

Decoding the therapeutic efficacy of under-researched plant from Asian sub-continent *Thysanolaena maxima* (roots) by pharmacognostical, phytochemical and pharmacological investigation

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Abstract

Objective: To evaluate *Thysanolaena maxima* with physico-chemical parameters and phytochemical analysis and to investigate the *in vitro* bioactivities of different plant extracts obtained from Sikkim, India. **Methods:** The ash value, extractive value, loss on drying (LOD), fluorescence analysis, powder microscopy, and phytochemical screening were conducted using the specific standard procedures. Antioxidant activity was evaluated using the 2, 2-diphenyl-1-picryl-hydrazyl-hydrate (DPPH) radical scavenging assay and the reducing power assay. Accordingly, antibacterial activity was investigated using the disk diffusion method. **Results:** Physico-chemical properties play a major role in determining drugs quality and purity. The ash value (total ash, acid insoluble and water soluble), extractive value, and LOD were found to be (6.4, 2.5, and 2.15%), (petroleum ether (1.26%), dichloromethane (1.68%), dichloromethane: methanol (1:1) (2.21%), chloroform water (3.84%), and 9.1%, respectively. In Fluorescence analysis, distinct colors were observed in the far and near ultra-violet ranges. The presence of fibers, stone cells, and graphite belonging to types A and C that is, random flake graphite and Kish graphite with hyper-eutectic composition, was observed in Powder microscopy. Phytochemical screening revealed the presence of Flavonoids, Alkaloids, steroids, phenol, and saponins. The dichloromethane: methanol (1:1) extract showed the highest reducing power as well as DPPH scavenging activity (74.966 ± 1.705) compared to other extracts. Almost all extracts showed mild anti-bacterial activity, with zones of inhibition ranging from 0.75 to 1.62, respectively. **Conclusion:** Systematic investigation of *T. maxima* preliminary showed a potent source of bioactive compounds that can be utilized as a lead for new drug discovery.

Key words: 2,2-diphenyl-1-picryl-hydrazyl-hydrate, bioactive compounds, drug discovery, reducing power, *Thysanolaena maxima*

INTRODUCTION

Natural products offer a diverse array of multi-dimensional chemical compositions, and their utilization as physiological function modifiers has become extremely prevalent.^[1] As a result, herbal remedies are extremely critical to the health of their consumers around the globe. This medicinal trait is related to the existence of specific chemical substances capable of causing physiological effects in the body.^[2]

Ethnobotany is a trans-disciplinary science that studies how plants and people interact with each

other^[3] while also anticipating the future. It preserves the history and current state of mankind. A traditional healthcare system is a conventional healthcare care system that is culturally organized in every ethnic group. In rural areas,

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health care appears to be the first and most important line of defense.^[4,5] Native tribes and tribal groups have established their own customs, cuisines, and healing practices throughout the world. Among these cultures, a variety of wild and domesticated plants play a crucial and vital role.^[6] Sikkim is a small hilly state in the Eastern Himalayas, covering a total area of 7096 square kilometers. Sikkim, which makes up only 0.2% of the country's land area, is home to various species like Rhododendrons (36), oaks (11), Bamboos (26), Ferns (326) and their Allies (326), Orchids (410), tree ferns (8), primulas (30), and approximately 242 medicinal plants discovered here, and boasts incredible biodiversity in the Eastern Himalayas. Ethnic groups like the Lepcha, Bhutia, Limboo, Sherpa, and Nepali have long practiced traditional medicine and relied heavily on herbal remedies.^[7-9]

Thysanolaena maxima, a perennial herb belonging to the Poaceae family, is found mainly in hilly areas of Nepal and India's northern and eastern states, including Sikkim, Bangladesh, and Bhutan. Various plant parts have been traditionally used by tribal populations across the world and within the Asian continent. *T. maxima* root preparation is often used as a remedy for boils, and the extract of the shrub is administered twice per day to eradicate intestinal worms.^[10,11] For bronchial symptoms, the Sikkimese ethnic group utilizes a single dosage of young root decoction twice a day. A poultice of young flowers is used for rheumatic pain and skin swelling. Lodhas, a traditional practitioner, prescribes a paste of flowers along with country liquor and honey as a contractive to women's.^[12] People in Thailand's western forest boil the entire *T. maxima* plant with water and drink it as a tonic. Crushed flowers and water are used as antiemetics and to cure gastrointestinal issues by the Kanda tribal people of Sylhet, Bangladesh. The Dimasa people of Assam, India, consume the tender parts of new leaves and flower buds fresh to heal flatulence and promote digestion. The Chakma folks of Chittagong, Bangladesh, use leaf-based pills twice a day for tuberculosis treatment.^[2]

A literature survey revealed that few research studies have been performed on this plant to evaluate its medicinal values and the active constituents responsible for its pharmacological activities. However, different solvent systems and conditions have distinct impacts on extracting bioactive compounds, resulting in differences in their bioactivities. Therefore, considering the traditional uses of the plant and facilities available for conducting the study, this research work was performed on different solvent extracts of the plant's roots available in the northeastern state of India, Sikkim.

MATERIALS AND METHODS

Chemicals and Solvents

All the chemicals and solvents used in the study were of analytical grade and were procured from Thermo Fisher

Scientific India Private Limited, Mumbai, and S. D. Fine Chemical Limited, Mumbai.

Authentication of Plant

The roots of *T. maxima* were collected from Linkey, Pakyong, Sikkim Himalayan region, on November 24th 2021, and authenticated by the Botanical Survey of India, Sikkim Himalayan Regional Centre, Gangtok, India. (Specimen number: SP-21-01).

Macroscopic Analysis

The macroscopical characteristics of the raw drug were studied. These include analysis of the dried root such as size, shape, color, etc. Organoleptic analysis of raw drugs was done through sensory-based characteristics like smell, taste, etc.^[13,14] were recorded.

Physico-chemical Analysis

The ash value, extractive value, and loss on drying (LOD) of the drug were assessed using the method provided in the British Pharmacopoeia^[15] for physicochemical investigations. The method developed by Kokoshi *et al.* and Das *et al.*^[16,17] was used to analyze the fluorescence of powdered drugs. A 1mg powder drug was placed on a microslide and subjected to UV and daylight to study the drug's luminous properties. Following that, the same amount of medication was placed on a second micro slide and treated individually with 50% H₂SO₄, 50% HNO₃, methanol, 1(N) HCl, cold water, hot water, picric acid, ammonia solution, chloroform, GAA, 5 percent iodine, Ferric chloride, and Pet ether, each 1 ml. To observe the fluorescence properties of powder, the slides were subjected to UV (short and long wavelengths) and daylight. For powder analysis, after being shade-dried, the roots were finely crushed and observed under the microscope. A small amount of powder was deposited on each slide independently, and each slide was then mounted with 2-3 ml of two distinct reagents, phloroglucinol, and HCl: water (1:1). Each slide was covered with a cover slip and inspected under a microscope^[18] and a picture was taken with a digital camera.

Extraction of Plant Materials

250g of powdered plant materials were immersed in 1liter of petroleum ether, dichloromethane, chloroform water, and dichloromethane: methanol (1:1) for 5 days with intermittent shaking and swirling in an airtight flat-bottom container. The majority of the plant materials' extractable components were dissolved in various solvents, which were collected and then evaporated using a rotary evaporator (HAHNAPOR, HS 3000) at a low temperature (40–50°C) and reduced pressure.

For future usage, the dried crude extracts were collected and kept at 4°C.

Phytochemical Screening

The existence of chemical components was qualitatively examined in the freshly generated extracts. Alkaloids were screened using Wagner's, Dragendorff's, Hager's, and Mayer's reagents; carbohydrates were screened using Molisch's, Barford's, and Fehling's reagents; flavonoids were screened using ferric chloride, alkaline reagent, and the Shinoda test; saponins were screened using the foam and forth test; Protein with the Ninhydrine and Biuret test; glycoside with the keller-killani and Legal test; cellulose with the iodine water test; tannins with ferric chloride and vanillin hydrochloride reagent; steroids with the Salkowski test; and phenols with ferric chloride and litmus paper using standard procedure.^[19,20] Using normal procedures, these were identified by distinctive color changes.

In-vitro Evaluation of Antioxidant Activity

In-vitro antioxidant studies of the plant extracts were carried out following two methods: the reducing power method (RP), which suggests an increase in absorbance of the reaction mixture leads to increase in antioxidant activity, and the DPPH radical method, which suggests a decrease in absorbance of DPPH in the presence of extracts or test samples infers antioxidant activity.

Reducing Power Method (RP)

The principle behind this approach is that the absorbance of the reaction mixtures increases. A rise in absorbance suggests that antioxidant activity has increased. In this technique, an antioxidant molecule forms a vibrantly colored complex at 700 nm with trichloroacetic acid, ferric chloride, and potassium ferricyanide. The reaction mixture's absorbance rises, indicating the samples' reducing power.^[21] In this method, 1 mL of sample diluted with distilled water is mixed with 2.5 mL of 0.2 Molar phosphate buffer (pH 6.6) and a 1 % w/v solution of potassium ferricyanide (2.5 ml). This mixture is then incubated for 20 min at 50°C. The mixture recovered after incubation is added to 2.5 ml of 10% C₂HCl₃O₂. After centrifuging the reaction mixture for 15 min at 3000 rpm, the upper supernatant layer of solution (2.5 ml) is withdrawn and mixed with distilled water (2.5 ml) and 0.1 percentage w/v ferric chloride (0.5 ml). The resulting solution is exposed to UV at 700 nm in reference to a blank.^[22]

DPPH Radical Method

To access the free-radical-scavenging activity of crude extracts, DPPH radical agents are often preferred and utilized. As described by Kondragunta *et al.*, the radical scavenging

abilities of four distinct plant extracts were investigated with minor modifications. In this study, the decrease in DPPH absorbance in the presence of the extract was measured. Each of the four crude extracts was combined with 10⁻⁴ mol/L DPPH in methanol and incubated at 37°C for 15 min in darkness. Ascorbic acid was used as a control for this study. For the baseline correction, methanol was utilized. Measurement of the absorbance was done at a wavelength of 517 nm. Antioxidant activity is evidenced by a decrease in absorbance. To estimate radical scavenging activity as a percentage inhibition of DPPH, the following equation is used:

$$\text{DPPH radicals scavenged (\%)} = [(Ab_{\text{control}} - Ab_{\text{sample}}) / Ab_{\text{control}}] \times 100, \text{ where } Ab_{\text{control}} \text{ is the absorbance of the control reaction and } Ab_{\text{sample}} \text{ is the absorbance of the tested extract samples.}^{[23,24]}$$

Antibacterial Assay

The disc diffusion method^[25,26] was used to conduct the antibacterial experiment against one Gram-positive (GM⁺) (*Bacillus subtilis*) (US 564) and one Gram-negative (GM⁻) (*Salmonella typhi* 89) bacterial strain. In this approach, 100 µl of suspension of each microbe, comprising around 100-150 CFU/ml, was seeded into nutritional agar medium plates. Filter paper discs (6 mm in diameter) soaked with 400 µg of different extracts were gently inserted on the agar plates after being dried and sterilized. After a 24-h incubation period at 37°C, the antibacterial activity of the extracts was measured by measuring the diameter of the zone of inhibition in millimeters.

RESULTS

Morphological Description

T. maxima (ROXB.) is a perennial forest shrub with a tall reed-like, non-invasive, and extremely vigorous perennial grass with bamboo-like leaves. It's a non-timber species that grows, in big, dense clumps, producing numerous unbranched, regular-jointed upright or arching stems (up to 10 mm thick). The leaves on these stems are big and alternately placed. Culms are robust, smooth, and rounded, reaching a maximum height of 4 m. Long lance-shaped leaves with pointy tips and whole edges are quite broad (25–60 cm long and 3–7 cm wide). They are mainly composed of a basal sheath that completely covers the stem and a developing leaf blade. The inflorescence, which is 30–90 cm long, looks like a foxtail. It grows well at low to medium heights, although it grows more quickly at higher elevations. Whole plant and Roots of *Thysanolaena maxima* were shown in Figure 1.

Organoleptic Evaluation

Organoleptic analysis of the root was carried out for the following sensory-based characters: smell, taste, color, and touch. The result obtained is reported in Table 1.

Physico-chemical Analysis

The values of all determinations are summarized in Tables 2 and 3. In this evaluations, the amount of water-soluble ash was higher than acid-insoluble ash, while the amount of total ash was nearly triple the amount of water-soluble ash. The Reducing power of different extracts of powdered roots of *Thysanolaena maxima* were presented graphically in Figure 2.

Fluorescence Analysis

The results are summarized in Table 4.

Powder Microscopy

Powder microscopy was done by taking 2–3 ml of two distinct reagents, phloroglucinol and HCl (1:1), and observed them under a microscope for their distinct characteristics. Fibers, stone cells, and graphite were identified as cell components, which are shown in Figure 3.

Phytochemical Screening

A phytochemical screening was carried out, and the results are shown in Table 5. These results represent the presence of alkaloids, saponins, flavonoids, steroids, phenols, etc. in the

roots of *T. maxima*. The extractive constituents present in the different solvent extracts are tabulated in Table 5.

Tests for Antioxidant Activity

Reducing Power Method (RP)

The results obtained are tabulated below in Table 6. The Mean % of DPPH scavenging activity of different extracts of powdered roots of *Thysanolaena maxima* where presented graphically in Figure 4.

DPPH Radical Method

The DPPH radical scavenging activity of the standard and tested samples is tabulated below in Table 7. The Mean % of DPPH scavenging activity of different extracts of powdered roots of *Thysanolaena maxima* where presented graphically in Figure 5.

Antibacterial Assay

The disc diffusion method was used to conduct the antibacterial experiment against one Gram-positive (*Bacillus subtilis*) and one Gram-negative (*S. typhi*) bacterial strain. The results obtained are tabulated below in Table 8.

Table 1: Physical characteristic of *Thysanolaena maxima* root

Color	Light yellow
Odor	Odorless
Taste	Starchy
Fracture	Fibrous

Table 2: LOD and ash values of powdered roots of *Thysanolaena maxima*

S. No.	Parameters	Values obtained in % w/w
1	LOD	9.1
2	Total ash	6.4
3	Acid insoluble ash	2.5
4	Water soluble ash	2.15

LOD: Loss on drying

Table 3: Extractive values of powdered roots of *Thysanolaena maxima*

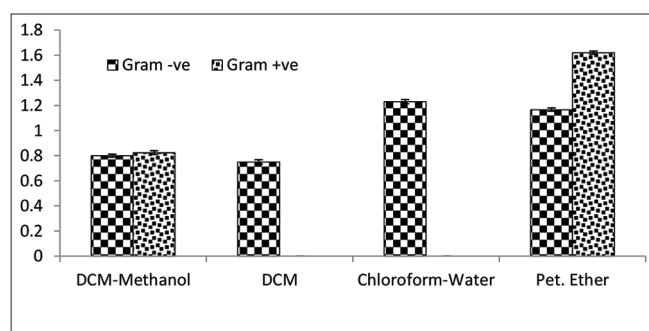
Solvent used	Physical nature	Color	% yield
Petroleum ether	Sticky	Light brown	1.26
Dichloromethane	Dried powder	Pale yellow	1.68
Dichloromethane: methanol (1:1)	Semisolid	Pale yellow	2.21
Chloroform water	Semi solid	Dark brown	3.84

DISCUSSION

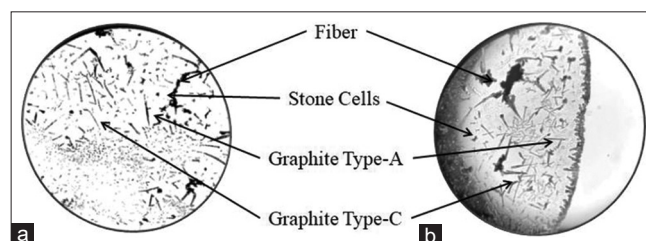
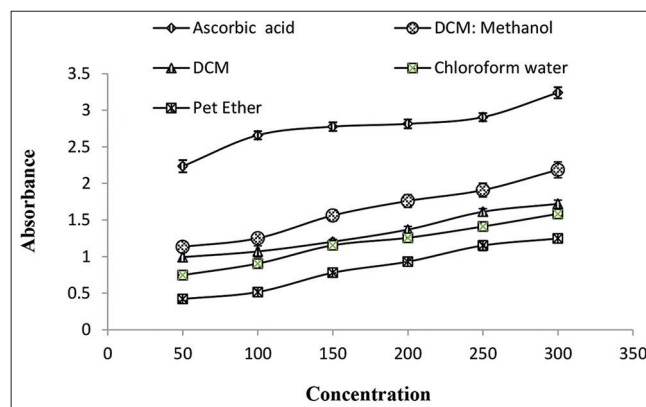
Standardization of herbal medicines is equally important, like quality checks in allopathic medicine, to ensure the drug's quality because alternative or counterfeit herbal components are frequently discovered in the marketed formulation. These examinations are intended to ensure the drug's identity, quality, purity, and safety for human consumption. Microscopic, macroscopic, physico-chemical, and fluorescent analyses are some of the factors investigated for *T. maxima*. Morphological evaluation was carried out to identify the plant. Morphological characters of the plant revealed that the plant used in the research is authentic and resembles the characters of *T. maxima* as found in various literature. The color of the crude drug was light yellow, with an odorless and starchy taste. Fractures were fibrous in nature. Moisture content plays a major role in crude drug deterioration and, in some cases, leads to active constituent loss due to interactions, which makes moisture content a major parameter for drug quality and purity. LOD was carried out to determine the moisture content and was found to be 9.1%w/w. The ash value is an important qualitative benchmark that can be used to verify sample authenticity and purity. The ash obtained is expressed in % and given as follows: total ash (6.4), acid-insoluble ash (2.5), and water-soluble ash (2.15).

Table 4: It shows fluorescence analysis of powdered roots of *Thysanolaena maxima*

S. No.	Treatment of powder	Visible rays	UV (254 nm)	UV (365nm)
1	Powder as such	Light brown	Black	Brown
2	Powder+50% H ₂ SO ₄	Greenish brown	Black	Black
3	Powder+50% HNO ₃	Brown	Colorless	Faded brown
4	Powder+methanol	Light brown	Dark brown	Brown
5	Powder+1(N) HCL	Light brown	Light brown	Light brown
6	Powder+cold water	Light brown	Dark brown	Brown
7	Powder+hot water	Light brown	Dark brown	Brown
8	Powder+picric acid	Light brown	Black	Black
9	Powder+NH ₃ sol ⁿ	Dark brown	Deep brown	Greenish dark brown
10	Powder+chloroform	Deep brown	Brown	Light brown
11	Powder+GAA	Brown	Black	Dark brown
12	Powder+5 % Iodine	Reddish brown	Black	Black
13	Powder+FeCl ₃	Light brown	Black	Black
14	Powder+Pet.ether	Brown	Black	Black

**Figure 1:** Pharmacognostical evaluation. (a) Whole plant *Thysanolaena maxima*. (b) Roots of *Thysanolaena maxima***Figure 2:** Graph representing zone of inhibition of different extracts of powdered roots of *Thysanolaena maxima*

The extractive value determines the amount of the active constituents in a given amount of plant material when extracted with a solvent. In the present study, the extractive value of *T. maxima* in petroleum ether, dichloromethane, dichloromethane: methanol (1:1), and chloroform water extract was determined. The chloroform water extractive value of *T. maxima* was found to be (3.84%), which is more than dichloromethane: methanol (1:1) (2.21%), dichloromethane (1.68), and Petroleum ether (1.26).

**Figure 3:** (a) Powder microscopy of root *Thysanolaena maxima* using 1:1 (HCl: Water) (b) Powder microscopy of root *T. maxima* using Phloroglucinol**Figure 4:** Reducing power of different extracts of powdered roots of *Thysanolaena maxima*

Various phytoconstituents present in plant materials exhibit fluorescence. In some cases, it glows in the visible range. Many natural materials that do not glow in daylight exhibit fluorescence when exposed to ultraviolet radiation. Although some substances that don't show fluorescence can be turned into fluorescent derivatives using various chemical reagents and chemicals. We can often determine the quality of some crude medications using

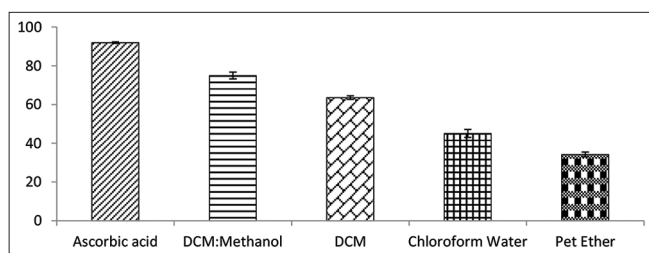


Figure 5: Mean % of DPPH scavenging activity of different extracts of powdered roots of *Thysanolaena maxima*

fluorescence, which is the most essential characteristic in pharmacognostical evaluation. Table 4 shows the findings of the fluorescence examination of *T. maxima* root powder, which showed characteristic coloring after treatment with a variety of chemical reagents.

For herbal pharmaceuticals, standardization is a vital tool, and the microscopic approach is one of the cheapest and simplest ways to start with establishing the right identification of the source material to verify its identity, purity, safety, and quality. Powder microscopy of the plant *T. maxima* root indicated the presence of fibers, stone cells, and graphite belonging to types A and C, that is, random flake graphite and Kish graphite/hyper-eutectic composition, respectively.

Preliminary phytochemical screening is a good approach for identifying bioactive chemicals. It's a quick, low-cost, and straightforward method that reveals the numerous phytochemicals found in the plant. The presence of phytochemicals indicates that the plant could be used to make synthetic pharmaceuticals as a source of precursors. The phytochemical analysis of extracts of *T. maxima* roots revealed the presence of Flavonoids, Saponins, Steroids, alkaloids, and phenols in all extracts, as well as the absence of carbohydrates, tannins, proteins, glycosides, and cellulose.

Oxidation driven by ROS can cause membrane protein damage, cell membrane disintegration, and Oncogenic transformation, which can further trigger or promote the onset of many diseases, such as liver and heart problems and cancer as well.^[27] So, there has been an increased interest in the engagement of reactive oxygen species (ROS) in numerous medical instances in recent years. As a result, antioxidants with radical-scavenging properties may play an important role in disease prevention and treatment. The ability of natural compounds to donate electrons can be measured using DPPH radical scavenging activity.^[28] In the present study, Dichloromethane: methanol (1:1) showed strong free radical scavenging activity with 74.966 ± 1.705 % DPPH radicals scavenged compared to other extracts, i.e., Dichloromethane, Chloroform water, and pet ether (63.570 ± 0.891 , 45.076 ± 2.067 , and 34.229 ± 1.207 , respectively). whereas, the standard ascorbic acid was found to be consistent with other literature findings (91.943 ± 0.373).

Table 5: Phyto-chemical screenings of extracts of powdered roots of *Thysanolaena maxima*

Plant extract	Alkaloid	Carbohydrates	Tannins	Flavonoids	Saponins	Proteins	Steroids	Phenols	Glycoside	Cellulose
Petroleum ether	+	-	-	+	+	-	++	+	-	-
Dichloromethane	+	-	-	++	+	-	+	++	-	-
Dichloromethane: methanol (1:1)	+	-	-	+++	+	-	+	+	-	-
Chloroform water	+	-	-	+	+++	-	+	+	-	-

+++ : Highly present, ++ : Moderately present, + : Slightly present, - : Absent

Table 6: Reducing power of different extracts of powdered roots of *Thysanolaena maxima*

Conc ⁿ µg/ml	Absorbance (700 nm)				
	Ascorbic acid (mean±SEM)	DCM: Methanol (mean±SEM)	DCM (mean±SEM)	Chloroform-water (mean±SEM)	Pet Ether (mean±SEM)
50	2.235±0.083	1.134±0.032	0.993±0.064	0.747±0.061	0.421±0.037
100	2.659±0.056	1.251±0.044	1.070±0.076	0.905±0.041	0.516±0.037
150	2.776±0.059	1.562±0.044	1.204±0.032	1.154±0.036	0.779±0.028
200	2.814±0.061	1.760±0.043	1.369±0.043	1.257±0.043	0.931±0.032
250	2.906±0.056	1.909±0.026	1.612±0.042	1.411±0.037	1.151±0.069
300	3.240±0.076	2.186±0.091	1.721±0.052	1.584±0.027	1.247±0.036

*SEM: Standard Error Mean, µg/ml: Micro gram per milliliter

Table 7: DPPH radical scavenging activity of different extracts of powdered roots of *Thysanolaena maxima*

Extracts	Conc ⁿ µg/ml	Absorbance 517 nm	DPPH radicals scavenged (%)
Ascorbic acid	100	0.078±0.004	91.943±0.373
DCM: Methanol (1:1)	100	0.243±0.017	74.966±1.705
DCM	100	0.353±0.009	63.570±0.891
Chloroform water	100	0.532±0.021	45.076±2.067
Pet. Ether	100	0.637±0.012	34.229±1.207

Table 8: It shows zone of inhibition of different extracts of powdered roots of *Thysanolaena maxima*

S. No.	Extracts	Dose (µg/ml)	Zone of inhibition GM –ve bacteria (<i>S. typhi</i>) (mean±SEM)	Zone of inhibition GM+ve bacteria (<i>Bacillus subtilis</i>) (mean±SEM)
1	Petroleum ether	400	1.167±0.013	1.620±0.012
2	Dichloromethane	400	0.750±0.018	NA
3	Dichloromethane: methanol (1:1)	400	0.800±0.012	0.824±0.015
4	Chloroform water	400	1.230±0.016	NA

*SEM: Standard error mean, µg/mL: micro gram per milliliter, GM+ve: Gram-positive, GM –ve: Gram-negative

The existence of reducing agents, which can exert antioxidant activity by splitting the radical chain reactions and contributing a hydrogen atom, determines a compound's reducing ability.^[29] The existence of reducing agents or antioxidants in all extracts of *T. maxima* (Dichloromethane: methanol (1:1), Dichloromethane, Chloroform water, and Pet Ether) was seen, which might be responsible for its reducing power. Dichloromethane: methanol (1:1) showed the highest reducing power compared to other extracts. The presence of Flavonoids and Phenolic compounds may be the reason behind the antioxidant activity of the extracts.

In the present study, the efficiency of various solvent extracts in the same concentration (Petroleum ether, Dichloromethane, Dichloromethane: methanol (1:1), and Chloroform water) of *T. maxima* roots against GM –ve bacteria (*S. typhi*) and GM +ve bacteria (*B. subtilis*) was tested using the disk diffusion method. The study revealed that Petroleum ether extract has shown the highest antimicrobial activity against both bacteria strains, with a zone of inhibition of 1.167 ± 0.013 for GM –ve and 1.620 ± 0.012 for GM +ve bacteria, followed by Chloroform water

extract with 1.230 ± 0.016 for the GM –ve bacterial strain, and no inhibition was seen for the other bacterial strains. Dichloromethane: methanol (1:1) with 0.800 ± 0.012 (GM –ve) and 0.824 ± 0.015 (GM +ve) with moderate inhibition, whereas Dichloromethane extract with the lowest inhibition for GM –ve with 0.750 ± 0.018 and no inhibition for GM +ve bacteria. Hence, *T. maxima* can be a potential antibacterial candidate.

CONCLUSION

The medicinal plant *T. maxima* was studied using morphological and anatomical powder microscopy, fluorescence analysis, phytochemical screening, antioxidant activity, and antibacterial activity. This study demonstrates that plants have distinct and crucial traits that can be used to identify them. *T. maxima* requires morphological and anatomical analysis because there are only a few publications accessible, which are insufficient for standardization and authenticity as a raw medication. The occurrence of graphite belonging to types A and C, which is random flake graphite and Kish

Graphite and hypereutectic composition are two unusual anatomical properties of this plant. The outcomes of all types of analyses aid in the establishment of quality control standards and the assurance of drug purity. Drug quality factors include photochemical ones as well. When employing the substance as folk medicine, these simple, affordable, and trustworthy standards might be useful even for the untrained. The existence of chemicals with strong antioxidant and antibacterial activity in the roots of *T. maxima* was discovered in this study. The various phenolics and flavonoids makeup of the *Thysanolaena maximum* extract can be attributed to their distinct actions. To identify the bioactive principle and understand the mechanism of action of distinct bioactivities, more research is needed.

REFERENCES

- Yuan H, Ma Q, Ye L, Piao G. The traditional medicine and modern medicine from natural products. *Molecules* 2016;21:559.
- Hoque N, Sohrab MH, Debnath T, Rana MS. Antioxidant, antibacterial and cytotoxic activities of various extracts of *Thysanolaena maxima* (Roxb) Kuntze available in Chittagong hill tracts of Bangladesh. *Int J Pharm Pharm Sci* 2016;8:168-72.
- Choudhary K, Singh M, Pillai U. Ethnobotanical survey of Rajasthan-an update. *Am Eur J Bot* 2008;1:38-45.
- Balakrishnan V, Prema P, Ravindran KC, Robinson JP. Ethnobotanical studies among villagers from Dharapuram taluk, Tamil Nadu, India. *Glob J Pharmacol* 2009;3:8-14.
- Verma KK, Gautam RK, Choudhary A, Gupta GD, Singla S, Goyal S. A review on ethnobotany, phytochemistry and pharmacology on *Rumex hastatus*. *Res J Pharm Technol* 2020;13:3969-76.
- Rahman MA. Indigenous knowledge of herbal medicines in Bangladesh. 3. Treatment of skin diseases by tribal communities of the hill tracts districts. *Bangladesh J Bot* 2010;39:169-77.
- Sharma BK, Jain AK. Ethnobotanical studies of Sikkim: A review. *Int Res J Biol Sci* 2021;10:1-6.
- Panda AK, Misra S. Health traditions of Sikkim Himalaya. *J Ayurveda Integr Med* 2010;1:183-9.
- Sharma S, Mistri R, Chettri A, Bhattarai B, Choubey M. Indigenous knowledge from livelihood perspectives in rural households of Sikkim: An analysis. *Res J Humanit Soc Sci* 2018;9:593-7.
- Ansari AA, Brahma KK, Panda AK. Medicinal uses of flowering plants by the local people of Samdong, East Sikkim. *J Drug Res Ayurvedic Sci* 2009;30:41-8.
- Hynniewta SR, Kumar Y. Herbal remedies among the Khasi traditional healers and village folks in Meghalaya. *Indian J Tradit Knowl* 2003;7:581-6.
- Maity D, Pradhan N, Chauhan AS. Folk uses of some medicinal plants from North Sikkim. *Indian J Tradit Knowl* 2004;3:66-71.
- Singh M, Singh S, Verma D. Morphological and pharmacognostical evaluation of *Moringa oleifera* lam. (Moringaceae): A plant with high medicinal value in tropical and subtropical parts of the world. *Pharmacogn Rev* 2020;14:138-45.
- Ghori SS, Khan M, Qureshi MS. Preliminary phytochemical screening and macroscopic analysis of *Ficus dalhousiae* Miq. *Res J Pharmacogn Phytochem* 2015;7:9-12.
- British Pharmacopoeia. Ash Values, Acid Insoluble Ash, Water Soluble, Extractive and Alcohol Soluble Extractive. Appendix XI. London: Her Majesty's Stationery Office; 1980.
- Kokoski CJ, Kokoski RJ, Slama FJ. Fluorescence of powdered vegetable drugs under ultraviolet radiation. *J Am Pharm Assoc Am Pharm Assoc* 1958;47:715-7.
- Das PK, Vaghela JS, Badore N. Pharmacognostical, phytochemical and fluorescence analysis of the plant *Bougainvillea spectabilis* (willd.). *Res J Pharm Technol* 2021;14:3733-8.
- Akbar S, Hanif U, Ali J, Ishtiaq S. Pharmacognostic studies of stem, roots and leaves of *Malva parviflora* L. *Asian Pac J Trop Biomed* 2014;4:410-5.
- Yadav RN, Agarwala M. Phytochemical analysis of some medicinal plants. *J Phytol* 2011;3:10-4.
- Prabhakar S, Taranjeet K, Vikram SR. Comparative phytochemical screening of six different plant species of Uttarakhand region. *Asian J Pharm Technol* 2014;4:50-2.
- Jayaprakasha GK, Singh RP, Sakariah KK. Antioxidant activity of grape seed (*Vitis vinifera*) extracts on peroxidation models *in vitro*. *Food Chem* 2001;73:285-90.
- Alam MN, Bristi NJ, Rafiquzzaman M. Review on *in vivo* and *in vitro* methods evaluation of antioxidant activity. *Saudi Pharm J* 2013;21:143-52.
- Karagöz A, Artun FT, Özcan G, Melikoğlu G, Anıl S, Kültür Ş, et al. *In vitro* evaluation of antioxidant activity of some plant methanol extracts. *Biotechnol Biotechnol Equip* 2015;29:1184-9.
- Kondragunta VK, Karuppuraj V, Perumal K. Antioxidant activity and Folic acid content in indigenous isolates of *Ganoderma lucidum*. *Asian J Pharm Anal* 2016;6:213-5.
- Balouiiri M, Sadiki M, Ibensouda SK. Methods for *in vitro* evaluating antimicrobial activity: A review. *J Pharm Anal* 2016;6:71-9.
- Preeti T. Antimicrobial activity of Amritarishta prepared by traditional and modern methods. *Asian J Pharm Res* 2014;4:114-6.
- Liao K, Yin M. Individual and combined antioxidant effects of seven phenolic agents in human erythrocyte membrane ghosts and phosphatidylcholine liposome systems: Importance of the partition coefficient. *J Agric Food Chem* 2000;48:2266-70.
- Umamaheswari M, Chatterjee TK. *In vitro* antioxidant activities of the fractions of *Coccinia grandis* L. leaf extract. *Afr J Tradit Complement Altern Med* 2007;5:61-73.
- Montoro P, Braca A, Pizza C, De Tommasi N. Structure-antioxidant activity relationships of flavonoids isolated from different plant species. *Food Chem* 2005;92:349-55.

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