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Beer enriched with "Lapins" cherry extracts: antioxidant activity and liver toxicity

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Abstract

Background: Beer can be considered a functional beverage and integrate innovative ingredients, namely sweet cherries, with different properties, such as antioxidant activity [1,2]. **Objective:** To evaluate the antioxidant activity, in vitro, and liver toxicity, in human hepatocarcinoma cells (HepG2), in beer after incorporation of aqueous (CAE) and ethanolic (CEE) extracts of cherry variety "Lapins". Methods: CAE and CEE (1mg/mL) were incorporated into commercial bottles of Imperial Stout beer (IS-N). The total phenolic content (TPC), expressed in mg of gallic acid equivalents (GAE)/g, was determined. The antioxidant capacity was evaluated by the 2,2'-azino-bis(3-ethylbenzothiazoline-6sulfonic acid) (ABTS) neutralization and hydrogen peroxide (H₂O₂) assays, both expressed in the concentration required to inhibit the activity by 50% (IC₅₀). Also, the ferric reducing antioxidant power (FRAP) assay was performed and expressed in μmol of trolox equivalent (TE)/mg. Cell toxicity was studied in HepG2 cells, with assessment of metabolic activity by the 3-(4,5-dimethyl-2-thiazolyl)-2,5diphenyl-2H-tetrazolium bromide (MTT) assay. Data were analysed using GraphPad Prism software, and significant differences were considered for p < 0.05. **Results:** The incorporation of CEE to IS-N beer significantly decreased TPC (4.2 \pm 0.1 mg GAE/g for IS-N + CEE; 8.3 \pm 0.2 mg GAE/g for IS-N; p < 0.05). There was an increase in the antioxidant capacity by the ABTS assay (IC₅₀ = 80.1 ± 1.1 μ g/mL for IS-N; IC₅₀ = 60.5 ± 1.5 μ g/mL for IS-N + CAE; IC₅₀ = 48.0 ± 0.6 μ g/mL for IS-N + CEE); however, the incorporation of cherry extracts was not promising in the H_2O_2 (27.0 \pm 1.5 µg/mL for IS-N; $IC_{50} = 58.7 \pm 0.4 \mu g/mL$ for IS-N + CEE; $IC_{50} = 78.8 \pm 1.6 \mu g/mL$ for IS-N + CAE;) and FRAP $(44.4 \pm 0.0 \mu mol/g \text{ for IS-N}; 42.7 \pm 0.0 \mu mol/g \text{ for IS-N} + \text{CAE}; \text{ and } 39.7 \pm 0.0 \mu mol/g \text{ for IS-N} +$ CEE) assays. Furthermore, IS-N + CEE showed greater antioxidant capacity than IS-N + CAE. After the incorporation of cherry extracts, cytotoxicity was observed in concentrations higher than 10 mg/mL (for IS-N + CAE, 24h incubation) and at the concentration of 500 mg/mL (for IS-N + CAE and IS-N + CEE, 48h incubation). IS-N + CEE showed the greatest increase in cell viability. Conclusions: The addition of cherry extracts to beer increased the antioxidant capacity by the ABTS assay, while TPC was reduced with the addition of CEE. The incorporation of both extracts showed promising potential, with low cytotoxicity in HepG2.

Keywords: antioxidant activity; liver toxicity; sweet cherry

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