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Comprehensive Assessment of the Biological and Chemical Properties of *Ficus carica* Leaf Ethanolic Extract

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ABSTRACT

Ficus carica L. is traditionally used for its medicinal properties. Despite its widespread use, the mechanisms underlying its biological activity remain insufficiently explored. This study aims to investigate the antioxidant, anti-inflammatory, and antimicrobial properties of the ethanolic leaf extract of *F. carica* harvested in Armenia, alongside its chemical composition.

The leaves of *F. carica* were collected at various growing periods, and ethanolic extracts were prepared. Radical-scavenging activity was assessed using DPPH assay. Thermal stability of extract components was tested at 70 °C and under autoclave conditions. Metal-chelating activity was also determined. Chemical profiling was conducted by LC-Q-Orbitrap HRMS analysis. The biological activity of the extract was tested in using *Escherichia coli* QC772 (*E. coli* K12 derivative, carrying soda-lacZ genes), and BV-2 *Wt* and Acyl-CoA oxidase 1 deficient (*Acox1*$^{-/-}$) cell lines, focusing on nitric oxide (NO) production, antioxidant enzyme activity (SOD, catalase), and lipid peroxidation. Antimicrobial activity was evaluated using the disk diffusion method and growth rate monitoring. To elucidate the potential mechanisms of the antibiotic activity of test extracts, the changes in H^+-fluxes across the cell membrane and their impact on the H^+-translocating F_OF_1-ATPase activity in *E. coli* explored. The extract showed strong antioxidant activity, which varied by leaf harvesting time. Bioactive compounds remained relatively stable at up to 70 °C, but were partially degraded under autoclaving. High total flavonoid and phenolic content strongly correlated with biological activity. The extract significantly reduced NO production and lipid peroxidation in *E. coli* and both microglial cell models, while enhancing antioxidant enzyme activity. Notable antimicrobial activity and metal-chelating capacity were observed. The results indicated that plant extract enhanced H^+-fluxes in the investigated bacterial strain and promoted ATPase activity, suggesting a potential role in altering bacterial membrane integrity. LC-Q-Orbitrap HRMS analysis identified over 140 distinct phytochemicals mainly belonging to flavonoids and phenolics. The ethanolic leaf extract of *F. carica* demonstrates potent antioxidant, anti-inflammatory, and antimicrobial properties, supported by rich phytochemical diversity. These findings suggest its therapeutic potential in oxidative stress- and inflammation-related conditions, warranting further pharmacological studies.

Keywords: phenolics and flavonoids, antoxidant activity, microglial cell lines, *Escherichia coli*

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