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ANTIMICROBIAL ACTIVITY OF AN ECOFRIENDLY BEE-DERIVED PRODUCT

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Introduction

Propolis (bee glue) is a complex mixture of resins, waxes, and gums, and it is a resinous exudate manufactured by honey bees by combining saps, resins, and mucilages extracted from different plant components, including flower buds, tree barks, and leaves, with beeswax and a number of bee enzymatic proteins.

The chemical composition of propolis reflects the flora used by bees and can vary according to regional seasonality, which can influence its potential for action and its physical, chemical, and biological properties. It is usually composed of 50% plant resin and balsam, 30% wax, 10% essential and aromatic oils, 5% pollen, and 5% other various substances, including organic residues. In general, propolis can contain flavonoids, modified carboxylic acids, higher hydrocarbons, alcohols, aromatic acids, higher fatty acids typical of waxes and their esters, ketones, flavones and flavanols, flavanones, chalcones and dihydrochalcones, terpenoids, steroids, amino acids, sugars, lignans, vitamins and minerals, compounds that have been linked with antioxidant, anti-inflammatory, anti-tumor and antimicrobial potential.

Propolis tincture is a concentrated liquid extract made by soaking raw bee propolis in a hydroalcoholic solution and depending on propolis characteristics it can have different antimicrobial properties. The aim of the present study was to evaluate the antimicrobial activity of a propolis tincture obtained from an ecological Romanian product from the Sibiu area.

Materials and methods

In this study we tested the antibacterial activity of a bee-derived product – propolis tincture, an ecological Romanian product from the Sibiu area.

Qualitative and quantitative evaluation of their antimicrobial activity was tested using standard microbial strains: *Enterococcus faecalis* ATCC 29212, *Escherichia coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 27853. The qualitative testing was performed using an adapted Kirby-Bauer method, briefly, on agar plates inoculated with bacterial suspensions 10µl propolis and alcohol were spotted in order to determine the inhibition zones after overnight incubation. Broth microdilution method was used to determine the minimum inhibitory concentration (MIC). This quantitative testing was performed in 96-well plates, in Mueller-Hinton broth serial two-fold dilutions of the tincture and alcohol were inoculated with standardized suspensions of

the target bacteria. The lowest concentration of the antimicrobial agent that stops visible growth of a microorganism is considered MIC.
The propolis tincture is ethanol based, so solvent-matched ethanol control at exactly the residua ethanol concentration present in each assay will be included.

Results and conclusions

In the agar diffusion assay, the qualitative evaluation, the propolis tincture produced an inhibitory effect against *E. faecalis*, whereas for the Gram negative bacteria, no inhibition zones were observed under the conditions tested. The ethanol control at the concentration corresponding to that present in the tincture did not produce an inhibitory effect.
Broth microdilution demonstrated antibacterial activity of the propolis tincture against all three tested strains. The MIC was 12.5% for both *E. coli* and *P. aeruginosa*, while a lower MIC of 3% was observed for *E. faecalis*. In comparison, the ethanol control showed a MIC of 50% against *E. coli* and *P. aeruginosa*, indicating that the inhibitory activity observed at 12.5% was not attributable solely to the ethanol solvent.
The tested propolis tincture demonstrated antibacterial activity against the investigated bacterial reference strains, with *E. faecalis* showing greater susceptibility than the Gram-negative bacteria tested. These preliminary findings support further investigation of Romanian propolis preparations as potential sources of antibacterial compounds. Additional studies, including testing against clinical isolates, characterization of the tincture's chemical composition, and assessment of its activity in relevant biological models, are needed to evaluate its potential therapeutic applications.

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