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Short communication

Evaluation of the antioxidant capacity of four Cannabis cultivars

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Introduction

Cannabis, commonly known as marijuana, is a product of the *Cannabis sativa* plant which has been used as an alternative medicine in many cultures for several centuries ago. Recently, its beneficial effects have been shown in the treatment of nausea and vomiting associated with cancer chemotherapy, anorexia and cachexia seen in HIV/AIDS patients as well as in neuropathic pain and spasticity in multiple sclerosis (Nagarkatti et al., 2010). Nowadays, this plant is being substantially used for medical purposes. The active compounds, cannabinoids, have been under the extensive investigation, and their potent antioxidant and inflammatory properties have been reported, although the detailed mechanisms of their actions have not been fully clarified (Kopustinskiene el al., 2022). On the other hand, it is assumed that the extraction of phenolic compounds as well as antioxidant capacity of *C. sativa* extracts were highly and significantly influenced by the type of extraction solvent (Aazza, 2021).

Regarding this, dried flowers from four different cannabis strains were extracted with two different solvents, methanol and ethanol and antioxidant activity of extracts were compared to see if the antioxidant capacity varies from one solvent to other.

Materials and methods

Plant materials

Plant material was consisted of dried flowers from four different cannabis cultivars: Amnesia Kush, Charlotte's Angels, Elleta Campana and Orange Hill.

Total phenolic content (TPC)

TPC of the methanolic and ethanolic extracts, prepared at concentration of 50 mg/ml in methanol and ethanol was determined using Folin-Ciocalteu reagent described by the Singleton method with some modifications. Gallic acid was used as a positive control.

Total flavonoid content (TFC)

TFC was evaluated according to Jiao method, using $AlCl_3$ with slight modifications. The flavonoid content was expressed as quercetin equivalent (mg QE/g) using the linear equation based on the standard calibration curve.

Antioxidative capacity

DPPH assay of the samples was conducted by Gyamfi method, with slight modification, while linoleic acid/β-carotene bleaching assay was used to determine the initial value of lipid peroxidation.

Ascorbic acid (AA) and butylated hydroxyanisole (BHA) were used as positive controls for both assays.

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Results and discussions

According to the results obtained in this study, both Amnesia Kush and Charlottes Angels samples showed higher values for TPC in ethanolic extracts, 4.88 ± 0.33 mg GAE/g and 5.05 ± 0.39 mg GAE/g, respectively compared to the values in methanolic extract, 4.29 ± 0.36 for Amnesia Kush are 3.19 ± 0.04 mg GAE/g for Charlottes Angel. Contrary to the previous results, methanolic extracts showed higher values for TPC for Eletta Campana and Orange Hill special extracts, with values at 4.26 ± 0.04 and 4.39 ± 0.25 mg GAE/g, compared to the values of ethanolic extracts, 3.15 ± 0.03 and 4.35 ± 0.28 mg GAE/g, respectively. This finding are consistent with that of Farrante et al. (2019) who determined the total phenolic content in flowers of *C. sativa* to be in a range of 4.7 to 8.1 mg GAE/g.

All four extracts showed higher values for TFC when methanol was used as solvent. The highest values for TFC were noticed in Charlottes Angel (4.68 ± 0.29), followed by Amnesia Kush (4.41 ± 0.44), Orange Hill special (3.48 ± 0.14) and Eletta Campana (3.22 ± 0.65). According to the Claudio et al. (2019), the determined TFC values for the water extracts prepared from cannabis inflorescence is up to 6.3 mg RE/g extract (Ferrante et al., 2019).

According to our findings, both methanolic and ethanolic extracts scavenged the DPPH free radicals. The IC_{50} values for DPPH radicals in methanolic and ethanolic extract of the Amnesia Kush were found to be 9.92 mg/mL and 15.56 mg/mL, respectively. Lower IC_{50} values for methanolic extracts were also noticed in Charlotte Angels extracts as values of 10.59 mg/mL and 9.90 mg/mL for methanolic and ethanolic extracts, respectively, were determined. Opposing to the previous results, higher IC_{50} values for ethanolic extracts were determined in Elleta Campana and Orange Hill extracts. Values of 15.56 mg/mL and 20.52 mg/mL were calculated for ethanolic extracts, while the IC_{50} values for methanolic extracts were lower and ranged from 9.92 mg/mL to 12.5 5mg/mL, respectively. The scavenging capacity of tested extracts was compared to BHA and AA, as positive controls. In this manner, all of the extracts showed significant higher IC_{50} values compared to BHA and AA, which were determined to be 0.0450 mg/mL and 0.014 mg/mL, respectively.

According to the *β*-carotene/linoleic acid bleaching test, all methanolic extracts at concentration of 1 mg/mL showed percentage of inhibition between 80% and 90%. Additionally, our research showed that the ethanolic extracts in the same concentration have lower percentage of inhibition compared with the methanolic extracts. In comparison to this, BHA at concentration of 1 mg/mL methanolic as well as ethanolic matrix, showed inhibition of 99.7%, and 91.7%, respectively. This finding is consistent with that of Smeriglio et al. (2016), who reported high antioxidative activity of Cannabis flower measured by β-carotene bleaching assay.

Conclusion

The type of the solvent which is used for the sample preparation influences the TPC and TFC, as well as affect the antioxidant capacity of the prepared Cannabis extracts. The obtained values for the tested antioxidative activity is probably due to the TPC and TFC values. Further examination, which take these variables into account, will need to be undertaken in order to see if there is a possible synergistic effects with some other compounds present in Cannabis flowers.

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