**Chemical Composition and the Anticancer, Antimicrobial, and Antioxidant Properties of *Acacia* Honey from the Hail Region: The in vitro and in silico Investigation**


## ABSTRACT

In consideration of the emergence of novel drug-resistant microbial strains and the increase in the incidences of various cancers throughout the world, honey could be utilized as a great alternative source of potent bioactive compounds. In this context, this study pioneers in reporting the phytochemical profiling and the antimicrobial, antioxidant, and anticancer properties of *Acacia* honey (AH) from the Hail region of Saudi Arabia, assessed using *in vitro* and molecular docking approaches. The phytochemical profiling based on high-resolution liquid chromatography-mass spectrometry (HR-LCMS) revealed eight compounds and three small peptide-like proteins as the constituents. The honey samples exhibited promising antioxidant activities (DPPH-$IC_{50}$ = 0.670 mg/mL; ABTS-$IC_{50}$ = 1.056 mg/mL; $\beta$-carotene-$IC_{50}$ > 5 mg/mL). In the well-diffusion assay, a high mean growth inhibition zone (mGIZ) was observed against *Staphylococcus aureus* (48.33 ± 1.53 mm), *Escherichia coli* ATCC 10536 (38.33 ± 1.53 mm), and *Staphylococcus epidermidis* ATCC 12228 (39.33 ± 1.15 mm). The microdilution assay revealed that low concentrations of AH could inhibit the growth of almost all the evaluated bacterial and fungal strains, with the minimal bactericidal concentration values (MBCs) ranging from 75 mg/mL to 300 mg/mL. On the contrary, high AH concentrations were required to kill the tested microorganisms, with the minimal bactericidal concentration values (MBCs) ranging from approximately 300 mg/mL to over 600 mg/mL and the minimal fungicidal concentration values (MFCs) of approximately 600 mg/mL. The AH exhibited effective anticancer activity in a dose-dependent manner against breast (MCF-7), colon (HCT-116), and lung (A549) cancer cell lines, with the corresponding $IC_{50}$ values of 5.053 $\mu$g/mL, 5.382 µg/mL, and 6.728 $\mu$g/mL, respectively. The in silico investigation revealed that the observed antimicrobial, antioxidant, and anticancer activities of the constituent compounds of AH are thermodynamically feasible, particularly those of the tripeptides (Asp-Trp-His and Trp-Arg-Ala) and aminocyclitol glycoside. The overall results highlighted the potential of AH as a source of bioactive compounds with significant antimicrobial, antioxidant, and anticancer activities, which could imply further pharmacological applications of AH.