



Research Article

In-Vitro Antioxidant Evaluation of Crassula ovata Leaf Extract Using DPPH Radical Scavenging Assay and Phytochemical Profiling¹Satheeshkumar N*, ¹V. Anbalagan, ¹S. Kannan, ¹B. Sangameswaran¹SSM College of Pharmacy, S.F. No. 834/1 & 2, Chinniyampalayam Pudur, Jambai Village, Bhavani**Keywords**

Crassula ovata, DPPH assay, phytochemical screening, antioxidant activity, IC₅₀, free radicals.

Abstract

Aims: The present in-vitro investigation assessed the antioxidant potential of Crassula ovata leaf extract through DPPH radical scavenging and preliminary phytochemical analysis. **Study Design:** Experimental laboratory-based in-vitro study. **Methods:** Leaf extract of C. ovata was screened for major phytochemical constituents. Antioxidant capacity was examined using the DPPH radical scavenging method by measuring absorbance changes at 517 nm across different extract concentrations. Percentage inhibition and IC₅₀ values were calculated to determine potency. **Results:** Phytochemical screening revealed the presence of alkaloids, steroids, saponins, and resins, indicating the presence of bioactive constituents capable of donating hydrogen atoms or electrons to neutralize free radicals. C. ovata demonstrated concentration-dependent scavenging of DPPH radicals, with percentage inhibition increasing progressively with dose. The calculated IC₅₀ value suggests moderate to strong antioxidant potential for the plant extract. **Conclusion:** The antioxidant activity exhibited by Crassula ovata may be attributed to its phytochemical constituents. These findings support the plant's potential application as a natural source of antioxidant compounds.

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ISSN: 3049-2955/The authors © 2025, under exclusive license to the Sprout Publication.

DOI: <https://doi.org/10.63785/2025.1.4.486490>**1. Introduction**

Oxidative stress arises from an imbalance between the generation of reactive oxygen species and the capacity of biological systems to neutralize these reactive intermediates [1]. Excessive free radical production leads to oxidative damage of cellular macromolecules, including lipid peroxidation, protein denaturation, and DNA strand breakage, thereby contributing to the onset and progression of numerous degenerative disorders such as cancer, cardiovascular diseases, neurodegenerative conditions, and aging related pathologies. Consequently, the identification of safe and effective antioxidants has become an important focus in pharmaceutical and biomedical research [2].

Natural products, particularly medicinal plants, represent a valuable source of antioxidant compounds due to their rich content of bioactive secondary metabolites [3]. Phytochemicals such as flavonoids, alkaloids, phenolic compounds, terpenoids, steroids, and resins are known to exhibit strong free radical scavenging, metal chelating, and cytoprotective

properties. The exploration of traditionally used plants for antioxidant activity provides scientific validation for ethnomedicinal claims and supports the development of plant based therapeutic agents [4].

Crassula ovata, a succulent plant belonging to the family Crassulaceae, has been traditionally used in folk medicine for the treatment of inflammation, wounds, and various systemic ailments [5]. The plant is reported to contain diverse secondary metabolites, including alkaloids, steroids, and resins, which are often associated with antioxidant and anti-inflammatory activities. Despite its widespread cultivation and traditional importance, limited scientific data are available regarding its antioxidant potential [6].

The present study aims to evaluate the in vitro antioxidant activity of *Crassula ovata* using the DPPH free radical scavenging assay and to perform a preliminary phytochemical screening to correlate its

chemical constituents with the observed antioxidant activity [7]. This investigation seeks to provide a scientific basis for the potential therapeutic application of *C. ovata* as a natural antioxidant source [8].

Figure 1 depicts a high-resolution botanical illustration of *Crassula ovata* (jade plant), characterized by thick, fleshy, oval-shaped leaves arranged in compact

clusters [9]. The leaves display a glossy green surface with distinctive reddish margins, reflecting typical morphological adaptations of succulent plants for water storage. The illustration highlights leaf texture, venation, and natural coloration, providing a clear visual representation suitable for academic and educational purposes [10].



Figure 1: *Crassula ovata* Leaf.

2. Materials and Methods

2.1 Plant Material and Extraction

Fresh leaves of *C. ovata* were washed, shade-dried, powdered, and subjected to solvent extraction using ethanol. The extract was concentrated and stored for further analysis.

2.2 Phytochemical Screening

The plant extract was subjected to qualitative phytochemical screening to identify the presence of major classes of secondary metabolites [11]. Standard phytochemical tests were employed to detect alkaloids, steroids, resins, saponins, carbohydrates, flavonoids, and glycosides [12]. The identification of these phytoconstituents provides preliminary insight into the chemical composition of the extract and suggests its potential for diverse biological activities [13]. The presence of multiple bioactive compound classes supports the rationale for further pharmacological evaluation and correlation with the observed antioxidant activity [14], [15].

2.3 DPPH Radical Scavenging Assay

The antioxidant activity of the plant extract was evaluated using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay [16]. Different concentrations of the extract were prepared and allowed to react with a freshly prepared DPPH solution (0.1 mM) [17], [18]. The reaction mixtures were incubated in the dark at room temperature to prevent photo-degradation of the DPPH radical [19]. After the incubation period, the absorbance of each sample was measured at 517 nm using a UV-visible spectrophotometer [20], [21]. The percentage of DPPH

radical scavenging activity was calculated using the following equation:

$$\text{Percentage inhibition} = \frac{(A_0 - A_1)}{A_0} \times 100$$

Where A_0 denotes the absorbance of the control solution and A_1 represents the absorbance in the presence of the extract. The half maximal inhibitory concentration (IC₅₀), defined as the concentration of extract required to scavenge 50% of DPPH radicals, was determined from the concentration–response curve and used as an indicator of antioxidant potency [22], [23].

3. Results

3.1 Phytochemical Profile

Qualitative phytochemical screening of the plant extract revealed the presence of several bioactive secondary metabolites. Alkaloids, steroids, saponins, and resins were detected in appreciable amounts, while carbohydrates were present in trace quantities. Flavonoids were slightly detectable, and glycosides showed a mild presence in the extract. The detection of these phytoconstituents suggests a diverse chemical profile that may contribute to the biological activity of the extract. Many of these compounds, particularly flavonoids, alkaloids, and saponins, are known to exhibit antioxidant properties through electron donation, free radical scavenging, and modulation of oxidative pathways. The observed phytochemical composition therefore provides a plausible basis for the antioxidant potential demonstrated by the extract in subsequent assays.

3.2 DPPH Radical Scavenging

The plant extract exhibited a clear concentration dependent increase in DPPH radical scavenging activity. As the concentration of the extract increased, a corresponding reduction in DPPH absorbance was observed, indicating effective neutralization of free radicals. Graphical analysis demonstrated a progressive decline in absorbance values with

increasing extract concentration, reflecting decreased availability of DPPH radicals. The extract showed a moderate IC_{50} value, suggesting appreciable antioxidant potential. These findings confirm the ability of the extract to donate electrons or hydrogen atoms to stabilize free radicals and support its role as a natural antioxidant source.

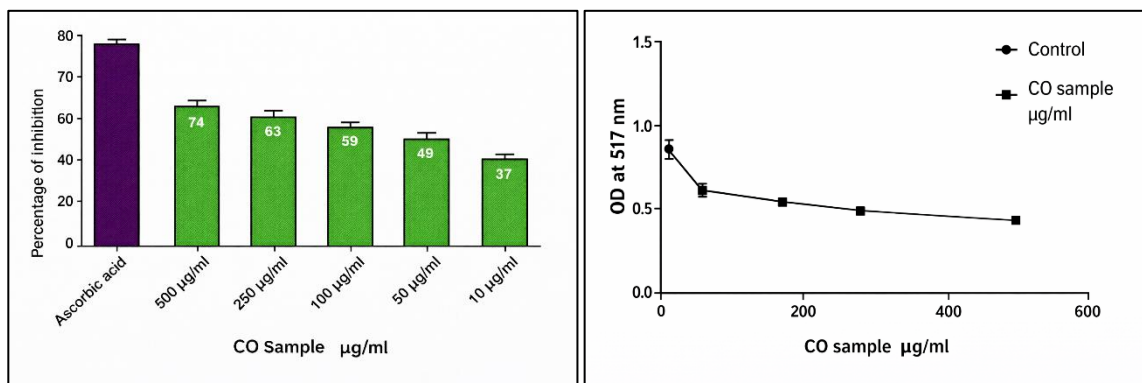


Figure 2: Dose-dependent antioxidant activity of the CO sample compared with ascorbic acid, showing percentage inhibition and corresponding reduction in OD at 517 nm.

Figure 2 illustrates the antioxidant activity of the CO sample evaluated using a free radical scavenging assay. The left panel shows the percentage of inhibition at different concentrations of the CO sample (10–500 µg/ml) compared with the standard antioxidant ascorbic acid, demonstrating a concentration-dependent increase in inhibitory activity. The highest inhibition was observed at 500 µg/ml, while lower concentrations exhibited reduced activity. The right panel depicts the corresponding decrease in optical density (OD) at 517 nm with increasing CO sample concentration, indicating effective free radical scavenging. Together, these results confirm the dose-dependent antioxidant potential of the CO sample in comparison to the reference standard.

4. Discussion

The antioxidant activity of *Crassula ovata* observed in the present study can be attributed to the presence of bioactive phytochemicals identified during qualitative screening. Secondary metabolites such as alkaloids, steroids, saponins, and flavonoids are known to participate in hydrogen donation and electron transfer mechanisms, which play a crucial role in neutralizing reactive free radicals. These compounds may act individually or synergistically to enhance the overall antioxidant capacity of the extract [24].

The DPPH radical scavenging assay, a widely accepted and reliable method for evaluating antioxidant activity, demonstrated that the extract possesses measurable free radical neutralizing potential. The concentration dependent increase in DPPH inhibition observed in this study indicates that higher levels of phytoconstituents enhance radical scavenging efficiency. The moderate IC_{50} value further supports the ability of the extract to effectively interact with and stabilize free radicals, thereby reducing oxidative stress [25].

The findings of this investigation are in agreement with

previous studies reporting antioxidant activity in members of the Crassulaceae family, which are rich in diverse secondary metabolites. The observed antioxidant potential of *C. ovata* suggests its possible application as a natural source of antioxidants. However, further studies involving quantitative phytochemical analysis, identification of active constituents, and *in vivo* antioxidant evaluation are necessary to substantiate its therapeutic relevance and to elucidate the underlying mechanisms of action [26].

Conclusion

The ethanolic extract of *Crassula ovata* demonstrated notable *in vitro* antioxidant activity, as evidenced by its concentration dependent DPPH radical scavenging capacity and supportive phytochemical profile. The presence of bioactive secondary metabolites such as alkaloids, steroids, saponins, and flavonoids likely contributes to its free radical neutralizing potential. These findings suggest that *C. ovata* may represent a valuable natural source of antioxidant compounds with potential applications in the development of pharmaceutical or nutraceutical formulations. Further studies involving quantitative phytochemical characterization, isolation of active constituents, and *in vivo* antioxidant evaluation are warranted to validate its therapeutic potential and ensure its safety and efficacy.

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Author Contribution

SN; Conceptualization of the review, **VA;** Literature survey, **SK;** Manuscript drafting, **BS;** Supervision and final approval.

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Ethical Approval

Not applicable

Data Availability

The data generated and analyzed during this study are available from the corresponding author upon reasonable request.

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Conflicts of Interest

There are no conflicts of interest

Declarations

The authors declare that they used AI language tools (ChatGPT and Grammarly Premium) to enhance this manuscript's linguistic clarity and readability. They carefully reviewed and edited all generated text to ensure accuracy and alignment with the research's intended meaning.

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