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Assessment of the antioxidant, anti-diabetic, and anti-obesity activities of a Sri Lankan "spice" mixture at different storage conditions used for treating obesity

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Obesity, diabetes, and hypertension have become prevalent health issues in Asian countries, affecting individuals at young age. Systemic oxidative stress and adipose tissue contribute significantly to the development of insulin resistance associated with obesity and type II diabetes. Due to the possible toxicity and carcinogenicity of synthetic antioxidants, there is a global need to discover safe antioxidants to use as food additives. To address these concerns, clinical trials were conducted in Sri Lanka using a spice mixture consisting of commonly used spices in Sri Lankan cuisine: *Cinnamomum zeylanicum*, *Cuminum cyminum*, *Piper nigrum*, *Murraya koenigii*, and *Allium sativum*. This study aimed to evaluate the antioxidant, anti-obesity, and anti-diabetic potential of a methanol extract derived from this Sri Lankan spice mixture, as well as to evaluate the above activity properties of five glass bottle samples stored at different storage conditions: the positive control (STD), the initial spice mixture sample (INT), the sample after three months at room temperature (RT), the sample after three months in a dark environment (BLK), and the sample after three months in a refrigerator (COOL). The IC_{50} value of the methanol extract of INT demonstrated significant antioxidant activity ($0.08 \pm 0.03 \times 10^{-2}$ mg/mL, $p < 0.05$) in the DPPH assay compared to the positive control, BHT ($0.07 \pm 0.05 \times 10^{-2}$ mg/mL). Similarly, the lower IC_{50} value of INT indicated higher antioxidant activity ($0.17 \pm 0.14 \times 10^{-1}$ mg/mL) in the ABTS assay when compared to the positive control BHT ($0.36 \pm 0.15 \times 10^{-1}$ mg/mL). In both assays, the IC_{50} values followed the order of INT < COOL < BLK < RT, signifying decreasing antioxidant activity in the same order. The methanol extract samples (1.0 mg/mL) in the FRAP assay showed increasing relative % reducing power in the order of RT < BLK < COOL < INT, with values of 30.37%, 32.14%, 32.74%, and 37.05%, respectively, which was lower compared to the positive control (BHT). The α-amylase inhibition assay revealed that the IC_{50} of the methanol extract of the INT was $0.29 \pm 0.21 \times 10^{-1}$ mg/mL compared to the positive control Acarbose (IC_{50}, $0.06 \pm 0.02 \times 10^{-1}$ mg/ml). The IC_{50} of the methanol extract of INT in the anti-lipase assay was $0.14 \pm 0.05 \times 10^{-1}$ mg/mL, while the positive control (Orlistat) had an IC_{50} of $0.09 \pm 0.01 \times 10^{-1}$ mg/mL. These results revealed that the IC_{50} value increased in the order of INT < COOL < BLK < RT, when each glass bottle sample was stored after three months according to their distinct conditions, suggesting a negative correlation between assay activity with light intensity, temperature, and storage time. Overall, the study concluded that the spice mixture used for reducing obesity exhibits significant antioxidant potential. Additionally, the results from the anti-lipase and α-amylase inhibition assays suggest that the bioactive constituents present in the methanol extract have the potential to inhibit lipase and α-amylase enzymes, providing anti-obesity and anti-diabetic benefits.

Keywords: Anti-diabetic, Anti-obesity, Antioxidant, Spice mixture