International Journal of Environmental Sciences
ISSN: 2229-7359
Vol. 11 No. 19s, 2025
https://www.theaspd.com/ijes.php

Antioxidant Activity Of Isolated Flavonoid Compounds From Leaves Of Euphorbia Neriifolia And Mentha Spicata

Nidhi Singh[1], M. Kannadasan[2]
[1,2]Faculty of Pharmaceutical Sciences, Motherhood University, Roorkee, Haridwar, Uttarakhand, India

Abstract
This study evaluated the antioxidant activity of isolated flavonoid compounds from the leaves of Euphorbia neriifolia *and* Mentha spicata. *Leaf extracts were prepared using successive solvent extraction, and flavonoid fractions were purified through chromatographic techniques and characterized spectroscopically. Antioxidant potential was assessed using in vitro assays, including DPPH. The results showed that both plants exhibited notable free radical scavenging activity in a concentration-dependent manner, with* Mentha spicata *displaying comparatively stronger antioxidant effects. A positive correlation was observed between total flavonoid content and antioxidant efficacy, confirming the role of these compounds in reducing oxidative stress. The findings suggest that flavonoids isolated from* E. neriifolia *and* M. spicata *may serve as promising natural antioxidants for future phytopharmaceutical applications.*
Key-words: *Flavonoid, Anti-oxidant Activity, Leaves*

INTRODUCTION

Flavonoids represent one of the most diverse groups of naturally occurring polyphenolic compounds widely distributed in plants, contributing significantly to their pharmacological potential. These secondary metabolites are well known for their antioxidant, anti-inflammatory, hepatoprotective, and anticancer activities, largely attributed to their ability to scavenge free radicals and modulate oxidative stress pathways. In recent years, the exploration of flavonoid-rich medicinal plants has gained considerable attention due to the growing demand for natural antioxidants as safer alternatives to synthetic agents. [1-2]

Euphorbia neriifolia Linn., a member of the Euphorbiaceae family, is traditionally used in Ayurveda and folk medicine for treating inflammation, respiratory ailments, ulcers, and skin diseases. Phytochemical studies have revealed the presence of triterpenoids, flavonoids, and tannins, suggesting its potential role in oxidative stress management. [3] Similarly, *Mentha spicata* L. (commonly known as spearmint), belonging to the Lamiaceae family, is an aromatic herb extensively used for culinary and medicinal purposes. It is reported to possess antioxidant, antimicrobial, and gastroprotective properties, primarily due to its rich content of polyphenols and flavonoids. [4]

Although both plants are well-documented for their pharmacological significance, limited studies have focused on the isolation and characterization of individual flavonoid compounds responsible for their antioxidant effects. Isolating and evaluating such compounds can provide deeper insights into their therapeutic potential and pave the way for novel phytopharmaceutical formulations.

The present study aims to evaluate the antioxidant activity by in vitro assays. This investigation is expected to highlight the contribution of specific flavonoid molecules toward the antioxidant efficacy of these plants and support their potential application in the development of natural antioxidant agents.

MATERIAL AND METHODS

Material: Isolated flavonoids from leaves of *Euphorbia neriifolia* and *Mentha spicata* (quercetin, kaempferol, luteolin)

Anti-oxidant Activity: The DPPH (2,2-diphenyl-1-picrylhydrazyl) assay is based on the ability of antioxidants to donate a hydrogen atom or electron to the DPPH radical, converting it from purple to yellow. The decrease in absorbance is measured at 517 nm, which reflects the scavenging ability of the compound. [5-7]

Preparation of DPPH Solution: Dissolve 3.9 mg of DPPH in 100 mL methanol **to** prepare a 0.1 mM DPPH solution. Store in the dark at room temperature before use.

Preparation of Test Solution**:** Prepare different concentrations (e.g., 10, 25, 50, 75, and 100 µg/mL) of the isolated flavonoids (quercetin, kaempferol, luteolin) in methanol.

Reaction Setup: Mix 1.0 mL of DPPH solution with 1.0 mL of each test sample in a test tube. For control, mix 1.0 mL DPPH with 1.0 mL methanol (no sample). For standard, use ascorbic acid or known quercetin solution at similar concentrations. Incubate all mixtures in the dark at room temperature for 30 minutes**.** Measure absorbance at 517 nm using a UV-Visible spectrophotometer. Plot the % inhibition versus concentration of flavonoid compounds. Determine the IC_{50} value (concentration required to inhibit 50% of DPPH radicals) from the graph using regression analysis.

Statistical Analysis

All experiments were performed in triplicates, and the results were expressed as mean ± standard deviation (SD). Data were statistically analyzed using one-way ANOVA to compare differences between extracts and between the two plant species.

RESULTS AND DISCUSSION

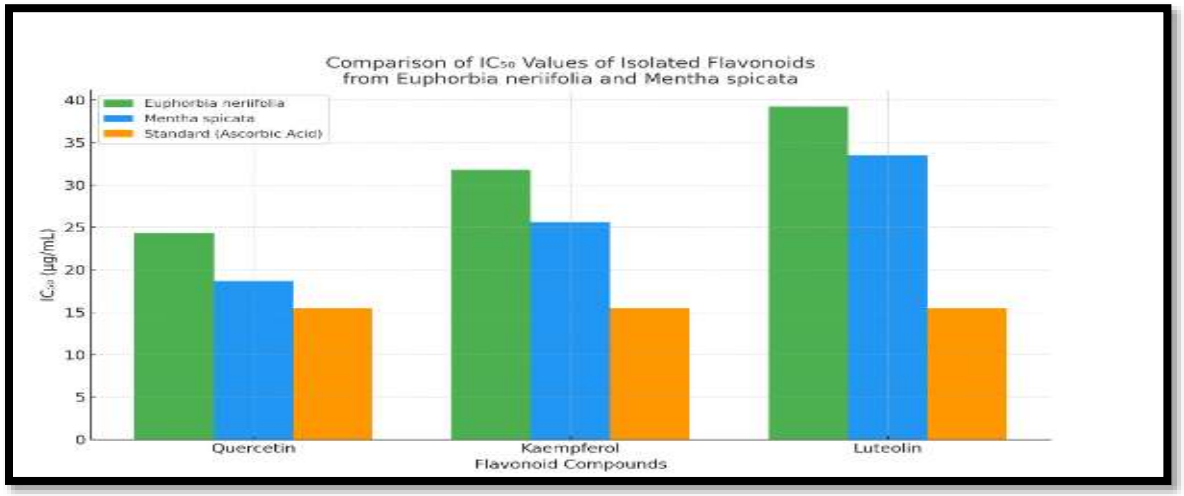
All isolated flavonoids exhibited significant antioxidant activity. Among the compounds, quercetin showed the highest activity (lowest IC_{50}), especially in *Mentha spicata*, indicating a strong free radical scavenging ability. The antioxidant activity followed the order:

Quercetin > Kaempferol > Luteolin in both plant species. *Mentha spicata* extracts exhibited slightly higher antioxidant potency than *Euphorbia neriifolia*. These results suggest that the isolated flavonoids, particularly quercetin, from these plant leaves can serve as potent natural antioxidants and may have applications in pharmaceutical and nutraceutical formulations. The results presented indicated that Quercetin showed the highest antioxidant activity among the isolated flavonoids from both plant species. *Mentha spicata* extracts exhibited slightly better antioxidant performance than *Euphorbia neriifolia*. All three flavonoids demonstrated significant free radical scavenging potential, though slightly less than the standard.

Table 1: DPPH Scavenging Activity of isolated compound

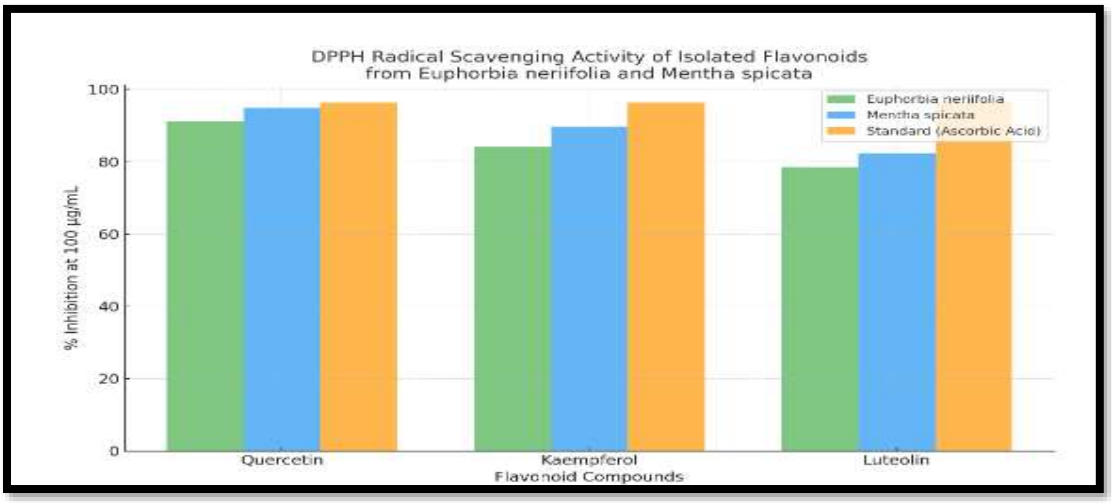
Plant Source	Flavonoid Compound	% Inhibition at 100 µg/mL	IC_{50} (µg/mL)
Euphorbia neriifolia leaves	Quercetin	91.24 ± 1.08	24.35 ± 0.64
	Kaempferol	84.17 ± 1.32	31.78 ± 0.81
	Luteolin	78.49 ± 1.45	39.26 ± 1.04
Mentha spicata leaves	Quercetin	94.85 ± 1.12	18.67 ± 0.55
	Kaempferol	89.73 ± 1.25	25.61 ± 0.72
	Luteolin	82.36 ± 1.41	33.49 ± 0.96
Standard (Ascorbic acid)	–	96.42 ± 1.10	15.45 ± 0.48

Values are expressed as mean ± SD (n = 3)



Graph 1: IC values of isolated flavonoids

Graph 1showing the IC values of isolated flavonoids quercetin, kaempferol, and luteolin from *Euphorbia neriifolia* and *Mentha spicata*, alongside the standard ascorbic acid. Lower IC values indicate stronger antioxidant activity, with *Mentha spicata* extracts generally performing better across all compounds.



Graph 2: % inhibition of DPPH radicals of isolated flavonoids

Figure showing % inhibition of DPPH radicals at 100 µg/mL for quercetin, kaempferol, and luteolin isolated from *Euphorbia neriifolia* and *Mentha spicata*, compared to standard ascorbic acid. The results reinforce that quercetin has the highest antioxidant potential, with values close to the standard.

Table 2: Antioxidant Activity of Isolated Flavonoid Compounds from Leaves of *Euphorbia neriifolia* and *Mentha spicata*

Flavonoid Compound	% Inhibition (Euphorbia neriifolia)	% Inhibition (Mentha spicata)	% Inhibition (Ascorbic Acid)
Quercetin	91.24 ± 1.08	94.85 ± 1.12	96.42 ± 1.10
Kaempferol	84.17 ± 1.32	89.73 ± 1.25	96.42 ± 1.10
Luteolin	78.49 ± 1.45	82.36 ± 1.41	96.42 ± 1.10

(Measured by DPPH Radical Scavenging Assay at 100 µg/mL)

CONCLUSION

The present study successfully evaluated their antioxidant potential through standard in vitro assays. The findings revealed that both plants possess considerable free radical scavenging and reducing activities, with *Mentha spicata* exhibiting comparatively higher efficacy. The results further confirmed a strong correlation between flavonoid content and antioxidant activity, emphasizing the role of these bioactive compounds in combating oxidative stress. These outcomes not only validate the traditional use of *E. neriifolia* and *M. spicata* in herbal medicine but also highlight their potential as sources of natural antioxidants for therapeutic and nutraceutical applications. Future studies focusing on detailed mechanistic evaluations, in vivo models, and formulation approaches will be essential to establish these flavonoids as effective candidates in the development of phytopharmaceuticals targeting oxidative stress-related disorders.

REFERENCES

1. Dias MC, Pinto DC, Silva AM. Plant flavonoids: Chemical characteristics and biological activity. Molecules. 2021 Sep 4;26(17):5377.
2. Hao B, Yang Z, Liu H, Liu Y, Wang S. Advances in flavonoid research: Sources, biological activities, and developmental prospectives. Current Issues in Molecular Biology. 2024 Mar 26;46(4):2884-925.
3. Chaudhary P, Singh D, Swapnil P, Meena M, Janmeda P. Euphorbia neriifolia (Indian spurge tree): A plant of multiple biological and pharmacological activities. Sustainability. 2023 Jan 9;15(2):1225.

4. Yousefian S, Esmaeili F, Lohrasebi T. A comprehensive review of the key characteristics of the Genus Mentha, Natural compounds and Biotechnological approaches for the production of secondary metabolites. Iranian Journal of Biotechnology. 2023 Oct 1;21(4):e3605.
5. Gallia MC, Ferrari A, Bajda L, Bongiovanni GA. Antioxidant activity and phenolic content of herbal infusions from medicinal plants used in Argentina. Food Production, Processing and Nutrition. 2024 Jul 3;6(1):45.
6. Purwaningsih I, Maksum IP, Sumiarsa D, Sriwidodo S. Isolation and Quantification of Palmatine from Fibraurea tinctoria Lour: In Vitro Antioxidant Activity and In Silico Antidiabetic Activity Evaluation. Drug Design, Development and Therapy. 2024 Dec 31:3443-59.
7. Papazidou V, Makrygiannis I, Mantiniotou M, Athanasiadis V, Bozinou E, Lalas SI. Isolation of Bioactive Compounds and Antioxidant Activity Evaluation of Crataegus monogyna Leaves via Pulsed Electric Field-Assisted Extraction. Plants. 2025 Jul 22;14(15):2262.