

A Comprehensive Analysis of the Phytochemical Composition and Therapeutic Potential of the Aerial Parts of *Mesosphaerum Suaveolens*

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ABSTRACT

Mesosphaerum suaveolens, a flowering plant from the Lamiaceae family, is traditionally utilized for its medicinal properties across various cultures. This study investigates the phytochemical composition and potential therapeutic benefits of *M. suaveolens* extracts. The phytochemical analysis, conducted using solvents such as ethanol, chloroform, petroleum ether, benzene, and water, identified significant quantities of alkaloids, cardiac glycosides, flavonoids, tannins, saponins, and phenolic compounds. Quantitative assessments revealed that cardiac glycosides were the most abundant, followed by alkaloids and phenolic compounds. The diverse bioactive compounds present in *M. suaveolens* exhibit significant pharmacological potential, including antioxidant, antimicrobial, anti-inflammatory, and cardiogenic properties. These findings underscore the plant's relevance in traditional medicine and its potential as a source of novel therapeutic agents. Further research is essential to optimize extraction methods, understand the mechanisms of action, and evaluate the bioavailability and toxicity of these compounds to facilitate their application in modern medicine.

KEYWORDS: *Mesosphaerum suaveolens*, phytochemical analysis, bioactive compounds, Traditional medicine, cardiac glycosides, antioxidant, Antimicrobial, Anti-inflammatory, Pharmacological potential, Therapeutic agents

INTRODUCTION

Plants play a crucial role in providing medicinal resources due to their rich array of bioactive compounds, which offer antioxidant, antibacterial, and antifungal properties [1, 2, 3].



Fig.1: Aerial part of Mesosphaerum suaveolens

A substantial portion of pharmaceuticals, around 25%, and a significant portion of global healthcare solutions are derived from plant sources [4, 5]. Natural antioxidants, in particular, are pivotal in mitigating oxidative stress induced by free radicals, thereby regulating physiological processes effectively and safely [6, 7, 8]. While conventional medicine predominantly employs synthetic or semi-synthetic antibiotics, the emergence of antibiotic-resistant pathogens and the associated financial burden have spurred interest in exploring plant extracts and derivatives as alternative therapeutic options [9, 10, 11]. Numerous studies have showcased the therapeutic potential of various botanicals in addressing chronic conditions such as cancer, diabetes, inflammation, stroke, and aging [12, 13, 14]. This importance of plants as reservoirs for novel drugs and therapeutic agents [15]. In contrast to plant cells, human cells often lack sufficient levels of antioxidants, necessitating the supplementation with synthetic alternatives [16, 17]. However, concerns regarding the toxicity of synthetic antioxidants have prompted the exploration of safer natural alternatives present in plants, including vitamins, phenolics, flavonoids, tannins, and carotenes [18, 19, 20]. Increased dietary intake of natural antioxidants has demonstrated beneficial effects on chronic diseases like heart disease and cancer, driving the demand for natural antioxidants in pharmaceuticals, nutraceuticals, and food additives [21, 22, 23]. Agriculture faces similar challenges, with fungal and bacterial infections posing significant threats to crop yields [24]. Throughout history, humans have relied on plants not only for sustenance but also for medicinal purposes and construction materials, with approximately 80% of all natural products having plant origins [25, 26, 27]. This highlights the enduring importance of plants in various facets of human life, including healthcare and agriculture.

Mesosphaerum suaveolens, which is also known as "Alfavaca-do-campo" or "Sambacaitá," or Vilati Tulsi is a flowering plant species belonging to the Lamiaceae family [28, 29]. Primarily native to various regions of Central and South America, including Brazil, Argentina, Paraguay, Uruguay, and rarely found in India, particularly in the Chotnagpur plateau, this plant is recognized for its fragrant foliage and petite, tubular

flowers spanning hues from white to lavender [30, 31]. Typically growing as a perennial herb reaching heights of up to one meter, it belongs in diverse habitats such as open fields, grasslands, and disturbed areas, displaying adaptability to both arid and moist environments. In traditional medicine, it has been extensively utilized for its therapeutic properties [32]. Different parts of the plant, including its leaves, stems, and roots, have been employed in herbal preparations to address various health concerns, ranging from digestive issues and respiratory ailments to skin conditions. Scientific inquiries into *Mesosphaerum suaveolens* have revealed its potential pharmacological activities, which include antimicrobial, anti-inflammatory, antioxidant, and analgesic properties [33, 34, 35]. These findings underscore the plant's significance in modern medicine and highlight its potential as a source of novel therapeutic agents [36, 37]. Despite its promising attributes, further research is warranted to comprehensively understand the chemical composition, pharmacological effects, and potential adverse reactions associated with *Mesosphaerum suaveolens*. Additionally, conservation efforts are imperative to ensure the sustainable utilization of this species, given its importance in traditional medicine and its potential value in pharmaceutical development.

Furthermore, *Mesosphaerum suaveolens* has been traditionally employed for various traditional purposes, including digestive aid, respiratory support, anti-inflammatory applications, skin care, and as a general tonic. In certain indigenous cultures, it holds symbolic significance beyond its medicinal utility, often incorporated into cultural and spiritual practices as a symbol of purification, protection, or reverence for nature [38, 39, 40].

Taxonomically, *Mesosphaerum suaveolens* is classified within the Plantae kingdom, Angiosperms clade, Eudicots clade, Asterids clade, Lamiales order, Lamiaceae family, and *Mesosphaerum* genus. Within the *Mesosphaerum* genus, it is specifically identified as the *suaveolens* species [41, 42].

EXPERIMENTAL SECTION

The investigations were carried out in the Medicinal Chemistry Laboratory located within the Faculty of Medical Science and Research at Sai Nath University, Ranchi. The experiments were conducted daily from 4:00 P.M. to 5:00 P.M. over a period of 1.5 months.

Plants materials and reagents

Freshly shoots of *M. suaveolens* were collected from the premises of Sai Nath University, located in Ranchi, Jharkhand, India (geographical coordinates 23° 29' 18.47" N, 85° 24' 28.73" E). Botanical identification and authentication procedures were conducted at Shibpur Botanical Garden in West Bengal, India. To eliminate any extraneous dirt particles, shoots underwent a gentle washing process with tap water, followed by air-drying in shaded areas. For the extraction process, various chemicals, including Ethanol, Petroleum ether, chloroform, and benzene, were utilized. Additionally, Sodium Hydroxide, concentrated Hydrochloric acid, Magnesium powder, Ferric chloride solution, Distilled water, Folin-ciocalteu reagent, Sodium bicarbonate, Potassium Iodide, Mercuric chloride, and Baljet's reagent were supplied by Sai Nath University, all of which met analytical grade standards. Working standards and samples were prepared by diluting the stock solution (1 mg/ml) in ethanol and double-distilled water, adjusting concentrations as required for the experiment. Moreover, the solvents used in the investigation were also of analytical grade.

Extraction procedure [43]

Freshly collected shoots of *M. suaveolens* underwent thorough cleansing by distilled water to remove any contaminants before being dried in a shaded area until completely dehydrated. Once dried, the shoots were

finely ground into powder using a blender. Subsequently, 150 grams of this powder underwent an extraction process using different solvents in a Soxhlet extractor for 72 hours. The solvents used included petroleum ether, benzene, chloroform, ethanol, and distilled water. Following extraction, the obtained extracts were concentrated and carefully stored in airtight containers for future use.

PHYTOCHEMICAL ANALYSIS

Qualitative Analysis

The phytochemical screening of *M. suaveolens* shoots was carried out using a standardized method to identify the presence of various compounds. The results of the screening revealed the presence of Flavonoids, Tannins, Saponins, Cardiac Glycosides, Phenolic Compounds and Alkaloids.

Determination of total Flavonoids [44]

The method involves the formation of a complex between flavonoids and aluminum, with its highest absorption occurring at 415nm. For the analysis, 100 μ l of plant extracts dissolved in methanol (10 mg/ml) were combined with 100 μ l of 20% aluminum trichloride in methanol and a drop of acetic acid. This mixture was then diluted with methanol to a final volume of 5ml. After 40 minutes, the absorbance at 415nm was recorded. Blank samples were prepared using 100 μ l of plant extracts, a drop of acetic acid, and methanol, then diluted to 5ml. Additionally, the absorbance of a standard rutin solution (0.5 mg/ml) in methanol was measured using the same procedure. All measurements were conducted in triplicate.

Determination of total Tannins [45]

The experiment conducted by weighing a precise 500 mg portion of the sample and placing it into a 50 ml plastic bottle. Then, 50 ml of distilled water was added to the bottle, and the contents were vigorously shaken for one hour using a mechanical shaker. Afterward, the mixture was filtered into a 50 ml volumetric flask, and the flask was filled up to the mark to ensure accurate volume measurement. Following filtration, 5 ml of the filtered solution was transferred into a test tube using a pipette. In the test tube, the solution was mixed with 2 ml of 0.1 M FeCl₃ solution in a mixture containing 0.1 N HCl and 0.008 M potassium ferrocyanide. The absorbance of the resulting solution was then measured at a wavelength of 120 nm for a duration of 10 minutes to evaluate its properties.

Determination of total Saponins [46]

Samples of 20 grams each were ground and placed into a conical flask. 100 cubic centimeters of a 20% aqueous ethanol solution were added to each flask. The flasks were then heated on a hot water bath, with continuous stirring at around 55°C, for 4 hours. After heating, the mixture underwent filtration, and the residue underwent another extraction using 200 milliliters of 20% ethanol. The extracts from both processes were combined and concentrated to 40 milliliters using a water bath at approximately 90°C. The concentrated solution was transferred to a 250 milliliter separatory funnel. Then, 20 milliliters of diethyl ether were added and vigorously shaken. The aqueous layer was separated and retained, while the ether layer was discarded. This purification step was repeated, followed by the addition of 60 milliliters of n-butanol. The combined n-butanol extracts were washed twice with 10 milliliters of a 5% aqueous sodium chloride solution. Then, the remaining solution was heated in a water bath. After evaporation, the samples were dried in an oven until a constant weight was achieved. The saponin content was calculated based on these dried samples.

Determination of total Cardiac glycosides [47]

To detect cardiac glycosides, a 10% extract from each batch and the overall seed extract were combined with 10 mL of freshly prepared Baljet's reagent. This reagent consists of 95 mL of 1% picric acid and 5

mL of 10% NaOH. After an hour, the mixture was diluted with 20 mL of distilled water, and the absorbance was measured at 495 nm using a Shimadzu UV/VIS spectrophotometer model 160A (Kyoto, Japan). For the standard curve, solutions with various concentrations (ranging from 12.5 to 100 mg/L) were prepared. The total glycosides obtained from triple replicates were quantified and expressed as milligrams of *M. suaveolens* per gram of dried extracts.

Determination of total Phenolic compounds [48]

A precisely measured sample extract weighing 100 milligrams was dissolved in 100 milliliters of triple distilled water (TDW). Following this, 1 milliliter of this solution was transferred to a test tube. Then, 0.5 milliliters of 2N Folin-Ciocalteu reagent and 1.5 milliliters of 20% Na₂CO₃ solution were added. The volume was adjusted to 8 milliliters with TDW, followed by vigorous shaking. The mixture was left to stand for 2 hours, after which the absorbance was measured at 765 nanometers. These absorbance readings were utilized to calculate the total phenolic content by referring to a standard calibration curve established using various diluted concentrations of gallic acid.

Determination of total Alkaloids [49]

A 5-gram portion of the sample was accurately measured and placed into a 250-milliliter beaker. Then, 200 milliliters of a 10% acetic acid solution in ethanol were added to the beaker, which was covered and left undisturbed for 4 hours. Following this, the mixture underwent filtration, and the resulting extract was concentrated on a water bath until it reached one-quarter of its original volume. To ensure complete precipitation, concentrated ammonium hydroxide was slowly added drop by drop to the concentrated extract. The entire solution was then allowed to settle to facilitate the formation of precipitate. The formed precipitate was carefully collected and subjected to washing with dilute ammonium hydroxide. Once washed, the precipitate was filtered, leaving behind a residue containing the alkaloid. Finally, the alkaloid residue was dried and weighed for further analysis.

RESULT

Qualitative phytochemical analysis

In a qualitative analysis of *M. suaveolens* extracts, researchers utilized five different solvents (aqueous, ethanol, chloroform, petroleum ether, and benzene) to evaluate various phytochemical properties, as detailed in **Table 1**. The results indicated that each solvent yielded positive outcomes in at least one of the six phytochemical tests. Notably, the ethanol, petroleum ether, aqueous, and chloroform extracts demonstrated positive results across all six tests, suggesting a wide array of chemical compounds present. Conversely, the benzene and aqueous extract showed positive results in five tests, indicating a slightly lower diversity of phytochemicals compared to ethanol. Overall, the ethanol extract exhibited the highest number of positive outcomes, followed by benzene, chloroform, aqueous, and petroleum ether extracts. The investigation primarily centered on screening for phytochemical compounds within the five solvent extracts. These compounds included alkaloids, phenolic compounds, flavonoids, saponins, tannins, and cardiac glycosides, all of which are recognized as significant secondary metabolites known for their medicinal properties within plants. Furthermore, researchers conducted additional analytical tests to quantify the presence of these phytochemical compounds in the extracts.

Table.1 Qualitative phytochemical analysis of Mesophaerum suaveolens

Compounds	Aqueous	Ethanol	Pet. Ether	Chloroform	Benzene	Corresponding Test	Results
Alkaloids	Present	Present	Present	Present	Present	Mayer's Test [50]	Cream precipitate arises
Cardiac Glycosides	<u>Absent</u>	Present	Present	Present	Present	Kedde's reagent Test [51]	Reddish brown colour arises
Flavonoid	Present	Present	Present	Present	Present	Shinoda Test [52]	Yellow precipitate arises
Tannins	Present	Present	Present	Present	Present	Ferric chloride Test [53]	Dark greenish black colour arises
Phenolic compounds	Present	Present	Present	Present	Present	Legal's reagent Test [54]	Greenish colour arises
Saponins	Present	Present	Present	Present	<u>Absent</u>	Foam Test [55]	Foam arises


Fig.1: Aerial part of Mesophaerum suaveolens

Fig. 2 Standard extract of M.Suaveolens



Fig. 3 Test for Alkaloids



Fig. 4 Test for Cardiac glycosides



Fig. 5 Test for Flavonoids

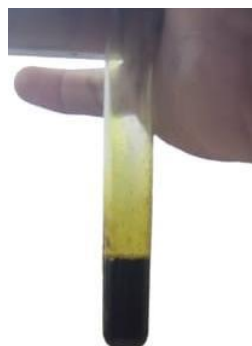


Fig. 6 Test for Tannins



Fig. 7 Test for Saponins



Fig. 8 Test for Phenolic Compounds

Quantitative phytochemical analysis

The investigation involved quantifying the phytochemicals present in the plant extract. The findings indicated varying levels of different phytochemicals across different extracts of *M. suaveolens*. Specifically, cardiac glycosides emerged as the primary constituents in the analyzed shoot parts. They were followed by alkaloids, phenolic compounds, tannins, and flavonoids, as outlined in **Table 2**. In contrast, the presence of saponins in this extract was noted to be minimal.

Table 2 Quantitative phytochemical analysis of *Mesosphaerum suaveolens*

Phytochemicals	Aqueous extract (mg.)	Ethanolic extract (mg.)	Pet. ether extract (mg.)	Chloroform extract (mg.)	Benzene extract (mg.)
Alkaloids	25.10 ± 0.35	27.13 ± 0.45	26.33 ± 0.54	24.31 ± 0.21	22.01 ± 0.45
Cardiac glycosides	<u>Absent</u>	40.40 ± 0.25	36.34 ± 0.10	38.60 ± 1	35.51 ± 0.37

Flavonoids	20.21 ± 0.25	22 ± 0.55	21.42 ± 0.43	23.33 ± 0.65	19.77 ± 0.43
Tannins	18.55 ± 0.7	20.01 ± 0.54	17.21 ± 0.23	19.65 ± 0.42	18.59 ± 0.07
Phenolic compounds	23.77 ± 0.32	24.05 ± 0.40	24.90 ± 0.15	22.44 ± .85	21.7 ± 0.61
Saponins	8.3 ± 0.10	9.09 ± 0.5	8.43 ± 0.25	7.77 ± 0.51	Absent

Upon analysis, the **aqueous extract** was determined to contain 25.10 mg of alkaloids, 20.21 mg of flavonoids, 18.55 mg of tannins, 23.77 mg of phenolic compounds, and 8.3 mg of saponins. Notably, cardiac glycosides were absent in this extract. Similarly, the **ethanol extract** exhibited 27.13 mg of alkaloids, 40.40 mg of cardiac glycosides, 22 mg of flavonoids, 20.01 mg of tannins, 24.05 mg of phenolic compounds, and 8.3 mg of saponins. The **petroleum ether extract** displayed 26.33 mg of alkaloids, 36.34 mg of cardiac glycosides, 21.42 mg of flavonoids, 17.21 mg of tannins, 24.90 mg of phenolic compounds, and 8.43 mg of saponins. Moreover, the **chloroform extract** contained 24.31 mg of alkaloids, 38.60 mg of cardiac glycosides, 23.33 mg of flavonoids, 19.65 mg of tannins, 22.44 mg of phenolic compounds, and 7.77 mg of saponins. Lastly, the **benzene extract** was found to comprise 22.01 mg of alkaloids, 35.51 mg of cardiac glycosides, 19.77 mg of flavonoids, 18.59 mg of tannins, and 21.7 mg of phenolic compounds. Saponins were not detected in this extract.

All the above data are combined and shown in the following **Charts**.

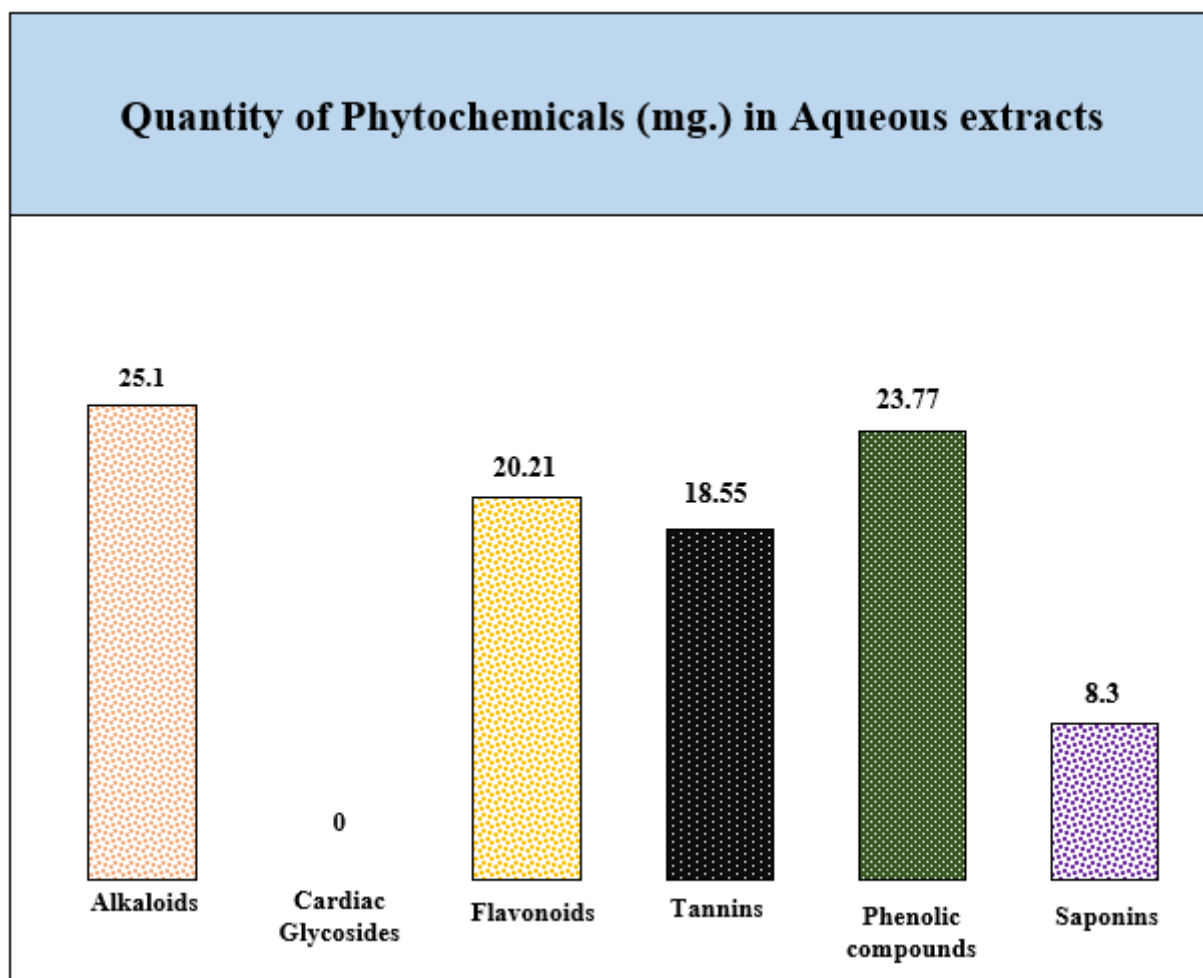


Chart No. 1 Quantitative phytochemical analysis of *Mesophaerum suaveolens* in aqueous extract

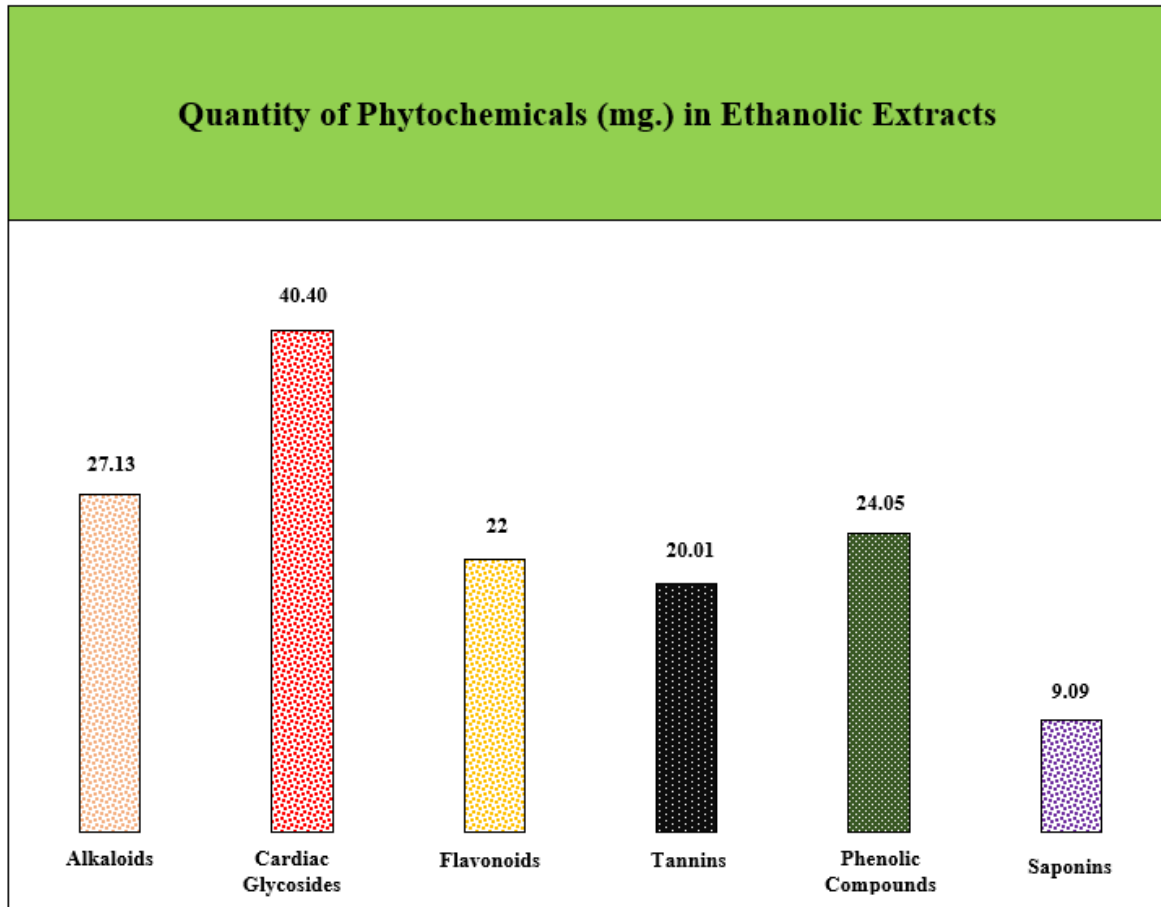


Chart No. 2 Quantitative phytochemical analysis of *Mesophaerum suaveolens* in ethanolic extract

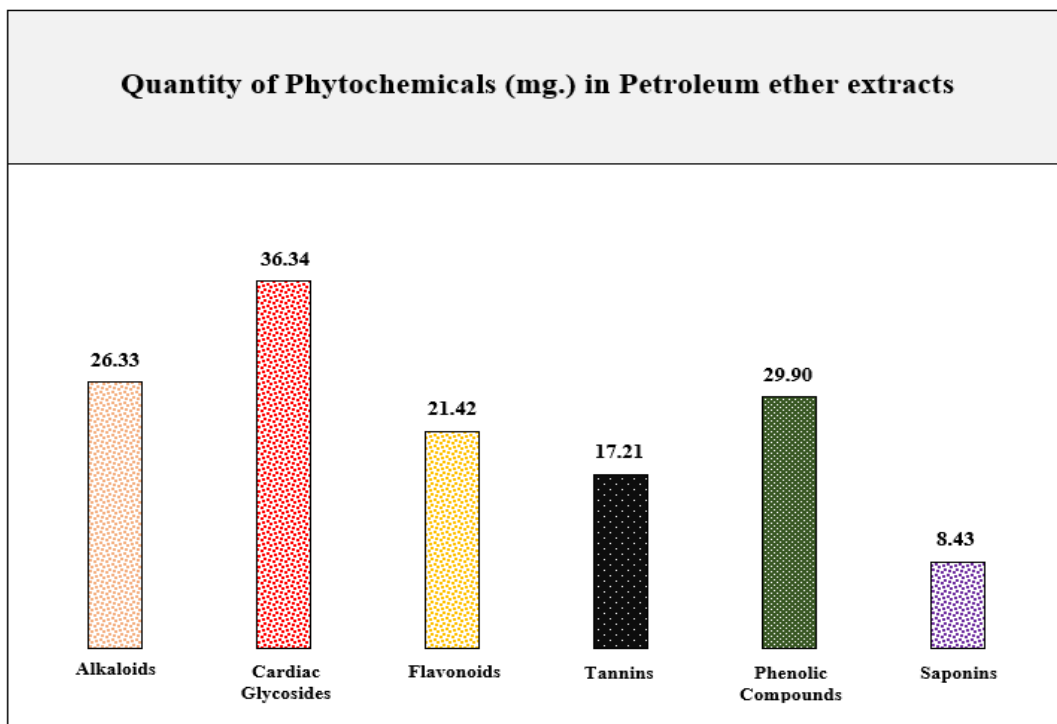


Chart No. 3 Quantitative phytochemical analysis of *Mesophaerum suaveolens* in Petroleum ether extract

DISCUSSION

The bioactive compounds identified in *Mesosphaerum suaveolens* exhibit significant medicinal potential due to the presence of active phytochemicals, including alkaloids, cardiac glycosides, flavonoids, tannins, saponins, and phenolic compounds.

The bioactive phytochemicals found in *Mesosphaerum suaveolens* demonstrate significant pharmacological potential. Alkaloids such as morphine are widely used for their analgesic and anti-inflammatory properties, while quinine and its derivatives are crucial for their antimalarial efficacy and antibacterial capabilities [56, 57]. Additionally, alkaloids like vincristine and vinblastine are employed in cancer chemotherapy [58]. Cardiac glycosides, including digoxin, are notable for their cardiotonic effects, enhancing heart contractions to treat heart failure and arrhythmias, and also exhibit diuretic properties beneficial for managing oedema [59, 60]. Flavonoids provide antioxidant protection by scavenging free radicals, possess anti-inflammatory effects by inhibiting inflammatory enzymes, and offer cardioprotective benefits by improving vascular function [61, 62, 63]. Tannins serve as astringents, useful in treating diarrhoea and skin conditions, and have antimicrobial and antioxidant activities [64, 65]. Saponins are known for their immune-boosting effects, cholesterol-lowering abilities by binding to cholesterol and bile acids, and anticancer properties through the induction of apoptosis in cancer cells [66, 67, 68]. Phenolic compounds contribute to antioxidant defence, reducing oxidative damage and the risk of chronic diseases, exhibit anti-inflammatory effects, and possess broad-spectrum antimicrobial activities [69, 70, 71]. Collectively, these phytochemicals underpin the medicinal potential of *Mesosphaerum suaveolens*, providing a diverse range of therapeutic benefits.

Future research on *Mesosphaerum suaveolens* should focus on optimizing extraction methods to maximize the yield of specific bioactive compounds, depending on their intended therapeutic applications. Given the variability in phytochemical content across different solvents, it is crucial to identify the most effective solvent for targeting particular compounds such as cardiac glycosides, alkaloids, or flavonoids. Additionally, further studies should investigate the synergistic effects of these compounds in complex biological systems to better understand their pharmacological potential. Exploring the mechanisms of action, bioavailability, and toxicity of these extracts will provide a more comprehensive understanding of their medicinal value. Advanced techniques like metabolomics and molecular docking studies could also be employed to identify novel compounds and predict their biological activities. Ultimately, the development of standardized extraction protocols and detailed pharmacokinetic studies will be essential for the potential commercialization of *Mesosphaerum suaveolens* extracts as therapeutic agents.

CONCLUSION

The study on *Mesosphaerum suaveolens* reveals its significant pharmacological potential due to its rich array of bioactive compounds, including alkaloids, cardiac glycosides, flavonoids, tannins, saponins, and phenolic compounds. Qualitative and quantitative phytochemical analyses indicated that the ethanol extract exhibited the highest diversity and concentration of these compounds, highlighting its promise as a potent source of therapeutic agents. The identified phytochemicals demonstrate a range of medicinal properties: alkaloids such as morphine and vincristine offer analgesic, anti-inflammatory, antimalarial, and anticancer benefits; cardiac glycosides like digoxin provide cardiotonic and diuretic effects; flavonoids contribute antioxidant, anti-inflammatory, and cardiovascular benefits; tannins act as astringents with antimicrobial and antioxidant activities; saponins are noted for their immune-boosting, cholesterol-lowering, and anticancer properties; and phenolic compounds enhance antioxidant defense,

reducing oxidative damage, and exhibit anti-inflammatory and antimicrobial activities. These findings validate the traditional use of *Mesosphaerum suaveolens* in herbal medicine and underscore its potential for developing new therapeutic agents. Further research is necessary to optimize extraction methods, explore synergistic effects, and elucidate the pharmacokinetics and mechanisms of these bioactive compounds. Conservation efforts are essential to ensure sustainable utilization of this species, given its therapeutic promise and traditional medicinal value.

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