

TITLE: EFFECT OF THE ETHYL ACETATE FRACTION OF GREEN PROPOLIS ON MICROBIAL GROWTH AND ITS APPLICATION IN COSMETIC EMULSION

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ABSTRACT:

Propolis is a resinous material produced by bees from exudates of plants, with variable chemical composition. The use of these raw materials of natural origin in several products is a promising trend for the industry, by the antioxidant and antimicrobial activities. The objective of this study was to verify the effect of the ethyl acetate fraction (EAF) of green propolis on microbial growth and its application in cosmetic emulsion. First crude ethanolic extract was obtained and, by liquid-liquid extraction, the EAF of propolis. The minimum inhibitory concentration (MIC) against *Candida albicans* (ATCC10231), *Staphylococcus aureus* (ATCC14458), *Staphylococcus aureus* MRSA (ATCC25923), *Escherichia coli* (ATCC10536), *Salmonella* Typhi (ATCC06539) and *Pseudomonas aeruginosa* (ATCC27853) were determined. Subsequently, a death curve was constructed against these microorganisms using EAF at concentrations related to MIC and MICx2. Four non-ionic oil/water emulsions were prepared, F1 and F2 controls (with and without synthetic preservatives, respectively), F3 and F4 containing 2 % EAF (with and without synthetic antioxidants, respectively). The emulsions were submitted to 40 °C, 5 °C and exposure to ultraviolet radiation (UVR) for 60 days, and physical-chemical and microbiological parameters were evaluated. The EAF had MICs of 2.5, 5.0, 10.0, 20.0, 20.0 and 20.0 mg mL⁻¹ for *C. albicans*, *S. aureus*, *S. aureus* MRSA, *E. coli*, *S. Typhi* and *P. aeruginosa*, respectively. In the death curve, was considered microbicidal activity a reduction > 3 log₁₀ colony-forming units per mL and, otherwise, microbiostatic activity. Thus, EAF against *C. albicans* was fungicide in MIC at 24 h; *S. aureus* was bactericidal in MIC at 12 h; *S. aureus* MRSA was bactericidal in MIC at 24 h; *E. coli*, *S. Typhi* and *P. aeruginosa* was bacteriostatic in MIC up to 24 h of exposure. In the stability tests, F2 emulsion showed significant reduction of pH after 30 days at 40 °C and UVR, significant increase in peroxides index after 60 days at 40 °C and increase in density and scatterability after 60 days at 40 °C and UVR. It was also observed fungal and bacterial growth in F2 at 40 °C after 60 days. Emulsions F3 and F4 remained stable for 60 days. In this way, it was possible to evaluate that the propolis fraction presented antimicrobial activity *in vitro* and the concentration of 2 % incorporated in the emulsion was effective in maintaining the stability of the product elaborated.

Keywords: Antimicrobial activity, microbial death kinetics, minimum inhibitory concentration, stability study