**Objectives:** To evaluate the *in vitro* activity of olorofim against thermally-dimorphic fungi cultured from South African patients over the past decade.

**Methods and Materials**: We included 50 isolates of thermally-dimorphic fungi submitted to a reference mycology laboratory for identification between 2010 and 2024. Macroscopic/ microscopic examination of cultured fungi and PCR/ sequencing of the internal transcribed spacer region were used for identification. Olorofim minimum inhibitory concentrations (MIC) were determined using custom-made plates in a broth microdilution method adapted from the Clinical and Laboratory Standards Institute (range 0.008-4 mg/L); MIC endpoints were read at 100% inhibition according to manufacturer’s instructions for both yeast and mould phases. Quality control strains of *Aspergillus fumigatus* ATCC204305 and *Aspergillus flavus* ATCC204304 were included in each run.

**Results:** Of the 50 tested isolates, 38 were *E. africanus*, 10 were *S. schenckii* sensu stricto, 1 was *E. pasteurianus*, and 1 was *T. marneffei* (from a patient with a travel history to China). Both the yeast and mould phases of *E. africanus* and *E. pasteurianus* had MIC values below 0.004 mg/L. The MIC values for *S. schenckii* sensu stricto ranged from 0.015 mg/L to 0.125 mg/L for the yeast phase and 0.031 mg/L to 0.25 mg/L for the mould phase. The single *T. marneffei* isolate had an MIC of 0.031 mg/L for the yeast phase and 0.015 mg/L for the mould phase.

**Conclusion:** Olorofim exhibited potent *in vitro* activity against all tested dimorphic fungi, with good activity against fungi in the genus *Emergomyces*. Olorofim may be an effective antifungal for treatment of a range of endemic mycoses. *In vivo* studies are needed to further assess the efficacy of this antifungal agent.