ABSTRACT

Background: E-101 solution (E-101) is a novel cell-free myeloperoxidase-mediated antimicrobial developed for topical application directly into surgical wounds. It is composed of 1) porcine myeloperoxidase (pMPO) and glucose oxidase (GO), 2) glucose, 3) sodium chloride, and 4) specific amino acids. Once activated, hydrogen peroxide (H₂O₂) is produced in situ by GO dehydrogenation of glucose and reduction of oxygen. The pMPO-catalyzed oxidation of chloride by H₂O₂ generates hypochlorous acid (HOCl). Once generated, HOCl reacts in a diffusion-controlled reaction with a second H₂O₂ molecule to yield singlet oxygen. The in vitro fungicidal activity of E-101 against Candida spp and other yeast-like organisms was investigated.

Methods: Ten clinical isolates of Candida spp. (C. albicans-1, C. dubliniensis-1, C. glabrata-2, C. guilliermondii-1, C. krusei-1, C. parapsilosis-2, C. tropicalis-2, C. albicans ATCC 90028, C. parapsilosis ATCC 22019, C. krusei 6258, Cryptococcus neoformans, and Saccharomyces cerevisiae) were evaluated. Time kill studies were performed using a modified suspension-neutralization method.

RESULTS

Fungal strains. Clinical strains of Candida species (10), Cryptococcus neoformans (1), and Saccharomyces cerevisiae (1) were selected based on their common frequency of isolation (Table 1) and resistance phenotype. C. albicans ATCC 90028, C. parapsilosis ATCC 22019, and C. krusei 6258 were also evaluated.

Antimicrobial agent. Stock solutions of E-101 enzyme solution and substrate solution were prepared at Exoxemis, Inc. (Little Rock, AR). E-101 contains pMPO, GO derived from Aspergillus Niger, and proprietary amino acids in an aqueous formulation vehicle consisting of 150 mM sodium chloride and 0.02% w/v polysorbate 80 in pH 6.5, 20 mM sodium phosphate buffer. The substrate solution contains 300 mM glucose in the same aqueous formulation as the enzyme solution. The enzyme and substrate solutions are packaged in two separate vials and mixed together to activate the system.

Time-kill assay. Time kill studies were performed using modified suspension-neutralization (3). Yeast suspensions were prepared from stationary phase growth on Sabouraud's dextrose agar. Reaction tubes were prepared to contain the appropriate inoculum (~5 x 10⁵ CFU/ml), substrate and enzyme solution. The final concentrations of pMPO in the enzyme solution were 150, and 300 GU pMPO/ml. Reaction tubes were incubated at room temperature and samples were removed at 0, 5, 15, 30, 60, and 120 minutes for quantitative culture. The log₁₀ CFU survivors were determined at each time point.

CONCLUSIONS

• Time-kill studies showed that E-101 solution is fungicidal against Candida species, Cryptococcus neoformans, and Saccharomyces cerevisiae in a time-dependant manner.

• The rapid rate of killing induced by E-101 solution is consistent with its oxidative oxygenation mode of action.

• Inoculum size did not have a major influence on in vitro activity of E-101 solution.

• These data further illustrates the broad spectrum of activity of E-101 solution and its potential fungicidal (candidicidal or yeast-killing) activity when used topically to prevent surgical site infections.

REFERENCES

