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Review article

Phytochemical determination and evaluation of antioxidant potential of leaves of *Moringa concanensis*

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ABSTRACT

The present study investigated the phytochemical composition and antioxidant potential of leaves of Moringa concanensis Nimmo. Mature leaves were collected from Kota, Rajasthan, in March 2026 and authenticated at the Herbarium, Department of Botany, University of Rajasthan, Jaipur, under Authentication No. RUBL22127. The leaves were separately extracted with water, methanol and petroleum ether 1. The maximum extractive yield of methanolic extract was 246mg/g of dry weight, followed by aqueous extract (123mg/g) and petroleum ether extract (22mg/g). Preliminary phytochemical screening showed the presence of carbohydrates and flavonoids in all three extracts. Proteins, tannins and alkaloids were found in water and methanol extracts, and phytosterols were observed in all the extracts. The methanol and petroleum ether extracts contained saponins and terpenoids, whereas only the aqueous extract contained glycosides. DPPH free radical scavenging assay was performed to determine antioxidant activity at different concentrations (20-100 µg/ml). All extracts showed antioxidant activity in a concentration-dependent manner. Maximum activity was found in the methanolic extract at most concentrations tested (9.23 ± 1.71% to 25.73 ± 5.66% for 20 µg/mL to 100 µg/mL, respectively) with an IC 50 value of 210.15 µg/mL. The IC50 values of aqueous and petroleum ether extracts were 227.41 and 286.48 µg/mL, respectively. The results indicated that methanol was the best solvent among the solvents used for the extraction of phytochemicals from M. concanensis leaves with antioxidant activity and the presence of diverse phytochemical constituents. Further studies are required to identify the active compounds responsible for the observed activity.

Keywords: *Moringa concanensis*, Leaf Phytochemical, antioxidant.

INTRODUCTION

India is indisputably a sanctuary for herbal flora. Indian literature contains several references concerning the usage of herbal remedies for the treatment of various ailments. The reason why India is called the 'Botanical Garden' of the world is due to the thousands of herbal plants thriving in the country. India has utilised up to eight thousand plant species for health care initiatives. Diverse botanical extracts comprising different compounds can lead to synergistic effects. The collaborative effects of phytochemicals may provide various benefits due to their diverse chemical components, such as enhanced efficacy, reduced dosage requirements, and a decrease in side effects. The flora currently serves an essential role in various pharmaceuticals such as Allopathic medicine, Natural remedies, Homeopathy, and Aromatherapy. The flowers of many medicinal plants are used as condiments and food ingredients. Many plants are cheap and easily available to the common people. This vegetation has no negative effects after application [1].

Some of the plants have been studied by several researchers for therapeutic properties and other uses. Moringa has been a popular medicinal tree grown in recent years for its nutritious pods, edible leaves and flowers. It provides a number of components used in the preparation of pharmaceuticals, coloration and cosmetic oils, amongst others. It is also used as livestock feed and for water purification [2].

The angiosperm family Moringaceae consists of 13 diverse species of the single genus Moringa, ranging from small plants to large trees, found in tropical and subtropical areas. Moringa is a medium-sized tree, growing to 10-15 m in height. *Moringa concanensis* Nimmo is a deciduous tree species with hard bark. The bark, cracked to a depth of 10cm, is a corky grey and devoid of hair save for younger sections and flowering regions. The leaves exhibit a bipinnate structure and are 45 cm in length. The plant possesses various common names across over eight local languages [3].

The foliage of Moringa is utilized as a vegetable. The Moringa tree produces pods that are utilized in various parts of Rajasthan. It contains elevated concentrations of vitamins, antimicrobial agents, and anti-carcinogenic compounds. Moringa leaves, flowers, and seeds are typically consumable and ingested by humans. The roots, bark and flowers have antibacterial properties and are commonly used in traditional medicines of Indian and African cultures. The Moringa seed is a cost-effective and low-risk coagulant for human health and the environment. Desiccated Moringa seeds aggregate particulates in water owing to their bioactive soluble protein, which functions as a natural cationic polyelectrolyte. All components of Moringa are beneficial in treating ailments such as ascites, rheumatism, and poisonous stings [4].

Phytochemical studies on the genus Moringa in several countries have revealed the leaves to be particularly rich in minerals, vitamins, essential amino acids and known antioxidants.

Ancient kings used to eat moringa leaves and fruits to keep their brains sharp and skin healthy. The forefathers of India used Moringa leaf extract on the battlefield. The Elixir drink was also believed to give extra vitality and relieve stress during the conflict. Currently, Moringa species are receiving considerable attention because of their great economic potential.

The identification of new pharmaceuticals has been possible through traditional empirical and scientific methodologies. The normal way is based on things that have been shown to work by experiment over a number of years in different cultures and medical systems. With these concerns in view, the present study is undertaken to explore the therapeutic efficacy of *M. concanensis* Nimmo, which has been used for its biological activities since ages but is still under-explored in Rajasthan, India [5].

MATERIALS AND METHODS

Collection and authentication of plant samples

The mature leaves of *M. concanensis* were collected from Kota, Rajasthan, India in March 2026. The authentication of the plant was carried out at the Herbarium, Department of Botany, University of Rajasthan, Jaipur (Authentication No.- RUBL22127).

Processing of the collected plant part

The collected healthy leaves were washed with running tap water to remove dust and dirt and then with distilled water to remove ionic impurities. Leaves were dried in air. Air-dried leaves were ground to make coarse powder and stored for further use.

Extraction of the processed plant part

Plant materials (leaves) were extracted in different polar and non-polar solvents. Selected solvents were water (highly polar), methanol (mid-polar) and petroleum ether (non-polar). For this purpose, 1 gm of plant part was taken in a flask and dipped into 10 mL of the solvent individually. A total of 3 flasks were prepared for this purpose. They were kept in a sonicator at 40°C for 10 minutes. After that, they were filtered. The filtrate was air-dried by evaporating the solvent. After complete drying, extracts were collected in glass vials for further use.

Screening of phytochemicals in the plant extracts

Preliminary phytochemical screening of different selected plants was conducted to confirm the presence or absence of different phytochemicals such as proteins, carbohydrates, phenols, flavonoids, alkaloids, phytosterols, phlobatannins, tannins, saponins, cardiac glycosides and terpenoids using standard methods with minor modifications.

Protein test: Ninhydrin test

0.5 mg of extract was taken, and 2 drops of freshly prepared 0.2% ninhydrin reagent were added and heated. The appearance of pink/ purple/yellow colour indicated the presence of proteins, peptides or amino acids.

Carbohydrate test: Fehling's test

2 ml of Fehling's solution was added to 0.5 mg of extract and boiled in a water bath. The presence of red colour indicated the presence of reducing sugars, while the appearance of blue colour indicated their absence.

Phenols: FeCl3 test

Phenol is a hydroxyl group (-OH) on an aromatic ring, or simply the hydroxy derivatives of aromatic compounds are known as phenols. Phenols are weaker acids than carboxylic acids. It undergoes substitution reactions easily. Phenol is one of the most versatile and important industrial organic chemicals.

An aqueous solution of phenol reacts with freshly prepared ferric chloride solution to give a coloured complex. Most phenols give dark-coloured solutions.

The chemical reaction is given below.

$$6C6H5OH + FeCl3 \rightarrow [Fe(C6H5O)6]3– \text{ (violet colour complex)} + 3HCl + 3H+$$

Flavonoids- lead acetate test

1 ml of extract was taken, and a few drops of 10% lead acetate solution were added. Appearance of a yellow-coloured precipitate indicates the presence of flavonoids.

Alkaloids: Mayer's test

Mayer's reagent is an alkaloidal precipitating reagent used for the detection of alkaloids in natural products. Mayer's reagent is freshly prepared by dissolving a mixture of mercuric chloride (1.36 g) and potassium iodide (5.00 g) in water (100.0 mL). Most alkaloids are precipitated from neutral or slightly acidic solution by Mayer's reagent (potassium mercuric iodide solution) to give a precipitate.

Phytosterol test: Liebermann-burchard's test

0.5 mg extract was dissolved in 2 ml acetic anhydride, and 1-2 drops of concentrated sulphuric acid were added along the side of the tube. An array of colour changes (pink/purple/blue/green/brown) was observed.

Tannin test: gelatin solution+ NaCl test

Extracted 0.1 g of the sample with 10 mL of water, then boiled for several minutes. Then filtered and the filtration was added with 2 mL of a 1% gelatin solution containing NaCl. Tannins cause precipitation of gelatin in the solution. If a precipitate is formed, it indicates the presence of tannins and phenolic components.

Saponins: honeycomb test

0.5 mg of extract was taken in a test tube, and a few drops of 5% sodium bicarbonate solution were added. The mixture was shaken vigorously and kept for 3 min. Formation of honeycomb-like froth shows the presence of saponins.

Evaluation of antioxidant potential of the plant extracts

The DPPH free radical scavenging activity assay was used to assess the sample's antioxidant capacity. The 1,1-diphenyl-2-picrylhydrazyl (DPPH) technique was used to assess the extracted materials' capacity to scavenge DPPH radicals. 3.9 mL of 0.1 mM DPPH (methanolic) solution was used to combine 100 µl aliquots of substances in varying doses (20–100 µg/mL in DMSO). After that, the mixture was vortexed and allowed to sit in the dark for half an hour. Using methanol as a blank, its optical density (OD) was determined at 517 nm [6-9].

The radical scavenging activity was determined by the ratio = (Abcontrol – Absample / Abcontrol) × 100

Whereas Absample represents the absorbance of the DPPH solution with sample, Abcontrol displays the absorbance of the DPPH solution.

IC50 values were calculated by plotting a linear relationship between concentration and percentage inhibition. With the aid of an inhibition curve, the antioxidant capability of each sample was displayed as IC50, which is defined as the concentration required to stop the production of DPPH radicals by 50% [10,11].

RESULTS

Extraction of plant material

The extractive values of Moringa concanensis leaves are shown in Table 1. Three solvents were used for extraction. These were water, methanol, and petroleum ether.

The methanolic extract showed the highest extractive value. It was 246 mg/g dry weight. The extract was brown in colour. It had a powdery consistency. It took 7 days to dry. The aqueous extract showed an extractive value of 123 mg/g dry weight. It was light brown in colour. It was obtained as a powder after drying. It took 9 days to dry. The petroleum ether extract showed the lowest extractive value. It was 22 mg/g dry weight. The extract was yellowish-brown in colour. It had a sticky consistency. It took 2 days to dry. Among the three solvents, methanol gave the highest extractive yield. This indicates that methanol extracted more soluble components from the leaves of *Moringa concanensis*.

Table 1: Extractive values, extract colour and appearance in leaves of the selected plant

Solvent	Weight of extract (mg/g.dw)	Drying time (days)	Colour	Consistency
Water	123	9	Light brown	powder
Methanol	246	7	brown	powder
Pet ether	22	2	Yellowish-brown	sticky

Phytochemical determination of plant material

The phytochemical screening of *Moringa concanensis* leaves was carried out using water, methanol, and petroleum ether extracts. The results are presented in the table.

The test for carbohydrates was positive in all three extracts. A red colour was observed in the water, methanol, and petroleum ether extracts. Proteins were detected in the water and methanol extracts. A blue colour was observed in both extracts. The petroleum ether extract showed a negative result. Tannins were present in the water and methanol extracts. A green colour was observed. The petroleum ether extract showed a negative result. The test for alkaloids was positive in the water and methanol extracts. A red colour was observed. The petroleum ether extract showed a negative result.

Flavonoids were detected in all three extracts. A yellow colour was observed in the water, methanol, and petroleum ether extracts. Phytosterols were present in all three extracts. A brown colour was observed in each extract. The test for saponins was negative in the water extract. No frothing was observed. The methanol and petroleum ether extracts showed frothing and gave positive results. Terpenoids were detected in the methanol and petroleum ether extracts. Purple colour was observed in both extracts. The water extract showed a negative result. Phenols were found in water and methanol extracts but were absent in the petroleum ether extract.

Glycosides were detected only in the water extract

The results showed that the leaves of *Moringa concanensis* contained several important phytochemicals. Methanol showed the presence of most of the tested phytoconstituents. This indicates that

methanol may be a suitable solvent for extracting different bioactive compounds from the leaves.

Evaluation of plant extracts for their antioxidant potential

The antioxidant activity of *Moringa concanensis* leaf extracts was evaluated at different concentrations. The results are presented in Table 3.

All three extracts showed antioxidant activity at all tested concentrations. The activity increased with an increase in extract concentration. The water extract showed activity of 8.32 ± 2.47 at 20 µg/ml. It increased to 24.39 ± 2.88 at 100 µg/ml.

The methanolic extract showed activity of 9.23 ± 1.71 at 20 µg/ml. The activity increased to 25.73 ± 5.66 at 100 µg/ml. It showed the highest activity among the three extracts at most concentrations. The petroleum ether extract showed the lowest activity. Its activity increased from 6.49 ± 0.38 at 20 µg/ml to 19.46 ± 0.66 at 100 µg/ml. The results showed a concentration-dependent increase in antioxidant activity. The methanolic extract showed better antioxidant potential than the water and petroleum ether extracts. This may be due to the presence of antioxidant phytochemicals that are more effectively extracted by methanol [12, 13].

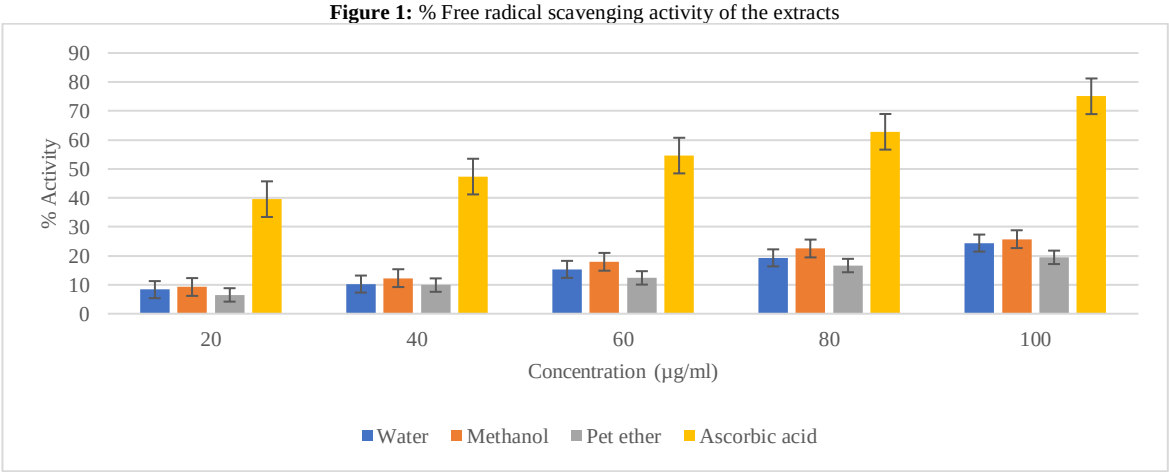
Table 2: Phytochemical determination of the extracts

Phytochemical test	Solvent	leaves	
		Observation	Result
Total carbohydrates (Fehling method)	Water	Red	+
	Methanol	Red	+
	Pet ether	Red	+
Protein (Ninhydrin)	Water	blue	+
	Methanol	blue	+
	Pet ether	brown	-
Tannins (FeCl3)	Water	Green	+
	Methanol	Green	+
	Pet ether	yellow	-
Alkaloids (Dragendorff's method)	Water	Red	+
	Methanol	Red	+
	Pet ether	yellow	-
Flavonoids (alkaline solution test)	Water	Yellow	+
	Methanol	Yellow	+
	Pet ether	Yellow	+
Phytosterols (Salkowski's test)	Water	Brown	+
	Methanol	Brown	+
	Pet ether	brown	+
Saponins (Honeycomb test)	Water	No frothing	-
	Methanol	frothing	+
	Pet ether	frothing	+
Terpenoids (Sofowora)	Water	pink	-
	Methanol	purple	+
	Pet ether	purple	+
Phenols	Water	Blue	+
	Methanol	Blue	+
	Pet ether	Layer	-
Glycosides	Water	Red	+
	Methanol	Blue	-
	Pet ether	Blue	-

Table 3: Antioxidant potential of extracts of various parts of the selected plant part

Solvent	20 µg/ml	40 µg/ml	60 µg/ml	80 µg/ml	100 µg/ml	IC50 value (µg/ml)
Water	8.32±2.47	10.24±2.60	15.31±1.67	19.29±2.12	24.39±2.88	227.41
Methanol	9.23±1.71	12.28±3.32	17.92±2.25	22.52±3.33	25.73±5.66	210.15
Pet ether	6.49±0.38	9.88±0.87	12.37±0.78	16.63±1.29	19.46±0.66	286.48
Ascorbic acid	49.55±0.59	67.34±0.99	74.59±0.55	82.80±0.41	85.06±0.74	9.43

Figure 1: % Free radical scavenging activity of the extracts



DISCUSSION

The extractive yield was different for all three solvents. The methanolic extract showed the highest yield of 246 mg/g dry weight. The aqueous extract showed a yield of 123 mg/g dry weight. The petroleum ether extract showed the lowest yield of 22 mg/g dry weight.

The higher yield with methanol suggests that more leaf material was soluble in this solvent. Methanol can pull out many different kinds of plant compounds. They may be phenolic compounds, flavonoids, tannins, alkaloids and other moderately polar compounds.

The aqueous extract gave a substantial yield as well. This indicates the presence of some water-soluble compounds in the leaves. Sugars, proteins and some phenolic compounds can be extracted by water.

Petroleum ether extract showed the lowest yield. This could be attributed to the lower number of compounds soluble in this non-polar solvent. Petroleum ether mainly extracts non-polar compounds, such as lipids, oils, and some other hydrophobic substances.

Phytochemical screening showed the presence of several important groups of compounds in the leaves of *Moringa concanensis*. The results were different for the three extracts. This implies that the solvent used for extraction affected the phytochemical profile. All three extracts were carbohydrate-containing. They were present in the methanol and petroleum ether extracts of the water. This indicates the presence of carbohydrate-related compounds in the leaf.

Proteins were identified in the water and methanol extracts. They were absent in the petroleum ether extract. This difference may be explained by the solubility of proteins in various solvents.

Tannins were detected in both the water and methanol extracts. They were not found in the petroleum ether extract. Tannins are usually more soluble in polar solvents. Therefore, their detection in the water and methanol extracts is expected.

Alkaloids were detected in the water and methanol extracts. The petroleum ether extract showed a negative result. This indicates that alkaloid-related compounds were more readily detected in the polar extracts.

Flavonoids were detected in all three extracts. Their presence in all the extracts suggests that flavonoid-related compounds were present in the leaves and could be extracted to some extent by each solvent.

All three extracts also contained phytosterols. This shows the presence of sterol-like compounds in leaf material. The detection of these compounds in the various extracts may be due to the presence of compounds with different solubility characteristics.

Saponins were found in methanol and petroleum ether extracts. They were absent in the aqueous extract. Terpenoids were detected in the methanol and petroleum ether extracts, but the water extract was negative.

The phytochemical screening of leaves of *M. concanensis* shows the presence of different phytoconstituents. It contains carbohydrates, proteins, tannins, alkaloids, flavonoids, phytosterols, saponins, and terpenoids. The present study indicates that the biological activities may be due to these compounds.

The differences between extracts are also relevant. Not every solvent detected all compounds. This demonstrates that the choice of extraction solvent can influence the phytochemical composition of the final extract. The results also indicate that the use of multiple solvents can give a broader insight into the chemical constituents present in the plant leaves.

The antioxidant potential of the three extracts was studied in the range of 20-100 µg/ml. The three extracts exhibited antioxidant activity at the concentrations tested. The activity was found to be concentration-dependent. The water extract showed an activity of 8.32 ± 2.47 at 20 µg/ml. The activity gradually increased with concentration. It reached 24.39 ± 2.88 at 100 µg/ml. This shows that the water extract had a concentration-dependent response.

The methanol extract showed an activity of 9.23 ± 1.71 at 20 µg/ml. The activity increased to 12.28 ± 3.32 at 40 µg/ml and 17.92 ± 2.25 at 60 µg/ml. It further increased to 22.52 ± 3.33 at 80 µg/ml. The highest value of 25.73 ± 5.66 was observed at 100 µg/ml.

The petroleum ether extract showed comparatively lower activity. Its activity was 6.49 ± 0.38 at 20 µg/ml. It increased to 9.88 ± 0.87 at 40 µg/ml and 12.37 ± 0.78 at 60 µg/ml. At 80 µg/ml, the activity reached 16.63 ± 1.29. The highest value of 19.46 ± 0.66 was observed at 100 µg/ml.

Among the three extracts, the methanolic extract showed the highest antioxidant activity at most of the tested concentrations. The water extracts also showed good activity. The petroleum ether extract showed comparatively less activity at all the concentrations tested.

The response of the methanolic extract was better, which may be due to the presence of several phytochemicals that were detected during preliminary screening. The antioxidant potential of the extract could be due to some other phenolic-related constituents, alkaloids, tannins and compounds like flavonoids. Methanol may have extracted more of those compounds or more of a broader range of those compounds.

All three extracts showed a definite increase in activity with increasing concentrations. Thus, a higher concentration of the extract provided more compounds, which could be responsible for the

observed antioxidant response.

The results also suggest that the solvent used for extraction can influence the antioxidant activity. Methanol gave a higher yield of extract and better antioxidant activity. This would indicate that the compounds responsible for the observed response may be more easily extracted by methanol.

To summarise, the results suggest that *Moringa concanensis* leaves have antioxidant activity. The methanolic extract gave the most promising response among the tested extracts. Additional studies are necessary to determine the particular compounds responsible for this activity and to determine their individual contribution to the antioxidant potential of the plant.

Moringa concanensis has been studied extensively for its medicinal and phytochemical properties. However, this species has not been studied as well as Moringa oleifera.

Pharmacognostic and phytochemical characters of *M. concanensis*. Their work provided information useful for the identification and authentication of the plant and supported the traditional medicinal importance of the plant.

Pharmacognostic, phytochemical and antimicrobial studies of *M. concanensis* leaves. The study recorded the presence of different phytochemical constituents and explored the antimicrobial potential of the leaf material.

The phytochemical composition (qualitative and quantitative) of *M. concanensis* leaves, flowers and seeds was determined using methanol extracts. Alkaloids, flavonoids, terpenoids, carbohydrates, proteins and amino acids were reported to be present. Leaves and flowers also contained phenols and saponins. The study revealed that the plant has several important phytochemical components.

Investigated the phytochemical composition, antimicrobial activity and cytotoxic effects of *M. concanensis* leaves. Their results also support the biological relevance of the leaves and their potential as a source of bioactive compounds [14].

Conflict of interest statement

The authors declare no conflicts of interest.

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