A pseudo-outbreak of echinocandin-resistant *Candida glabrata*: The Unreliability of Caspofungin Testing in Predicting Echinocandin Susceptibility

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Objectives

Clinical isolates of *Candida* species typically undergo susceptibility testing (to azoles and echinocandins). Multiple echinocandins are commercially available for in vitro susceptibility testing using an agar gradient diffusion method, including micafungin, caspofungin and anidulafungin. After institutional formulary (and therefore susceptibility testing) was changed from micafungin to caspofungin, we experienced an abrupt increase in echinocandin resistance among clinical isolates of *C. glabrata*. In this study, we determined and quantified discrepancies in echinocandin testing among three agents and found that caspofungin testing results had overestimated echinocandin resistance.

Methods

We queried the laboratory information system for caspofungin-resistant *C. glabrata* isolates, and retrospectively tested these isolates for sensitivity to micafungin and anidulafungin, using an agar diffusion method. We compared these results with those generated prior to the institutional change to caspofungin, and to those generated after adopting the European Committee on Antimicrobial Susceptibility Testing (EUCAST) testing guidelines (inferring caspofungin sensitivity from a combination of results from micafungin and anidulafungin). The study period was a total of 44 weeks. Breakpoints were determined by Clinical & Laboratory Standards Institute (CLSI) guidelines. Additionally, we performed FKS sequencing on six isolates, all of which were wild-type FKS1 and FKS2 (save for non-significant sporadic variations) (Figure 1). Descriptive statistics were used.

Results

A total of 44 *C. glabrata* isolates from various body sites were tested (mostly from sterile sites). The baseline echinocandin non-susceptibility rate prior to the change to caspofungin testing was 5.3% (*n* 19), which increased to 91.7% (*n* 12) coinciding with the change to caspofungin testing (p <0.0001). Institution of EUCAST guidelines resulted in a return to baseline echinocandin non-susceptibility rates of 7.7% (*n* 13). Retrospective testing of micafungin/anidulafungin-sensitive isolates with caspofungin confirmed erroneous detection of echinocandin resistance based on caspofungin MICs (Figure 2).

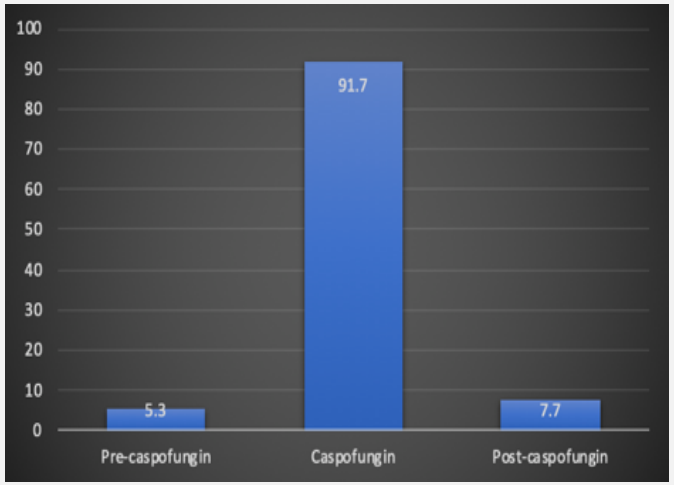

Figure 2. Percentage of *Candida glabrata* clinical isolates non-susceptible to echinocandins stratified based on the phase of laboratory MIC testing.

Isolate	FKS1 Hotspot 1 (FLILSLRDP)	FKS1 Hotspot 2 (DWVRRYTL)	FKS2 Hotspot 1 (FLILSLRDP)	FKS2 Hotspot 2 (DWIRRYTL)
1	SYYFLILSLRDPIRI	PAIDWVRRYTLSIF	YFFLILSLRDPIRI	SPAIDWIRRYTLSIF
2	SYYFLILSLRDPIRI	PAIDWVRRYTLSIF	YFFLILSLRDPIRI	SPAIDWIRRYTLSIF
3	SYYFLILSLRDPIRI	PAIDWVRRYTLSIF	YFFLILSLRDPIRI	SPAIDWIRRYTLSIF
4	SYYFLILSLRDPIRI	PAIDWVRRYTLSIF	YFFLILSLRDPIRI	SPAIDWIRRYTLSIF
5	SYYFLILSLRDPIRI	PAIDWVRRYTLSIF	YFFLILSLRDPIRI	SPAIDWIRRYTLSIF
6	SYYFLILSLRDPIRI	PAIDWVRRYTLSIF	YFFLILSLRDPIRI	SPAIDWIRRYTLSIF

Figure 1. Genotype analysis of FKS1 & FKS2 genes Hotspots demonstrates wild type sequence.

Conclusion

This study quantifies and emphasizes the unreliability of caspofungin testing by agar diffusion for determining echinocandin resistance in *C. glabrata*, which may affect patient management and antifungal choice.

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