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Evaluation of antioxidant and anti-inflammatory properties of leaves of *Impatiens repens*

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Impatiens repens, a plant species highly valued for its medicinal properties, has been used in traditional medicine to treat various ailments, such as epilepsy, insanity, hemorrhoids, and gonorrhoea. This species contains phytochemicals such as tannins, flavonoids, and phenols that are believed to possess antioxidant and anti-inflammatory properties, making it a potential source for developing novel anti-inflammatory drugs. This study aimed to investigate the medicinal properties of *I. repens* and evaluate its potential as a valuable source of natural anti-inflammatory compounds. The research focused on exploring the antioxidant and anti-inflammatory activities of *I. repens* leaf extracts. Phytochemicals of *I. repens* leaves, first extracted into hexane by cold extraction were sequentially extracted into ethyl acetate and methanol, respectively. The antioxidant potential of three solvent extracts (hexane, ethyl acetate, and methanol) was evaluated using the ferric reducing antioxidant power (FRAP) and 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay. The methanol extract demonstrated the highest radical scavenging activity, with an IC₅₀ value of 407.63 ± 12.24 µg/mL, indicating its ability to effectively act as an antioxidant. Similarly, in the FRAP assay the methanol extract exhibited the highest reducing power, with a FRAP value of 661.56 ± 16.69 µg (Fe²⁺)/g extract. Furthermore, the total phenolic content (TPC) and total flavonoid content (TFC) of the extracts were determined. The methanol extract exhibited the highest TPC (120.52 ± 4.47 mg GAE/g extract) and TFC (17.77 ± 4.47 mg CE/g extract) among the solvent extracts. In addition, the radical scavenging properties of all solvent fractions were investigated using DPPH scavenging assay and their correlations with phenolics and flavonoids were analyzed using Pearson's correlation. Strong and positive correlation coefficients of 0.99 and 0.99 were observed respectively. These results reflected that both phenolics and flavonoids contributed to elicit antioxidant properties of *I. repens* leaves. To assess the anti-inflammatory potential of *I. repens*, two assays were performed: the egg albumin denaturation assay and the Human red blood cell (HRBC) membrane stabilization assay. In the egg albumin denaturation assay, the methanol extract exhibited marked and concentration-dependent anti-inflammatory activity, with an IC₅₀ value of 346.58 ± 26.93 µg/mL. In the HRBC membrane stabilization assay, the hexane extract demonstrated significant inhibition of protein denaturation with an IC₅₀ value of 387.73 ± 4.42 µg/mL, comparable to the standard drug used, o-acetylsalicylic acid. These results suggest that leaves of *I. repens* possesses potent anti-inflammatory properties, making it a promising candidate for the development of new anti-inflammatory drugs. The findings of this study validate the use of *I. repens* in traditional medicine for various ailments and highlight its potential as a valuable medicinal plant. The rich content of phenolic compounds and flavonoids in the methanol extract further supports its antioxidant activities. Given the increasing demand for effective and safe anti-inflammatory drugs, *I. repens* holds promise as a natural alternative to conventional treatments.

Keywords: Anti-inflammatory, Antioxidants, Egg albumin denaturation, HRBC membrane, *Impatiens repens*