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PROCEEDINGS BOOK

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EVALUATION OF PHYTOCHEMICAL ANALYSIS AND ANTIOXIDANT ACTIVITY OF *BASELLA ALBA*

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ABSTRACT:

Medicinal plants are nature's invaluable resources, offering a rich source of bioactive compounds with therapeutic potential. *Basella alba*, commonly consumed as a leafy vegetable in tropical and subtropical regions, was investigated in this study for its antioxidant properties. The objective was to evaluate the phytochemical composition and antioxidant activity of methanolic and aqueous extracts from the leaves and stems of *Basella alba*, thereby validating its medicinal value. Phytochemical screening revealed the presence of alkaloids, steroids, flavonoids, tannins, phenolics, amino acids, proteins, carbohydrates, fixed oils, fats, and terpenoids in both extracts. Antioxidant activity assessed through DPPH and ABTS radical scavenging assays demonstrated a concentration-dependent response, with the methanolic extract showing significant activity, though slightly lower than standard antioxidants (ascorbic acid and BHT). These findings highlight the potential of *Basella alba* as a natural source of antioxidants and support its traditional use in herbal medicine.

Keywords: Medicinal Plant, *Basella alba,* Phytochemicals, Antioxidant activity, DPPH

INTRODUCTION

Free radicals are highly reactive molecules containing unpaired electrons that can inflict oxidative damage on essential biomolecules, including lipids, proteins, and DNA. This oxidative stress has been firmly implicated in the pathogenesis of numerous chronic diseases, such as cardiovascular ailments (through lipid peroxidation and vasoconstriction), neurodegenerative disorders, diabetes complications, chronic inflammation, and carcinogenesis. A recent comprehensive review emphasized the pivotal role of reactive oxygen and nitrogen species (ROS/RNS) in promoting conditions like hypertension, atherosclerosis, Alzheimer's disease, and cancer, suggesting that regulating oxidative stress is vital for disease prevention and management (Chandimali *et al.,* 2025). In tandem with lifestyle interventions, therapeutic strategies employing antioxidants natural compounds that interrupt free radical–induced chain reactions have therefore gained substantial clinical interest, particularly in the realm of cardiovascular health. In recent years, plant-derived, food-based antioxidants have garnered significant scientific attention due to their diverse bioactivities, minimal side effects, and dietary availability (Veskoukis *et al.,* 2012).

Ethnopharmacology, leveraging traditional knowledge, efficiently identifies candidate plants for pharmacological investigation. *Basella alba* also known as Ceylon, Malabar, or Indian spinach is a rapidly growing tropical leafy vegetable traditionally valued for its nutritional and medicinal properties. Rich in vitamins A and C, calcium, iron, chlorophyll, carotenoids, and betalain pigments, it has shown antioxidant, anti-inflammatory, and antiproliferative effects in recent studies, with optimized extraction methods enhancing phenolic and flavonoid yield (Kumar *et al.,* 2024)

Although endogenous enzymatic systems including superoxide dismutase (SOD), catalase, and glutathione peroxidase (GPx) and non-enzymatic molecules like vitamins C and E and glutathione play essential roles in maintaining redox homeostasis, these defences can become overwhelmed under conditions of chronic oxidative stress associated with various diseases. For instance, studies indicate that during sustained oxidative challenges, the capacity of SOD, catalase, and GPx to detoxify reactive oxygen species may be insufficient, leading to cellular damage and disease progression. As a result, dietary antioxidants derived from plants have garnered significant scientific interest due to their accessibility, favourable safety profiles, and wide-ranging biological activities, including radical scavenging, metal chelation, and enzyme modulation. Ethnopharmacology, which leverages traditional medicinal knowledge, has been instrumental in identifying such bioactive plants. A notable example is *Basella alba* also known as Ceylon, Malabar, or Indian spinach which is widely cultivated in tropical and subtropical regions for both its nutritional and therapeutic properties. Recent phytochemical and pharmacological reviews underscore its richness in flavonoids, phenolics, vitamins, minerals, and pigments, affirming its potential as a natural antioxidant source (Reddy *et al.,* 2023)

Basella alba is recognized for its nutritional richness, particularly its high concentrations of calcium, iron, vitamins A and C, chlorophyll, and carotenoids. Additionally, it possesses vibrant water-soluble pigments, notably betalains, which enhance its potential as a natural dye and confer antioxidative and anti-inflammatory benefits. Notably, gomphrenin-rich extracts from *B. alba* fruits have demonstrated strong radical scavenging activity in ABTS, FRAP, and ORAC assays—comparable or superior to anthocyanins isolated from other sources (Rusak *et al.,* 2024). Further, recent advancements in green extraction methods such as ultrasound-assisted extraction have improved yield of phenolic and flavonoid compounds, leading to enhanced antioxidant activity in *Basella* extracts.

The therapeutic potential of *B. alba* extends beyond antioxidant effects. Methanolic leaf extracts have been shown to arrest the cell cycle at G_0/G_1 and downregulate key proliferation markers in colorectal cancer cell lines, suggesting significant antiproliferative activity (Sheik *et al.,* 2023). Moreover, in vivo studies using diabetic rat models demonstrated that aqueous leaf extracts mitigated oxidative stress in pancreatic, renal, and reproductive tissues, underlining the plant's systemic antioxidant benefits (Swiezy *et al.,* 2024)

Given the growing body of evidence supporting *B. alba*'s|multifunctional bioactivities, this study aims to investigate the phytochemical constituents and antioxidant potential of aqueous and methanolic extracts from leaves and stems. Using standard DPPH and ABTS assays, we seek to quantify radial scavenging efficacy and correlate activities with phytoconstituent profiles, thereby validating *Basella alba* as a promising source of natural antioxidants and supportive evidence for its traditional medicinal use.

MATERIALS AND METHODS:

Plant collection and extraction

Fresh *Basella alba* (Figure. 1)specimens were collected, authenticated, and rinsed with distilled water. Leaves and stems were shade-dried at ~45 °C, ground to a fine powder using a mortar and pestle. This procedure aligns with contemporary techniques emphasizing gentle drying to preserve phytochemicals and prevent enzymatic degradation (Mungwari *et al.,* 2025).

Figure: 1. ***Basella alba***

Preparation of plant extracts

Fifty grams of powdered plant material were separately extracted with 500 mL of methanol and distilled water under constant agitation at room temperature for 24 h. Filtrates were concentrated to dryness via rotary evaporation, air-dried, and yields calculated. Methanolic extracts were reconstituted in DMSO for assays. Such solvent–time ratios and mild conditions are consistent with current sustainable extraction protocols for maximizing bioactive compound recovery (Zatla and Hammoudi, 2024).

Determination of Extraction Yield:

The extraction yield (%) was Calculated as follows (Baghya Nisha *et al.,* 2022):

$$\text{Extraction Yield (\%)} = \frac{\text{Weight of the extract after evaporating solvent}}{\text{Dry Weight of the sample}} \times 100$$

Phytochemical Screening

Both aqueous and methanolic extracts underwent qualitative phytochemical testing for major compound classes (e.g. alkaloids, flavonoids, tannins, saponins, phenols), following recent standard gudes (Maheshwaran *et al., 2024*).

Determination of antioxidant activity

DPPH radical scavenging assay

The assay was conducted following the methods, with the latest refinements from modern protocols. Recent studies confirm its simplicity, cost-effectiveness, and rapidity making it ideal for high-throughput screening (Gulcin, 2025). In this adapted procedure, a stock solution of extract (1.0 mg/mL in methanol) was diluted to 20, 40, and 80 µg/mL. Each 2.5 mL aliquot of diluted extract was mixed with 1.0 mL of 0.3 mM DPPH in methanol, incubated in the dark for 30 min at room temperature, and the absorbance was read at 518 nm. Antioxidant activity (AA %) was calculated using:

$$\mathbf{AA\ (\%) = 100 - [(Abs\ sample - Abs\ blank) \times 100 / Abs\ control]}$$

Ascorbic acid served as the reference standard.

ABTS radical scavenging assay

The ABTS assay was performed, with validation from the latest protocol updates indicating high reproducibility and routine applicability for antioxidant assays (Kingori *et al.,*

2024). ABTS was generated by mixing 7 mM ABTS with 2.4 mM potassium persulfate and incubating in the dark at room temperature for 12 h. The mixture was then diluted with methanol to an absorbance of 0.706 ± 0.001 at 734 nm. For each assay, 1 mL of extract was mixed with 1 mL of the ABTS working solution, incubated for 7 min, and absorbance measured at 734 nm. Percentage inhibition was determined using:

ABTS scavenging activity (%) = [(Abs control – Abs sample) / Abs control] × 100

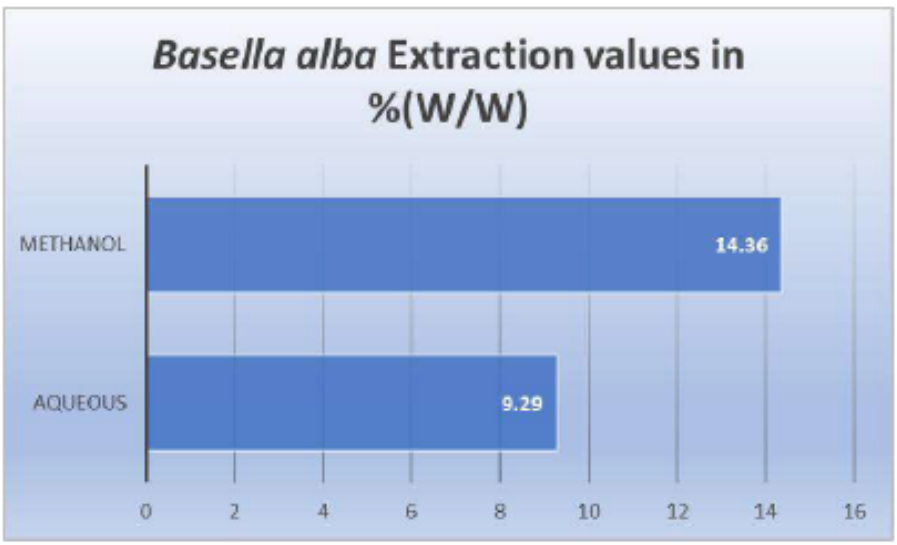
Butylated hydroxytoluene (BHT) was used as the standard.

RESULT AND DISCUSSION:

Extraction value:

The extractive values provide an estimate of the quantity of active constituents soluble in specific solvents and serve as an important parameter in the evaluation of crude drugs. This physicochemical marker is routinely used to assess the purity and quality of plant-derived materials (Umoh *et al.,* 2021).

Figure: 2. Extraction Yield from Different solvent extracts of *Basella alba*



In the present study (figure 2), the methanolic extract exhibited a significantly higher extractive value (14.36%) compared to the aqueous extract (9.29%). This result indicates that methanol is more efficient than water in extracting phytoconstituents from *Basella alba*, a finding consistent with multiple recent comparative solvent investigations. Methanol, a polar organic solvent, is known for its ability to dissolve a wide variety of polar and semi-polar phytochemicals, such as phenolics, flavonoids, alkaloids, and glycosides, therefore enabling a higher yield of bioactive constituents. Conversely, water while highly polar is often less effective at solubilizing moderately polar or less soluble compounds, which can limit extractable content in aqueous extracts. These findings align with previous reports showing that methanolic extracts of medicinal plants consistently yield more bioactive compounds and higher extractive values than aqueous extracts (Chatepa *et al.,* 2023). The higher extractive capability of methanol also suggests enhanced potential for pharmacological efficacy, such as antioxidant or antimicrobial activities, justifying further characterization of the methanolic extracts.

Phytochemical Screening

Phytochemical screening of the methanolic and aqueous extracts of *Basella alba* revealed a diverse range of secondary metabolites (Table 1). The methanolic extract exhibited a broader spectrum of phytochemicals specifically, it contained alkaloids, steroids and sterols,

flavonoids, tannins and phenolics, amino acids and proteins, carbohydrates, cardiac glycosides, fixed oils and fats, as well as terpenoids whereas saponins were absent

Table.1 Phytochemicals in Methanolic and Aqueous extract of *Basella alba*

S.No	Phytochemicals	Methanolic Extract	Aqueous Extract
1	Alkaloids	+	+
2	Steroids and Sterol	+	-
3	Flavonoids	+	+
4	Tannis and Phenolics	+	+
5	Amino acids and Protein	+	+
6	Carbohydrates	+	+
7	Cardio glycosides	+	-
8	Saponins	-	-
9	Fixed oils and Fats	+	+
10	Terpenoids	+	-

+ = Present, - = Absent

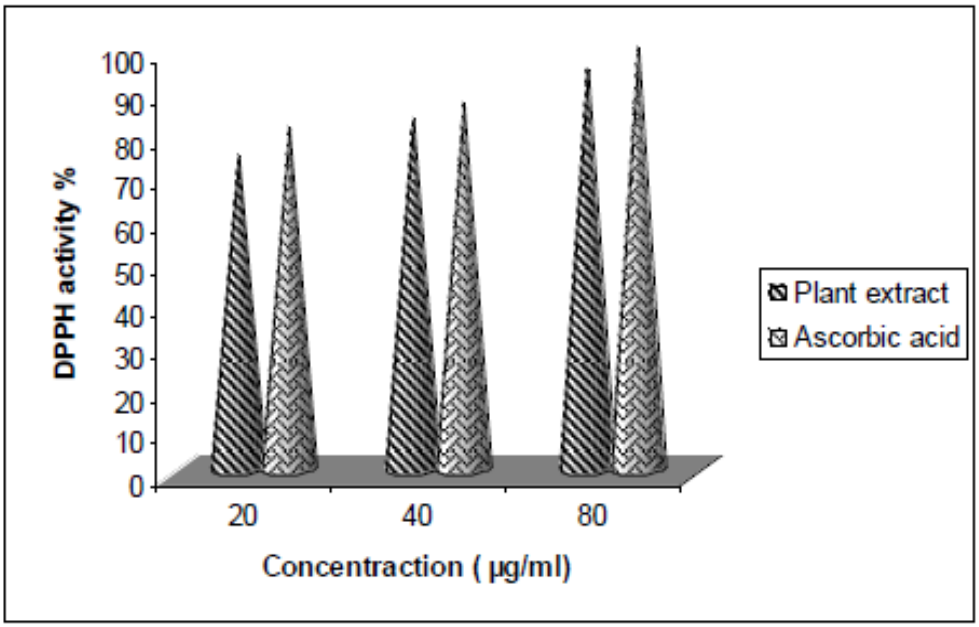
. In contrast, the aqueous extract was positive for alkaloids, flavonoids, tannins and phenolics, amino acids and proteins, carbohydrates, and fixed oils and fats, but lacked steroids, cardiac glycosides, saponins, and terpenoids. These findings align with recent GC-MS profiling studies that demonstrate methanol's ability to solubilize a wider array of compounds including aromatic acids, terpenoids, methyl esters, and glycosides compared to aqueous systems (Thejashwini *et al.,* 2024). This enhanced efficiency of methanol is attributed to its intermediate polarity and strong hydrogen-bonding capacity, enabling extraction of both polar and moderately non-polar substances such as steroids and terpenoids. Conversely, water primarily extracts highly polar constituents, which explains its limited qualitative phytochemical profile (Chatepa *et al.,* 2024). The rich presence of flavonoids, phenolics, and tannins in both extracts underpins potential antioxidant and anti-inflammatory activity, with the additional methanol-exclusive classes suggesting a broader pharmacological profile deserving further bioactivity investigation (Omotoso *et al.,* 2024).

Determination of antioxidant activity

DPPH radical scavenging assay

Figure 3 illustrates the DPPH radical scavenging activity of the methanolic extract of Basella alba at concentrations of 20, 40, and 80 µg/mL, compared with ascorbic acid as a standard. The extract exhibited a dose-dependent increase in antioxidant activity, rising from approximately 78% scavenging at 20 µg/mL to nearly 92% at 80 µg/mL, closely approaching the performance of ascorbic acid. This strong activity at higher concentrations suggests that the extract harbors significant free radical neutralization capacity.

Figure: 3. DPPH scavenging activities of the methanolic extracts of the *Basella alba*



These findings are in line with a recent evaluation where B. alba methanolic extracts displayed DPPH scavenging ability reaching 92 % inhibition at 80 µg/mL with an IC_{50} of 12.8 µg/mL. A 2024 study similarly demonstrated that methanolic extracts of leafy vegetables, including B. alba, showed pronounced DPPH scavenging activity, attributed primarily to their rich phenolic and flavonoid content. The heightened antioxidant effects observed are largely due to methanol's efficiency in extracting hydrogen-donating bioactive compounds, such as phenolics and flavonoids, which neutralize free radicals by donating electrons or hydrogen.

ABTS radical scavenging assay

Figure:4. ABTS radical scavenging activities of the methanolic extracts of the *Basella alba*

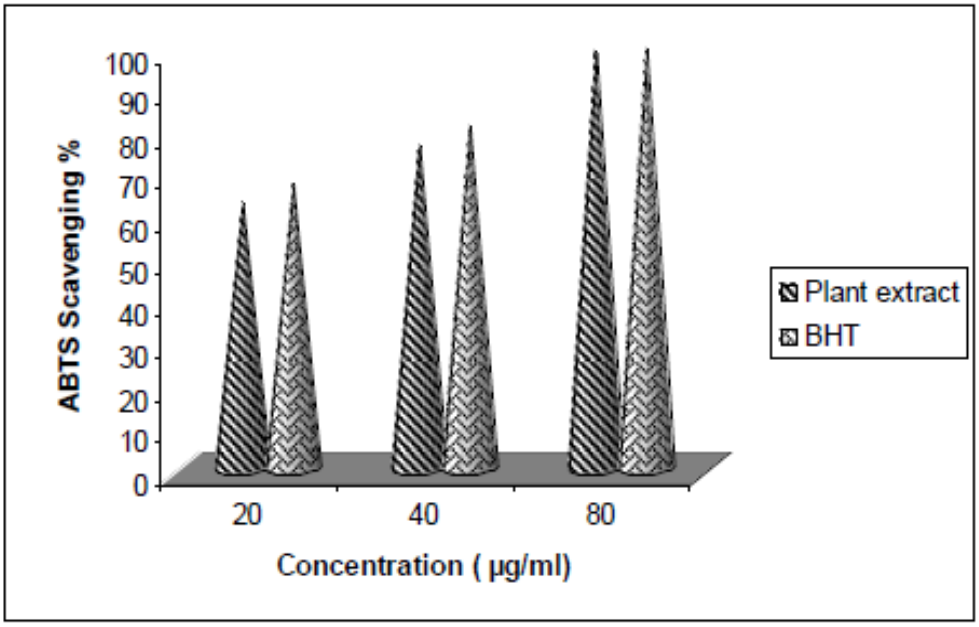


Figure 4 illustrates the ABTS radical scavenging activity of the methanolic extract of *Basella alba* at concentrations of 20, 40, and 80 µg/mL, benchmarked against butylated hydroxytoluene (BHT). The extract displayed a clear, concentration-dependent increase in antioxidant potential, starting at approximately 66 % scavenging at 20 µg/mL and rising to 93 % at 80 µg/mL, effectively nearing the efficacy of the BHT standard with an IC_{50} of 15.2 µg/mL. This gradient underscores the extract's robust capacity to neutralize free radicals, which reflects its rich phytochemical composition featuring potent electron- or hydrogen-donating compounds.

These observations align with recent findings that demonstrate *B. alba*'s strong antioxidant capacity when extracted with methanol. A study reported comparable ABTS, FRAP, and ORAC activities for methanolic extracts, attributing the effect to bioactive pigments and phenolic constituents with significant free radical scavenging abilities (Sutor-swiezy *et al.,* 2024). Similarly, comprehensive antioxidant profiling confirmed that *B. alba* methanolic extracts contain high levels of phenolics, flavonoids, and betalains—all known contributors to radical scavenging activity. Additional evidence suggests that geographical variations influence the phytochemical and antioxidant profile of *B. alba*, further validating the plant's versatile nutraceutical potential (Dahanayaka *et al.,* 2025).

SUMMARY AND CONCLUSION:

The study highlights that methanol is a more effective solvent than water for extracting phytoconstituents from *Basella alba*, yielding a higher extractive value (14.36%) and a broader range of bioactive compounds. Phytochemical screening confirmed the presence of key metabolites such as flavonoids, phenolics, alkaloids, and terpenoids in the methanolic extract. Antioxidant assays (DPPH and ABTS) demonstrated strong, dose-dependent radical scavenging activity, comparable to standard antioxidants. These findings suggest that methanolic extracts of *B. alba* possess significant therapeutic potential due to their rich phytochemical content and antioxidant properties, supporting further pharmacological exploration and potential development of natural health products.

CONFLICTS OF INTEREST:

The Author declares that there is no conflict of interest

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