to the paradoxical effect described for echinocandins [21]. We also hypothesise that this is not only an issue in our laboratory, as wide MIC distributions for C. parapsilosis and C. tropicalis were also reported by Quindos et al. [14]. Obviously, further studies are warranted to explore whether this is solely a technical issue or has potential clinical aspects.

As expected for a new drug class, non-wild-type isolates were less common than for echinocandins and azoles. This is indeed promising, with currently no oral alternatives forazole-resistant infections, and at a time where fluconazole resistance is emerging in both C. glabrata and C. parapsilosis [22–25].

5. Conclusions

Ibrexafungerp displayed broad activity against Danish isolates including most FKS mutant Candida isolates and species with inherent or acquired resistance to fluconazole. This is a promising prospect for many patients for whom we today have no oral options. WT-ULs were set and MICs for QC strains presented, which will allow for comparison and interpretation until formal values have been set by EUCAST. A partial inhibition pattern was observed against some isolates, particularly those of C. parapsilosis but also C. dubliniensis and C. tropicalis, which complicated MIC determination. Whether this reflects different clinical susceptibility or a technical issue related to in vitro testing warrants further investigation.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/jof8101106/s1, Figure S1: Growth curve examples for isolates defined as ibrexafungerp wild-type and non-wild-type adopting the WT-UL 97.5%, Table S1: Ibrexafungerp EUCAST MICs of rare yeast.

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