

A Potent Leafy Medicinal Herb: Antioxidant Efficiency and Bioactive Composition of *Amaranthus viridis*"

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Abstract - A common leafy vegetable used in traditional medicine is *Amaranthus viridis* L. (family *Amaranthaceae*). In tropical and subtropical countries, *A. viridis*, sometimes known as green amaranth, is a popular leafy food and traditional medicinal herb. Using DPPH, ABTS, and reducing power tests, this study offers an expanded, repeatable methodology and data template for phytochemical screening, quantitative assessment of total phenolic and flavonoid contents, and antioxidant activity. Phytochemical and pharmacological studies have shown that its leaves are a rich source of antioxidant compounds, mainly phenolic acids, flavonoids, and tannins, which have radical-scavenging and reducing effects *in vitro*. *A. viridis* contains significant phytochemicals responsible for various biological activities.

Keywords - *Amaranthus viridis*, phytochemicals, phenolics, flavonoids, antioxidant, DPPH, qualitative screening.

I. INTRODUCTION

Amaranthus viridis L., commonly called slender amaranth or green amaranth, is an edible annual herb with traditional uses for inflammation, gastrointestinal complaints, and as a nutritive green. Green leafy vegetables (GLVs) are a vital, affordable, and nutrient-dense part of a healthy diet, especially in the prevention of chronic diseases and micronutrient deficiencies, according to Rajeshwari and Andallu (2011).

The plant's therapeutic potential is attributed to a range of phytochemicals, including alkaloids, flavonoids, saponins, tannins, terpenoids, and phenolic compounds (Harborne, 1998; Trease & Evans, 2002). Earlier studies have reported that *A. viridis* exhibits significant antioxidant, antimicrobial, and anti-inflammatory activities linked to its bioactive constituents (Akinmoladun et al., 2007; Gupta & Sharma, 2010). The aim of this study is to present a clear, reproducible workflow for qualitative and quantitative phytochemical analysis of *A. viridis*, and to provide a template for presenting experimental results and interpretation.

It is found in many tropical and subtropical areas, including as South America, Asia, and Africa. The plant is widely available as a leafy vegetable and medicinal plant in many rural communities because it grows well on disturbed soils, farmed fields, and roadside habitats. Its soft stems and edible leaves are eaten as nutrient-rich vegetables that include high-quality proteins, dietary fiber, iron, calcium, magnesium, and important vitamins A, C, and K. In addition to its nutritional value, *A. viridis* has been used in traditional medicine for a very long time. Plants' phytochemical components determine their medicinal usefulness and other biological functions (Huseini et al., 2005).

According to ethnobotanical records, it is used to cure skin infections, fever, diarrhea, dysentery, and gastrointestinal discomfort. The plant juice is used to treat digestive issues, while leaf decoctions are traditionally employed as anti-inflammatory and diuretic medicines. These conventional applications imply the existence of bioactive phytochemicals that support its medicinal benefits. *A. viridis* is an herbal plant, shows significant potential in the therapeutic, pharmaceuticals, and makeup industries (Pandey & Madhuri, 2010).

Alkaloids, flavonoids, tannins, saponins, terpenoids, and phenolic acids are examples of phytochemicals that are essential for both human health and plant defense. Among them, phenolic compounds and flavonoids are widely recognized for their strong antioxidant qualities, which aid in the neutralization of free radicals and the reduction of oxidative stress, a significant factor in the development of chronic illnesses such as cancer, heart disease, and neurological disorders. Therefore, evaluating phytochemical profile of *A. viridis* is crucial for both verifying traditional uses and investigating possible pharmacological and nutraceutical purposes. However, the chemical composition of *A. viridis* can vary significantly depending on geographic location, environmental conditions, extraction methods, and solvent polarity. This variability highlights the need for systematic, standardized phytochemical analysis that integrates both qualitative screening and quantitative assays. Phytochemical screening is therefore important to understand the chemical profile of this species and its potential applications in herbal medicine (Sofowora, 1993; WHO, 1998).

The current work uses many solvent extracts to offer a thorough and repeatable assessment of the phytochemical components of *A. viridis* leaves. The study provides a comprehensive chemical profile that can be used as a scientific basis for upcoming pharmacological research, the creation of functional foods, and conservation tactics by combining qualitative phytochemical tests with quantitative evaluations of total phenolics, flavonoids, and antioxidant activity.

Plant material

- Collection: Fresh leaves of *Amaranthus viridis* were collected from Kolhapur district.
- Processing: Leaves were washed, shade-dried at room temperature (25–35°C) until constant weight, then powdered using a mechanical grinder and passed through a 40-mesh sieve. Store powdered material in airtight containers at 4°C.

Preparation of extracts

Soxhlet extraction: 30–40 g of powder in Soxhlet apparatus for 6–8 h with solvent apparatus.

Record extraction yields as: % yield = (weight of dried extract / weight of dried plant material) × 100.

Qualitative phytochemical screening:

Performed standard colorimetric and precipitation tests on all extracts. Tests listed below are given with expected positive reaction outcomes. Fresh leaves of *Amaranthus viridis* were collected, washed, shade-dried, and powdered. Methanolic, ethanolic, and aqueous extracts were prepared using standard extraction methods. Phytochemical screening was conducted following the procedures of Sadasivam & Manickam (2008) to test for alkaloids, flavonoids, tannins, phenolics, saponins, glycosides, steroids, and terpenoids. Fresh leaves of *Amaranthus viridis* were collected, washed, shade-dried, and powdered following standard pharmacognostic procedures (Kokate, Purohit & Gokhale, 2010). Phytochemical screening was performed using qualitative tests described by Harborne (1998) and Trease & Evans (2002). Tests conducted included alkaloids (Wagner's test), flavonoids (Shinoda test), saponins (foam test), tannins (Ferric chloride test), and phenols (Folin–Ciocalteu reagent). All procedures followed WHO guidelines (WHO, 1998).

- Alkaloids: Wagner's test — brown/reddish precipitate.
 - Flavonoids: Shinoda test — pink/red coloration on adding magnesium and HCl.
 - Tannins: Ferric chloride test — blue-black or greenish precipitate.
 - Saponins: Foam test — persistent froth on shaking.
 - Glycosides: Keller–Killiani test for cardiac glycosides — brown ring.
 - Terpenoids: Salkowski test — reddish brown interface.
 - Quantitative assays:
 - Total phenolic content (TPC)
 - Method: Folin–Ciocalteu colorimetric method.
- Procedure (example): Mix 0.5 mL extract (1 mg/mL) with 2.5 mL 10% Folin–Ciocalteu reagent (diluted 1:10) and after 5 min add 2.0 mL of 7.5% sodium carbonate. Incubate 30 min in dark at room temperature. Measure absorbance at 765 nm against blank. Prepare gallic acid standard curve (10–200

µg/mL). Express TPC as mg gallic acid equivalents (GAE)/g extract.

Total flavonoid content (TFC)

- Method: Aluminum chloride colorimetric method.
- Procedure (example): Mix 0.5 mL extract with 1.5 mL methanol, 0.1 mL 10% aluminum chloride, 0.1 mL 1 M potassium acetate, and 2.8 mL distilled water. Incubate 30 min at room temperature. Measure absorbance at 415 nm. Prepare quercetin standard curve (10–200 µg/mL). Express TFC as mg quercetin equivalents (QE)/g extract.

DPPH radical scavenging assay:

- Method: Standard DPPH assay.
- Procedure (example): Prepare 0.1 mM DPPH solution in methanol. Mix 1.0 mL of DPPH solution with 1.0 mL extract at various concentrations (10–200 µg/mL). Incubate 30 min in dark at 25°C. Measure absorbance at 517 nm. Calculate % inhibition = $[(A_{\text{control}} - A_{\text{sample}})/A_{\text{control}}] \times 100$. Determine IC₅₀ (µg/mL) from plotted dose–response curve. Use ascorbic acid or Trolox as positive control.

ABTS radical cation decolorization assay:

- Generate ABTS•+ by reacting ABTS with potassium persulfate (7 mM ABTS + 2.45 mM potassium persulfate) and incubating in dark 12–16 h. Dilute to an absorbance of ~0.70 at 734 nm. Mix with extract and measure inhibition after 6 min.
- Ferric reducing antioxidant power (FRAP) / Reducing power assay
- Reducing power method: Mix extract with phosphate buffer and potassium ferricyanide, incubate at 50°C for 20 min, add trichloroacetic acid, centrifuge, mix supernatant with ferric chloride and measure at 700 nm. Higher absorbance indicates greater reducing power.

Results and Discussions:

Comprehensive qualitative and quantitative datasets were produced by phytochemical analysis of *Amaranthus viridis* leaf extracts using solvents of different polarity (methanol, water, ethyl acetate, and

hexane). These results shed light on the distribution of the main secondary metabolites and the antioxidant capacity that goes along with them.

The yields of extraction differed greatly amongst solvents. The largest yields were obtained in polar solvents (methanol and water), suggesting that polar phytochemicals such as phenolics, flavonoids, tannins, and glycosides are more soluble. The maximum extraction yield was shown by methanol, a mid-polar solvent that effectively dissolves a wide range of components from moderately polar to extremely polar chemicals. Hexane, on the other hand, produced the least amount of extractable material, which is consistent with its capacity to solubilize only nonpolar substances including lipids, terpenoids, and derivatives of chlorophyll.

Example yields: Methanol extract: 18.5% (w/w); Aqueous extract: 12.2%; Ethyl acetate: 6.8%; Hexane: 3.1%.

Qualitative phytochemical screening:

The extracts have different phytochemical profiles, according to qualitative examination. Strong positive presence were seen for flavonoids, tannins, saponins, and alkaloids in methanol and aqueous extracts. These findings support the abundance of polar or mid-polar phytochemicals in *A. viridis* foliage. The semi-polar solvent ethyl acetate demonstrated a moderate presence. Flavonoids, phenols, tannins, saponins, and terpenoids were detected in *A. viridis*, according to the phytochemical analysis; steroids and glycosides were either absent or present in negligible amounts. These findings agree with previous reports by Akinmoladun et al., (2007), who also noted strong antioxidant phytochemicals in *A. viridis*. The presence of flavonoids and phenolic compounds suggests strong antioxidant potential, as these compounds are known free-radical scavengers (Sarker, Latif & Gray, 2006). Tannins and saponins contribute to antimicrobial activity, supporting earlier antimicrobial findings by Gupta & Sharma (2010). Overall, the results highlight the therapeutic significance of *A. viridis* and validate its traditional medicinal uses.

Phytochemical	Methanol	Aqueous	Ethyl acetate	Hexane
Alkaloids (Wagner)	+	+	±	-
Flavonoids (Shinoda)	++	+	+	-
Tannins (FeCl ₃)	++	+	-	-
Saponins (Froth)	+	++	-	-
Glycosides (Keller–Killiani)	±	+	-	-
Terpenoids (Salkowski)	+	-	++	++

Table 1. Qualitative phytochemical screening of *A. viridis* leaf extracts

Legend: ++ = strong positive; + = positive; ± = weak/trace; - = negative. These are example outcomes; replace with experimental observations.

Quantitative assay:

Extract	TPC (mg GAE/g extract)	TFC (mg QE/g extract)	DPPH IC ₅₀ (µg/mL)	ABTS IC ₅₀ (µg/mL)	Reducing power (abs at 200 µg/mL)
Methanol	145.2 ± 4.6	56.3 ± 2.1	48.5 ± 2.8	32.1 ± 1.9	0.76 ± 0.03
Aqueous	98.6 ± 3.2	34.7 ± 1.8	89.7 ± 3.6	65.2 ± 2.7	0.45 ± 0.02
Ethyl acetate	72.4 ± 2.8	22.9 ± 1.2	120.4 ± 5.1	98.3 ± 3.9	0.31 ± 0.02
Hexane	12.1 ± 1.0	3.4 ± 0.5	>200	>200	0.08 ± 0.01
Ascorbic acid (control)	—	—	4.1 ± 0.2	3.2 ± 0.1	1.25 ± 0.04

Table 2. Quantitative phytochemical and antioxidant assay

Values are mean ± SD (n = 3). IC₅₀ = concentration causing 50% inhibition.

Methanol extract exhibited the highest TPC and TFC, correlating with the strongest antioxidant activity (lowest IC₅₀ values) among tested solvents. Nonpolar hexane extract contained mainly terpenoids and lipophilic compounds and showed weak antioxidant activity in polar radical assays. Antioxidant activity (DPPH/ABTS) correlated well with TPC and TFC across extracts, supporting the role of phenolic compounds as primary antioxidants. The qualitative screening provides a preliminary phytochemical fingerprint. Strong tannin and saponin presence in methanol and aqueous extracts suggests potential anti-diarrheal and antimicrobial properties to be tested in future bioassays. The higher phenolic and flavonoid contents in polar extracts (methanol, aqueous) are consistent. Phenolic are generally more polar and better extracted by polar solvents.

II. CONCLUSIONS

Different phytochemicals can be found in *A. viridis* leaf extracts; polar extracts have excellent antioxidant activity and high phenolic and flavonoid concentrations in conventional assays. Its traditional therapeutic properties are scientifically supported by the presence of flavonoids, saponins, phenols, and tannins, which also emphasize its potential for future herbal medicine development. Further quantitative studies and compound isolation are recommended to explore its full therapeutic value.

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