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# *In Vitro* Activity of Manogepix (APX001A) and Comparators against Contemporary Molds: MEC Comparison and Preliminary Experience with Colorimetric MIC Determination

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**ABSTRACT** Manogepix (APX001A) is the active moiety of the drug candidate fosmanogepix (APX001), currently in clinical development for the treatment of invasive fungal infections. We compared manogepix EUCAST minimum effective concentrations (MECs) to MICs of five comparators and CLSI MECs and MICs by a colorimetric method against contemporary molds. EUCAST susceptibility testing was performed for 161 isolates. Interlaboratory and intermethod reproducibility were determined by comparison with published manogepix MECs. Colorimetric MICs (measuring metabolic activity) were evaluated using three *Aspergillus fumigatus* isolates and one *Aspergillus flavus* isolate with four inocula at 24 to 48 h of incubation and 1 to 3 h 2,3-bis-(2-methoxy-4-nitro-5-sulfophenyl)-2H-tetrazolium-5-carboxanilide salt (XTT)/menadione (MEN) exposure. Manogepix modal MECs (range in mg/liter) against *Aspergillus* species were 0.03 to 0.06 (0.008 to 0.125) and unaffected by itraconazole resistance. Manogepix was as active against two *Fusarium* isolates but inactive against *Trichophyton interdigitale*, *Lichtheimia ramosa*, and *Rhizomucor pusillus* isolates (MECs >0.5). Modal MEC/MICs were ≥3 2-fold dilutions apart without overlapping ranges comparing manogepix with amphotericin B, isavuconazole, and voriconazole against *Aspergillus* isolates. Manogepix and posaconazole MECs/MICs correlated for *Aspergillus niger* (Pearson's $r = 0.711$; $P = 0.0044$). The MEC at which 50% of the isolates tested are inhibited ($MEC_{50}$), mode, and $MEC_{90}$ values were within ±1 dilution in all cases compared with published EUCAST and CLSI data. The colorimetric method showed excellent agreement with the MECs when plates were inoculated with the lowest inoculum ($1 \times 10^2$ CFU/ml to $2.5 \times 10^2$ CFU/ml), incubated for 24 h, and exposed for 1 to 3 h to XTT/MEN. Broad-spectrum *in vitro* activity of manogepix against clinically relevant molds was confirmed with excellent agreement across EUCAST and CLSI methods reported from experienced mycology laboratories. Colorimetric MIC determination warrants further investigation as a potential alternative that is less dependent on mycology expertise.

**KEYWORDS** APX001, APX001A, *Aspergillus*, CLSI, EUCAST, antifungal susceptibility testing, fosmanogepix, manogepix, mold

Manogepix (formerly APX001A) is the active moiety of the first-in-class small-molecule methyl-phosphate prodrug fosmanogepix (formerly APX001). By targeting the conserved fungal inositol acyltransferase enzyme Gwt1, manogepix prevents glycosylphosphatidylinositol (GPI)-anchored protein maturation, thereby compromising fungal growth (1). Manogepix has broad-spectrum activity that includes *Aspergillus* and rare molds (2–8). Fosmanogepix has demonstrated promising efficacy in several animal models of invasive pulmonary aspergillosis and scedosporiosis, as well as

**Citation** Jørgensen KM, Astvad KMT, Arendrup MC. 2020. *In vitro* activity of manogepix (APX001A) and comparators against contemporary molds: MEC comparison and preliminary experience with colorimetric MIC determination. Antimicrob Agents Chemother 64:e00730-20. https://doi.org/10.1128/AAC.00730-20.



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**Received** 15 April 2020
**Returned for modification** 20 May 2020
**Accepted** 2 June 2020
**Accepted manuscript posted online** 8 June 2020
**Published** 22 July 2020

disseminated fusariosis (9–12). Fosmanogepix is currently in phase 2 clinical trials for invasive aspergillosis.

Previous studies have shown that manogepix *in vitro* susceptibility testing is similar to that of the echinocandins, which utilizes MIC endpoints for yeast and minimum effective concentration (MEC) endpoints for molds (1, 6, 7). The *in vitro* activity of manogepix against molds using the European Committee on Antimicrobial Susceptibility Testing (EUCAST) E.Def 9.3.1 method was evaluated in 3 studies investigating *in vitro* activity against up to 23 isolates of each of the 4 most prevalent *Aspergillus* species (6), *Fusarium* and *Scedosporium* (7, 8), and rare molds (8), respectively, in comparison with the CLSI method. Two additional studies reported the *in vitro* activity against *Aspergillus*, *Fusarium*, and *Scedosporium* with the CLSI method (1, 4). MEC determination is subjective, requires expertise, and is not easily implemented in a routine laboratory of clinical microbiology. An alternative automated spectrophotometer colorimetric method was recently found superior for susceptibility testing of echinocandins against *Aspergillus* (13). This method evaluates fungal metabolic activity after the addition of XTT/MEN to the incubated microtiter plate.

Here, we present EUCAST manogepix MECs and comparator MICs for 161 prospective *Aspergillus* isolates and some other clinical mold isolates received for identification and EUCAST susceptibility testing and compared to published data for the EUCAST and CLSI methods. In addition, we present preliminary results for the determination of manogepix susceptibility using a colorimetric assay for metabolic activity.

## RESULTS

**Manogepix *in vitro* activity by EUCAST and CLSI.** For the *Aspergillus* species, the modal (range) MECs (mg/liter) were 0.03 to 0.06 (0.008 to 0.125), with *Aspergillus niger* and *Aspergillus nidulans* being 1 2-fold dilution more susceptible than *Aspergillus fumigatus* (Table 1). Ten *A. fumigatus* isolates were itraconazole resistant and harbored the following Cyp51A alterations: P216L ($n = 3$), G54E ($n = 1$), G54R ($n = 2$), G54A ($n = 1$), G432S ($n = 1$), $TR_{34}$/L98H ($n = 1$), and $TR_{34}^{3}$/L98H ($n = 1$). Manogepix was equally active against itraconazole-resistant (geometric mean MEC [GM-MEC], 0.056; range, 0.03 to 0.125 mg/liter) and -susceptible *A. fumigatus* (GM-MEC, 0.053; range, 0.016 to 0.125). For the other molds, manogepix was as active against the two *Fusarium* isolates (*Fusarium dimerum* and *Fusarium solani* MECs, 0.06 and 0.03 mg/liter, respectively) as against *A. fumigatus*, whereas no activity was seen for *Trichophyton interdigitale*, *Lichtheimia ramosa*, and *Rhizomucor pusillus* within the concentration range tested (MECs $>0.5$ mg/liter).

Manogepix was more active against *Aspergillus* isolates than amphotericin B, isavuconazole, and voriconazole on a milligrams per liter basis, with modal MECs/MICs $\geq 3$ dilution steps apart or no overlapping ranges (Table S1 in the supplemental material). Manogepix was also more effective than any of the comparators for the two *Fusarium* isolates. A correlation was seen between manogepix MECs and posaconazole MICs for *A. niger* (Pearson's $r = 0.711$; $P = 0.0044$; 14/18 isolates included in the calculations) but not for any other manogepix drug combinations.

EUCAST MECs obtained in this study were systematically $\sim$1 dilution higher than those previously reported against *Aspergillus* and *Fusarium* with the CLSI method (Table 1). However, the differences were minor, with the MEC at which 50% of the isolates tested are effective ($MEC_{50}$), modal MEC, and $MEC_{90}$ values within $\pm 1$ dilution in all cases across EUCAST and CLSI reference methods.

**Colorimetric MIC determination.** Inhibition of the metabolic activity was investigated using the standard EUCAST inoculum as well as three 10-fold dilutions hereof (corresponding to $1 \times 10^5$ to $2.5 \times 10^5$ to $1 \times 10^2$ to $2.5 \times 10^2$ CFU/ml; CLSI recommends $0.2 \times 10^4$ to $2.5 \times 10^4$) after both 24 h and 48 h of incubation and after another 1, 2, and 3 h of incubation with 2,3-bis-(2-methoxy-4-nitro-5-sulfophenyl)-2H-tetrazolium-5-carboxanilide salt (XTT)/menadione (MEN) (Fig. S1 to S3). For *A. fumigatus*, the lowest inoculum (corresponding to $1 \times 10^2$ to $2.5 \times 10^2$ CFU/ml or 20 to 50 CFU/well) resulted in complete inhibition of metabolic activity in the highest concen-

**TABLE 1** Summary of EUCAST and CLSI MECs for clinical mold isolates in this and previous studies[a]

| Species | Method | No. of complexes | $MEC_{50}$ (mg/liter) | Mode (mg/liter) | $MEC_{90}$ (mg/liter) | Range (mg/liter) | Source or reference no. |
|---|---|---|---|---|---|---|---|
| *A. flavus* | EUCAST | 8[b] | (0.03) | (0.03) | (0.06) | 0.016 to 0.06 | This study |
| | EUCAST | 20 | 0.03 | 0.03 | 0.06 | 0.016 to 0.06 | 6 |
| | CLSI | 20 | 0.016 | 0.016 | 0.03 | ≤0.008 to 0.06 | 6 |
| | CLSI | 18 | 0.016 | 0.03 | 0.03 | ≤0.008 to 0.06 | 4 |
| | CLSI | 4 | (0.016) | (0.016) | (0.125) | 0.016 to 0.125 | 1 |
| *A. fumigatus* | EUCAST | 121 | 0.06 | 0.06 | 0.125 | 0.016 to 0.125 | This study |
| | EUCAST | 22 | 0.03 | 0.03 | 0.06 | 0.016 to 0.06 | 6 |
| | CLSI | 22 | 0.03 | 0.03 | 0.06 | ≤0.008 to 0.06 | 6 |
| | CLSI | 182 | 0.016 | 0.016 | 0.03 | ≤0.008 to 0.06 | 4 |
| | CLSI | 20 | 0.06 | | 0.125 | 0.03 to 0.125 | 1 |
| *A. nidulans* | EUCAST | 2[c] | | 0.03 | | | This study |
| *A. niger* | EUCAST | 18[d] | 0.03 | 0.03 | 0.06 | 0.008 to 0.06 | This study |
| | EUCAST | 13 | 0.03 | 0.03 | 0.06 | 0.016 to 0.06 | 6 |
| | CLSI | 13 | ≤0.008 | ≤0.008 | 0.016 | ≤0.008 to 0.016 | 6 |
| | CLSI | 23 | ≤0.008 | ≤0.008 | 0.016 | ≤0.008 to 0.03 | 4 |
| | CLSI | 2 | | | | 0.016 to 0.03 | 1 |
| *Aspergillus terreus* | EUCAST | 6[e] | (0.06) | (0.06) | (0.125) | 0.016 to 0.125 | This study |
| | EUCAST | 23 | 0.03 | 0.03 | 0.06 | 0.016 to 0.06 | 6 |
| | CLSI | 23 | 0.016 | 0.016 | 0.03 | ≤0.008 to 0.06 | 6 |
| | CLSI | 10 | 0.016 | 0.016 | 0.03 | ≤0.008 to 0.03 | 4 |
| | CLSI | 1 | | | | 0.125 | 1 |
| *F. dimerum* | EUCAST | 1 | | | | 0.06 | This study |
| *F. solani* | EUCAST | 1 | | | | 0.03 | This study |
| | EUCAST | 15 | 0.125 | 0.125 | 0.125 | 0.03 to 0.125 | 7 |
| | CLSI | 15 | 0.03 | 0.03 | 0.06 | 0.016 to 0.06 | 7 |
| | CLSI | 7 | (0.03) | (0.03) | (>32) | 0.016 to >32 | 1 |
| *Fusarium oxysporum* | EUCAST | 15 | 0.06 | 0.06 | 0.125 | 0.03 to 0.25 | 7 |
| | EUCAST | 10 | 0.25 | | 16 | 0.016 to 16 | 8 |
| | CLSI | 15 | 0.06 | 0.06 | 0.25 | 0.016 to 0.5 | 7 |
| | CLSI | 10 | 0.016 | | 16 | 0.016 to 16 | 8 |

[a]$MEC_{50}$, mode, and $MEC_{90}$ are presented in parentheses when set for less than 10 isolates.
[b]Three *A. flavus sensu stricto* and five *A. flavus* complexes.
[c]One *A. nidulans* complex and one *Aspergillus spinulosporus* (both with a manogepix MEC of 0.03 mg/liter).
[d]Fourteen *A. niger*, three *A. tubingensis*, and one *Aspergillus luchuensis* complex.
[e]Two *A. terreus sensu stricto* and four *A. terreus* complexes.

trations. Decreasing and partial inhibition were observed with increasing inoculum concentrations, increasing incubation time, and time of XTT/MEN exposure (Fig. S1 to S3). With the undiluted EUCAST inoculum, no inhibition was observed for the three *A. fumigatus* isolates included after 24 h of incubation independent of the duration of XTT/MEN incubation. The same was true if allowing 48 h of incubation before XTT/MEN addition, although the curves were somewhat U-shaped with no inhibition at high and low concentrations and modest inhibition observed in the 0.008 to 0.03 mg/liter range, most pronounced after 1 h XTT/MEN exposure. Overall, the best agreement with EUCAST MECs was achieved with the 1:1,000 inoculum dilution and 24 h of incubation of the microtiter plate followed by 1 or 2 h of incubation with XTT/MEN. For the three *A. fumigatus* isolates, this allowed a full-inhibition MIC spectrophotometric endpoint for the lowest inoculum, comparable to the microscopically determined MEC within one dilution (Table 2).

For *A. flavus*, the inhibition pattern was somewhat different. A gradual decrease in inhibition of the metabolic activity was observed in the entire manogepix concentration range tested, which complicated the endpoint definition. Application of a 50% inhibition endpoint after 24 h of incubation resulted in MICs ranging from ≤0.002 to

**TABLE 2** Colorimetric manogepix MICs (mg/liter) and EUCAST MEC against three *A. fumigatus* and one *A. flavus* isolate[a,b]

| Species and strain | EUCAST MEC (mg/liter) | MIC (mg/liter) after 24 h incubation with exposure to XTT/MEN for: | | | MIC (mg/liter) after 48 h incubation with exposure to XTT/MEN for: | | |
|---|---|---|---|---|---|---|---|
| | | 1 h | 2 h | 3 h | 1 h | 2 h | 3 h |
| *A. fumigatus* | | | | | | | |
| SSI-7706 | 0.06 | 0.06 | 0.06 | 0.06 | 0.125 | 0.125 | 0.125 |
| SSI-7707 | 0.06 | 0.03 | 0.03 | 0.06 | 0.06 | 0.06 | 0.06 |
| SSI-7709 | 0.06 | 0.03 | 0.03 | 0.03 | NA[c] | NA | 0.125 |
| *A. flavus* | | | | | | | |
| SSI-7908[d] | 0.03 | ≤0.002 | ≤0.002 | 0.002 | 0.03 | 0.016 | 0.016 |

[a]Determined with a 1:1,000 diluted EUCAST standard inoculum.
[b]MICs were read as the last dilution on the visually horizontal part of the spectrophotometer curve (representing a complete inhibition endpoint but ignoring potential trailing growth) for *A. fumigatus* (Fig. S1 and S3). For *A. flavus*, a 50% inhibition MIC endpoint was adopted. MECs (mg/liter) obtained by the EUCAST E.Def 9.3.1 method for these isolates are included for comparison.
[c]NA, not applicable. A complete inhibition endpoint could not be determined due to a slight but continued increase in absorbance that prohibited definition of a reproducible MIC determination.
[d]Fifty percent inhibition endpoint.

0.002 mg/liter with increasing XTT/MEN incubation and from 0.03 to 0.016 mg/liter after 48 h of incubation, compared to the EUCAST MEC of 0.03 mg/liter.

## DISCUSSION

The EUCAST MECs obtained in this study against contemporary mold isolates confirmed the uniform *in vitro* activity of manogepix with low MECs across *Aspergillus* and *Fusarium*. We also demonstrated an excellent interlaboratory and intermethod agreement comparing the data in this study and the data from five prior reports (1, 4, 6–8). In all cases, EUCAST MECs were ~1 2-fold dilution elevated compared to those obtained with CLSI. However, $MEC_{50}$, modal MECs, and $MEC_{90}$ values were within ±1 dilution, thus meeting the stringent EUCAST criteria for accepting MIC/MEC data set for aggregation during the process for breakpoint setting outlined in EUCAST standard operating procedure (SOP) 10.1 (http://www.eucast.org/documents/sops/). Manogepix MECs were lower than the MICs found for the comparators, although MECs and MICs may not be directly comparable, and manogepix maintained activity against itraconazole-resistant *A. fumigatus*.

For *Candida*, a correlation between manogepix and fluconazole susceptibility has been observed at the species level and for some isolates with acquired fluconazole resistance (3). The underlying mechanism has been ascribed to efflux pumps in two spontaneous laboratory mutants (14), although the clinical relevance of the correlation is still unknown. In *A. fumigatus*, the most common azole resistance mechanism is target gene mutations, and indeed, all 10 itraconazole-resistant isolates in this study harbored target gene mutations, which may explain why manogepix activity was unaffected. A correlation was observed between manogepix and posaconazole for *A. niger*. However, none of the manogepix MECs were above the modal MEC plus 1 2-fold dilution against *A. niger*, and all *A. niger* were wild type for posaconazole (epidemiological cutoff [ECOFF], 0.5 mg/liter). These data suggest this correlation is either an artifact due to the low number of isolates or related to differential inoculum sizes or growth rates among the isolates and thus would have little clinical impact.

MEC determination requires microscopic identification of the well containing the lowest concentration that confers aberrant hyphal growth. This method is subjective, requires expertise, is impracticable in a routine laboratory of clinical microbiology, and has been associated with failure to correctly identify clinically relevant echinocandin-resistant *A. fumigatus* (15). The colorimetric method investigated here has recently proven superior for susceptibility testing of echinocandins against *Aspergillus* (13). For *A. fumigatus*, our pilot study suggested that manogepix MIC determination was feasible already after 24 h when plates were inoculated with the lowest inoculum tested ($1 \times 10^2$ to $2.5 \times 10^2$) before addition of XTT/MEN. The obtained MICs were stable within a 1 to 3 h of incubation with XTT/MEN and in agreement with the corresponding

reference MEC. For *A. flavus*, the endpoint reading was challenged by a slow but gradually increasing color development over a wide concentration range that complicated endpoint definition. However, if adopting a 50% inhibition endpoint after 48 h of incubation before XTT/MEN addition, MICs were again in agreement with the standard MECs.

This study has strengths and limitations. The MEC readings of contemporary molds were done over a 12-month period as part of the clinical routine. This suggests a robust performance in a reference laboratory. Another strength is that contemporary clinical isolates were included, including isolates with acquired resistance, and thus our findings suggest a broad "real-life" susceptibility. Limitations are that *A. fumigatus* formed the majority of the clinical molds received, and thus, numbers were limited for several other *Aspergillus* species as well as for other molds. Moreover, not all *Aspergillus* species were tested in the pilot study of the XTT/MEN method, something that needs to be done before considering evaluation of this method as an alternative in a larger multicenter study. Finally, as no isolates with confirmed acquired resistance to manogepix were available, the performance with respect to correct identification of such isolates remains to be investigated.

In conclusion, our study confirmed the broad-spectrum and uniform *in vitro* activity of manogepix against most clinically relevant molds, including itraconazole-resistant strains. An excellent agreement of MEC endpoints across EUCAST and CLSI methods was observed when conducted by experienced mycology laboratories. Moreover, a pilot study suggested that an automated colorimetric MIC determination may warrant further examination as a potential alternative objective method less dependent on mycology expertise.

## MATERIALS AND METHODS

**Isolates.** A total of 161 clinical mold isolates from 149 patients obtained from August 2016 to September 2017 were included. The same species isolates from the same patient were excluded from the study if sampled ≤21 days apart and identical MICs (within ±1 dilution step) for comparators were seen. The majority of isolates were derived from airways ($n = 129$) or eye/ear ($n = 20$). Identification was done by classical techniques, including thermotolerance (incubation at 50°C) for discriminating *A. fumigatus sensu stricto* from cryptic species, which underwent $\beta$-tubulin sequencing (16). The use of the term "complex" is acknowledged for *Aspergillus* species other than *A. fumigatus* in the absence of detailed molecular identification, although for simplicity, it is not used throughout the manuscript. Internal transcribed spacer (ITS) and translation elongation factor (TEF) sequencing were adopted for other molds and *Fusarium* species specifically (17, 18).

**Susceptibility testing.** *A. fumigatus* isolates were screened for azole resistance according to EUCAST E.Def 10.1 using a four-well screening plate (Balis Laboratorium VOF, Boven-Leeuwen, the Netherlands) (19). EUCAST E.Def 9.3.1 susceptibility testing was performed for all isolates for amphotericin B (Sigma-Aldrich) and manogepix (Amplyx Pharmaceuticals, San Diego, CA, USA) and posaconazole, voriconazole, itraconazole (all from Sigma-Aldrich) and isavuconazole (Basilea Pharmaceutica) for non-*A. fumigatus* molds and on nonsensitive and a selection of sensitive *A. fumigatus* isolates ($n = 35$) (20). Filtration (11-nm filter) of the inoculum was standard. *A. flavus* ATCC 204304 and *A. fumigatus* ATCC 204305 were used as controls. *CYP51A* sequencing was performed for non-wild-type/resistant *A. fumigatus* and *Aspergillus terreus* isolates (21).

**Colorimetric microdilution MIC determination.** Three WT *A. fumigatus sensu stricto* and one WT *A. flavus* complex were selected for a pilot study using a colorimetric assay for determination of manogepix activity. With the same inoculum preparation, each isolate was inoculated in double to allow addition of XTT (XTT sodium salt BioChemica (PanReac AppliChem GmbH, Darmstadt, Germany)/menadione (MEN) (Sigma-Aldrich, Denmark) after both 24 h and 48 h of incubation. The standard EUCAST inoculum and 1:10, 1:100, and 1:1,000 dilutions (corresponding to $1 \times 10^5$ to $2.5 \times 10^5$, $1 \times 10^4$ to $2.5 \times 10^4$, $1 \times 10^3$ to $2.5 \times 10^3$, and $1 \times 10^2$ to $2.5 \times 10^2$ CFU/ml, respectively) of the standard EUCAST inoculum were tested. Each inoculum dilution had two positive controls and one negative control. XTT/MEN preparations were prepared, added, and incubated at 37°C and read at 1, 2, and 3 h at 450/630 nm as previously described (13).

**Data management.** MIC ranges, modal MIC (the most common MIC), $MIC_{50}$ (the MIC value that includes 50% of the isolates), and $MIC_{90}$ (the MIC value that includes 90% of the isolates) values were calculated. Correlations between manogepix MECs and azole MICs were determined with Pearson's analysis for each drug after $\log_2$ transformation using GraphPad Prism version 8.3.0 (GraphPad Software).

## SUPPLEMENTAL MATERIAL

Supplemental material is available online only.

**SUPPLEMENTAL FILE 1**, PDF file, 0.8 MB.

## ACKNOWLEDGMENTS

We thank research technician Birgit Brandt for excellent technical assistance.

This study was supported by an unrestricted grant from Amplyx Pharmaceuticals. The funder had no influence on the study design nor on the analysis of the results.

Outside this work, the authors have the following potential conflicts to declare. M.C.A. has, over the past 5 years, received research grants/contract work (paid to the SSI) from Amplyx, Basilea, Cidara, F2G, Gilead, Novabiotics, Scynexis, and T2Biosystems and speaker honoraria (personal fee) from Astellas, Gilead, Novartis, MSD, and Seges. She is the current chairman of the EUCAST-Antifungal susceptibility testing (AFST). K.M.T.A has, over the past 5 years, received travel grants from Basiliea, Pfizer, and the Nordic Society of Medical Mycology and speaker honoraria (personal fee) from Pfizer. K.M.J. has received travel grants from Amplyx Pharmaceuticals and F2G and a meeting grant from MSD.

## REFERENCES

1. Miyazaki M, Horii T, Hata K, Watanabe NA, Nakamoto K, Tanaka K, Shirotori S, Murai N, Inoue S, Matsukura M, Abe S, Yoshimatsu K, Asada M. 2011. In vitro activity of E1210, a novel antifungal, against clinically important yeasts and molds. Antimicrob Agents Chemother 55:4652–4658. https://doi.org/10.1128/AAC.00291-11.
2. Berkow EL, Lockhart SR. 2018. Activity of novel antifungal compound APX001A against a large collection of Candida auris. J Antimicrob Chemother 73:3060–3062. https://doi.org/10.1093/jac/dky302.
3. Arendrup MC, Chowdhary A, Astvad KMT, Jørgensen KM. 2018. APX001A in vitro activity against contemporary blood isolates and candida auris determined by the EUCAST reference method. Antimicrob Agents Chemother 62:1–9. https://doi.org/10.1128/AAC.01225-18.
4. Pfaller MA, Huband MD, Flamm RK, Bien PA, Castanheira M. 2019. In vitro activity of APX001A (manogepix) and comparator agents against 1,706 fungal isolates collected during an international surveillance program in 2017. Antimicrob Agents Chemother 63:e00840-19. https://doi.org/10.1128/AAC.00840-19.
5. Hager CL, Larkin EL, Long L, Zohra Abidi F, Shaw KJ, Ghannoum MA. 2018. In vitro and in vivo evaluation of the antifungal activity of APX001A/APX001 against Candida auris. Antimicrob Agents Chemother 62:e02319-17. https://doi.org/10.1128/AAC.02319-17.
6. Pfaller MA, Duncanson F, Messer SA, Moet GJ, Jones RN, Castanheira M. 2011. In vitro activity of a novel broad-spectrum antifungal, E1210, tested against Aspergillus spp. determined by CLSI and EUCAST broth microdilution methods. Antimicrob Agents Chemother 55:5155–5158. https://doi.org/10.1128/AAC.00570-11.
7. Castanheira M, Duncanson FP, Diekema DJ, Guarro J, Jones RN, Pfaller MA. 2012. Activities of E1210 and comparator agents tested by CLSI and EUCAST broth microdilution methods against Fusarium and Scedosporium species identified using molecular methods. Antimicrob Agents Chemother 56:352–357. https://doi.org/10.1128/AAC.05414-11.
8. Rivero-Menendez O, Cuenca-Estrella M, Alastruey-Izquierdo A. 2019. In vitro activity of APX001A against rare moulds using EUCAST and CLSI methodologies. J Antimicrob Chemother 74:1295–1299. https://doi.org/10.1093/jac/dkz022.
9. Alkhazraji S, Gebremariam T, Alqarihi A, Gu Y, Mamouei Z, Singh S, Wiederhold NP, Shaw KJ, Ibrahim AS. 2019. Fosmanogepix (APX001) is effective in the treatment of immunocompromised mice infected with invasive pulmonary scedosporiosis or disseminated fusariosis. Antimicrob Agents Chemother 64:1–12. https://doi.org/10.1128/AAC.01735-19.
10. Hata K, Horii T, Miyazaki M, Watanabe N-A, Okubo M, Sonoda J, Nakamoto K, Tanaka K, Shirotori S, Murai N, Inoue S, Matsukura M, Abe S, Yoshimatsu K, Asada M. 2011. Efficacy of oral E1210, a new broad-spectrum antifungal with a novel mechanism of action, in murine models of candidiasis, aspergillosis, and fusariosis. Antimicrob Agents Chemother 55:4543–4551. https://doi.org/10.1128/AAC.00366-11.
11. Gebremariam T, Alkhazraji S, Gu Y, Singh S, Alqarihi A, Shaw KJ, Ibrahim AS. 2019. Galactomannan is a biomarker of fosmanogepix (APX001) efficacy in treating experimental invasive pulmonary aspergillosis. Antimicrob Agents Chemother 64:e01966-19. https://doi.org/10.1128/AAC.01966-19.
12. Zhao M, Lepak AJ, Marchillo K, Vanhecker J, Sanchez H, Ambrose PG, Andes DR. 2019. APX001 pharmacokinetic/pharmacodynamic target determination against aspergillus fumigatus in an in vivo model of invasive pulmonary aspergillosis. Antimicrob Agents Chemother 63:e02372-18. https://doi.org/10.1128/AAC.02372-18.
13. Meletiadis J, Siopi M, Kanioura L, Jørgensen KM, Perlin DS, Mouton JW, Arendrup MC. 2020. A multicentre study to optimize echinocandin susceptibility testing of Aspergillus species with the EUCAST methodology and a broth microdilution colorimetric method. J Antimicrob Chemother. https://doi.org/10.1093/jac/dkaa102.
14. Kapoor M, Moloney M, Soltow QA, Pillar CM, Shaw KJ. 2019. Evaluation of resistance development to the Gwt1 inhibitor manogepix (APX001A) in Candida species. Antimicrob Agents Chemother 64:e01387-19. https://doi.org/10.1128/AAC.01387-19.
15. Arendrup MC, Perkhofer S, Howard SJ, Garcia-Effron G, Vishukumar A, Perlin D, Lass-Flörl C. 2008. Establishing in vitro-in vivo correlations for Aspergillus fumigatus: the challenge of azoles versus echinocandins. Antimicrob Agents Chemother 52:3504–3511. https://doi.org/10.1128/AAC.00190-08.
16. Glass NL, Donaldson GC. 1995. Development of primer sets designed for use with the PCR to amplify conserved genes from filamentous ascomycetes. Appl Environ Microbiol 61:1323–1330. https://doi.org/10.1128/AEM.61.4.1323-1330.1995.
17. Schoch CL, Seifert KA, Huhndorf S, Robert V, Spouge JL, Levesque CA, Chen W, Fungal Barcoding Consortium. 2012. Nuclear ribosomal internal transcribed spacer (ITS) region as a universal DNA barcode marker for fungi. Proc Natl Acad Sci U S A 109:6241–6246. https://doi.org/10.1073/pnas.1117018109.
18. O'Donnell K, Sutton DA, Rinaldi MG, Sarver BAJ, Balajee SA, Schroers H-J, Summerbell RC, Robert VARG, Crous PW, Zhang N, Aoki T, Jung K, Park J, Lee Y-H, Kang S, Park B, Geiser DM. 2010. Internet-accessible DNA sequence database for identifying fusaria from human and animal infections. J Clin Microbiol 48:3708–3718. https://doi.org/10.1128/JCM.00989-10.
19. Guinea J, Verweij PE, Meletiadis J, Mouton JW, Barchiesi F, Arendrup MC, Subcommittee on Antifungal Susceptibility Testing (AFST) of the ESCMID European Committee for Antimicrobial Susceptibility Testing (EUCAST), Arendrup MC, Arikan-Akdagli S, Barchiesi F, Castanheira M, Chryssanthou E, Friberg N, Guinea J, Järv H, Klimko N, Kurzai O, Lagrou K, Lass-Flörl C, Mares M, Matos T, Meletiadis J, Moore CB, Mouton JW, Muehlethaler K, Rogers TR, Andersen CT, Velegraki A. 2019. How to: EUCAST recommendations on the screening procedure E.Def 10.1 for the detection of azole resistance in Aspergillus fumigatus isolates using four-well azole-containing agar plates. Clin Microbiol Infect 25:681–687. https://doi.org/10.1016/j.cmi.2018.09.008.
20. Arendrup MC, EUCAST-AFST, Cuenca-Estrella M, Lass-Flörl C, Hope W. 2012. EUCAST technical note on the EUCAST definitive document EDef 7.2: method for the determination of broth dilution minimum inhibitory concentrations of antifungal agents for yeasts EDef 7.2 (EUCAST-AFST). Clin Microbiol Infect 18:E246–7. https://doi.org/10.1111/j.1469-0691.2012.03880.x.
21. Jensen RH, Hagen F, Astvad KMT, Tyron A, Meis JF, Arendrup MC. 2016. Azole resistant Aspergillus fumigatus in Denmark: a laboratory based study on resistance mechanisms and genotypes. Clin Microbiol Infect 22:570.e1–570.e9. https://doi.org/10.1016/j.cmi.2016.04.001.