



Phytoconstituents Screening and Analysis of Antioxidant, Membrane Stabilizing, and Thrombolytic Properties of Leaves and Rhizomes of *Schumannianthus dichotomus* (Roxb.) Gagnep

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ABSTRACT

The initial screening of various leaf and rhizome extracts of *Schumannianthus dichotomus* (Roxb) indicated the presence of saponins, phenols, flavonoids, glycosides, phytosterols, and tannins. To verify the therapeutic potential of this plant, known locally as Shital Pati, the antioxidant activity of the different extracts from its leaves and rhizomes were evaluated using eight methodologies. Using a Soxhlet extraction device, solvents like methanol, ethanol, and chloroform were used to extract the powdered leaves and rhizomes of this plant. The membrane stabilizing and thrombolytic potential of the extracts were also assessed using the clot lysis method. In the DPPH scavenging test, the ethanol extracts of the leaf and rhizome showed the highest levels of free radical scavenging activity (IC₅₀ 22.54 and 19.78 µg/mL, respectively), compared to the standard (IC₅₀ 19.51 µg/mL). In the H₂O₂ scavenging assay, the chloroform leaf extract and methanol rhizome extract exhibited the highest inhibition with IC₅₀ values of 2.03 and 1.85 µg/mL, respectively, compared to the standard L-Ascorbic acid at 1.69 µg/mL. The leaf and rhizome extracts displayed promising potential in total antioxidant, total phenolic content, and cupric-reducing antioxidant capacity. The results indicated that all leaf and rhizome extracts have significant potential for membrane stabilizing activity compared to the standard. The methanol leaf and ethanol rhizome extracts of *S. dichotomus* have demonstrated better thrombolytic activity with 23.74±10.71 and 23.56±1.65% clot lysis, respectively, compared to the standard streptokinase at 24.14±9.5%. This investigation confirms the therapeutic potential of *S. dichotomus*'s leaves and rhizome.

Keywords: *Schumannianthus dichotomus* (Roxb) Gagnep, Preliminary screening, Antioxidant, Membrane stabilizing, Thrombolytic activity.

Introduction

Medicinal plants have emerged as a significant element of the global healthcare ecosystem for both human and animal populations. They are not only utilized in the treatment of diseases, but also serve as valuable reservoirs for sustaining overall health. Phytochemicals, which are biologically active chemical compounds present in plants, play a pivotal role in augmenting human health.¹ *Schumannianthus dichotomus* (Roxb) Gagnep (Family: Marantaceae) is used to make Shitalpati, commonly called the cool mat, and is utilized as a traditional bed mat in Bangladesh and Eastern India. It is a rhizomatous plant with a glossy and erect green stem, reaching a height of 3- 5m.² *S. dichotomus* is a newly added medicinal plant in Thailand, the leaf, stem, rhizome, and root of which are phytochemically useful plant components. This plant's rhizomes have historically been used as a diuretic, and treatment for skin conditions, and fever.³ Excessive reactive oxygen species (ROS) are produced in response to various stimuli.

When they surpass the body's antioxidant capacity, they can lead to various health issues such as genotoxicity, cancer, diabetes, and inflammation. Moreover, these ROS can quickly initiate the peroxidation of membrane lipids. Foods can also degrade due to ROS-induced lipid peroxidation.⁴ The use of antioxidants especially natural antioxidants to reduce lipid peroxidation and defend against free radical damage has received a lot of interest.⁵ The circulatory system can experience impediments to blood flow due to the formation of thrombi within blood arteries. This occurrence may lead to outcomes such as hypertension, heart attacks, anoxia, and other related complications.⁵ While thrombolytic medications are commonly utilized in the management of cerebral venous sinus thrombosis, it is important to acknowledge their associated limitations. Although there is evidence suggesting that medicinal plants and their derivatives exhibit antithrombotic properties, there is a noticeable absence of documentation regarding plant-based agents suitable for thrombolysis.⁶ The membrane stabilizing property is utilized to limit the inflammatory response and several studies have shown that extracts of some plants have this property.⁷ This rigorous study involved conducting a series of eight tests to accurately evaluate the antioxidant activity, thrombolytic activity, and membrane-stabilizing properties of plant extracts from the leaves and rhizomes.

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Materials and Methods*Plant materials collection*

A specimen of the *Schumannianthus dichotomus* plant was obtained from Jakiganj, Sylhet, Bangladesh on November 21, 2015, prior to the plant's blooming phase. The plant sample was submitted to Bangladesh

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National Herbarium, Mirpur, Dhaka for identification. The accession no of this plant is 42761.

Preparation of plant samples

The leaves and rhizomes were carefully separated from the plant and subjected to a thorough cleaning and washing process. Following this, the dried plant parts were processed into a coarse powder using a high-efficiency grinder (Mixer Grinder, Japan).

Extraction of plant materials

To obtain the extracts from powdered leaves and rhizomes, 30 g of each sample were successively extracted with 300 mL of methanol, ethanol, and chloroform by Soxhlet extraction apparatus. The extraction process took over six to seven hours at a temperature below the boiling point of each solvent. After extraction, each extract was filtered through filter paper and then transferred to petri dishes. The solvents were allowed to evaporate from the extracts, resulting in dried extracts. The weight of each extract was recorded, as methanol extract, ethanol extract, and chloroform extract. The crude extracts were dried and stored at 4°C in a refrigerated environment with proper labeling until they were needed for analysis.

Preliminary phytochemical screening

The leaves and rhizomes of the plant were extracted using methanol, ethanol, and chloroform. Using standard screening protocols, the extracts were then tested for the presence of various phytochemicals such as alkaloids, carbohydrates, flavonoids, glycosides, saponins, phytosterols/steroids, and tannins.⁸

Determination of DPPH free radical scavenging activity

Plant extracts and diluted solutions with concentrations ranging from 6.25 µg/mL to 200 µg/mL were tested for their capacity to scavenge free radicals using DPPH. Each test tube had 2 mL of 0.004% DPPH solution, added, and the mixtures were then incubated for 30 minutes at room temperature. Absorbance was measured at 517 nm using a spectrophotometer, with L-ascorbic acid used as the standard.^{8,9} Free radical scavenging activity of the extracts was evaluated as percent inhibition and/or IC₅₀ using the following equation:

$$\% \text{ Inhibition} = \left(\frac{A_{\text{control}} - A_{\text{sample}}}{A_{\text{control}}} \right) \times 100$$

$$\text{Or, } \% \text{ Inhibition} = \left(1 - \frac{A_1}{A_0} \right) \times 100$$

Where;

A₁= Absorbance of the extract or standard (sample) and A₀= Absorbance of the control.

Determination of hydrogen peroxide scavenging assay

A standardized procedure was used to assess the effectiveness of plant extracts in removing of hydrogen peroxide, with L-ascorbic acid as the reference standard. The test required filling a test tube with 1 mL of plant extract or a standard solution with concentrations ranging from 6.25 µg/mL to 200 µg/mL. After that, 2 milliliters of H₂O₂ solution were mixed with phosphate-buffered saline (pH 7.4), and incubated at 37°C for ten minutes. The absorbance of the solution was then measured using a spectrophotometer.¹⁰ The percentage of hydrogen peroxide scavenging is calculated as follows:

$$\% \text{ scavenged (H}_2\text{O}_2) = \left\{ \frac{(A_i - A_t)}{A_i} \right\} \times 100$$

Where;

A_i = the absorbance of control and
A_t = the absorbance of test.

Determination of Nitric oxide scavenging assay

The standard used was L-ascorbic acid. Two milliliters of 10-milligram sodium nitroprusside in half a liter of phosphate buffer saline (pH 7.4), prepared according to the BP protocol, were mixed with half a milliliter of extract or sub-fraction of various diluted quantities (ranging from 6.25 µg/ml to 100 µg/ml). Allow the mixture to sit at room temperature for five minutes¹¹. Then, 1 mL of sulphanimide was added to the extracted 0.5 mL of the mix. Allow the mixture to stand at room temperature for another five minutes. Next, a solution containing 1%

naphthyl ethylenediamine dihydrochloride was then added. The mixture was then incubated at room temperature for thirty minutes. Absorbance was measured at 546 nm. Percent inhibition of the NO free radical was measured by using the following equation:

$$\% \text{ Inhibition} = \left(\frac{A_{\text{control}} - A_{\text{sample}}}{A_{\text{control}}} \right) \times 100$$

$$\text{Or, } \% \text{ Inhibition} = \left(1 - \frac{A_1}{A_0} \right) \times 100$$

Where;

A₁= Absorbance of the extract or standard (sample) and
A₀= Absorbance of the control.

Determination of Cupric reducing antioxidant capacity

A 0.5 ml sample of the extract or standard at various diluted concentrations (6.25 µg/µL to 200 µg/µL) was used. Next, a 0.01M Copper (II) Chloride solution (CuCl₂ · 2H₂O) containing 1 milliliter was added, followed by, 1 milliliter of ammonium acetate buffer (pH 7.0). The 0.0075M neocuprione solution was then mixed with 1 mL, and 0.6 milliliters of distilled water was added to reach the final volume of the mixture. The mixture was incubated at room temperature for one hour, and absorbance was measured at 450 nm.¹²

Reducing power assessment

The reducing power of the plant extracts was determined by following a specific protocol. One millilitre (1 mL) of extract or standard of different diluted (ranging from 6.25 µg/µl to 200 µg/µL) was used. To each test tube, 2.5 mL of 0.2 M Phosphate buffer was added, followed by 2.5 mL of 1% Potassium ferrocyanide. Absorbance was measured at 700 nm.¹³ Ascorbic acid at different concentrations was used as the standard. An increase in absorbance of the reaction mixture indicates an increase in reducing power.

$$\% \text{ increase in reducing power} = \left(\frac{A_{\text{test}}}{A_{\text{blank}}} - 1 \right) \times 100$$

Where;

A_{test} = Absorbance of the extract or standard (sample) and A_{blank} = Absorbance of the control.

Determination of total phenolic content

Total phenolic contents in the extracts were determined using the Folin-Ciocalteu reagent.¹⁴ Gallic Acid was used as the standard. One millilitre (1 mL) of extract or standard at various dilutions (6.25 µg/µL to 200 µg/µL) was placed in test tubes. Absorbance was measured at 765nm. The total content of phenolic compounds in plant extracts in Gallic acid equivalents (GAE) was calculated using the following equation:

$$C = \frac{(c \times V)}{m}$$

Where;

C = total content of phenolic compounds, mg/gm plant extract, in GAE,
c = the total concentration of Gallic acid established from the calibration curve (mg/mL),
V = the volume of extract in mL,
m = the weight of crude plant extract in g.

Determination of total flavonoid content

The colorimetric method using aluminum chloride for analysis¹⁵ was utilized to determine the flavonoid content. One millilitre (1 mL) of extract or standard with varying concentrations (6.25 µg/µL to 200 µg/µL) was placed. To each test tube, 3 mL of methanol was added, followed by 0.2 mL of aluminum chloride, 0.2 mL of 1 M potassium acetate, and finally, 5.6 mL of distilled water. The mixture was then incubated at room temperature for 30 minutes, absorbance was measured at 415nm.

Determination of total antioxidant capacity

Test tubes were filled with 1 mL of extract or standard at various diluted concentrations (6.25 µg/µL to 200 µg/µL). A reagent solution (0.6 M sulphuric acid, 28 mM sodium phosphate, and 4 mM ammonium molybdate) was combined with 0.3 mL of the extract. The tubes

containing the reaction solutions were then incubated at 95°C for 90 minutes. The absorbance of the solution was measured at 695 nm.¹⁵

Determination of *in vitro* membrane stabilizing activity

For the *in vitro* membrane stabilizing activity erythrocyte suspension preparation, hypotonic solution-induced hemolysis, and heat-induced hemolysis were carried out according to the conventional method.¹⁶

Preparation of erythrocyte suspension

To prepare the erythrocyte suspension, whole blood was obtained from a healthy human volunteer and placed in Falcon tubes containing 3.1% Na-citrate as an anticoagulant. The blood was centrifuged, and the blood cells were washed three times with a solution of 154 mM NaCl in a 10 mM sodium phosphate buffer (pH 7.4) through centrifugation at 3000 rpm for 10 min.

Hypotonic solution-induced hemolysis

The percentage inhibition of either hemolysis or membrane stabilization was calculated using the following equation-

$$\% \text{ inhibition of hemolysis} = \text{OD}_1 - \frac{\text{OD}_2}{\text{OD}_1} \times 100$$

Where;

OD₁ = optical density of hypotonic-buffered saline solution alone (control)

OD₂ = optical density of test sample in hypotonic solution.

Heat-Induced hemolysis

The percentage inhibition of hemolysis in the test was calculated according to the equation:

$$\% \text{ inhibition of hemolysis} = \left[1 - \frac{\text{OD}_2 - \text{OD}_1}{\text{OD}_3 - \text{OD}_1} \right] \times 100$$

Where, OD₁ = optical density of unheated test sample

OD₂ = optical density of heated test sample and

OD₃ = optical density of heated control sample.

In-vitro thrombolytic activity

The thrombolytic activity of all extracts was evaluated using an *in vivo* clot lysis method¹⁷ with slight modifications, with streptokinase (SK) as the standard substance. Thrombolytic activity was determined by calculating the percentage of clot lysis using the following equation.

$$\% \text{ of clot lysis} = \left[\frac{W_2 - W_3}{W_2 - W_1} \right] \times 100$$

Where, W₁ = weight of micro-centrifuge tube alone

W₂ = weight of micro-centrifuge tube with blood clot

W₃ = weight of micro-centrifuge tube after clot lysis

Statistical analysis

Some data are expressed as Mean ± SEM in Excel, 2016 (Standard Error of Mean). Statistical analysis was conducted using a student t-test assuming unequal variances for two samples. Results with P values less than 0.05 were considered as statistically significant, while those with P values less than 0.001 were deemed statistically highly significant.¹⁸

Results and Discussion

Phytochemical constituents

The preliminary phytochemical screening of different solvent extracts of leaves and rhizomes of *S. dichotomus* (Roxb) revealed the presence of saponins, phenols, glycosides, flavonoids, and steroids, imparting medicinal value to the plant. (Table 1).¹⁹

Nitric oxide scavenging activity

Overproduction of nitric oxide can lead to inflammation and toxic effects. In a study, it was discovered that the chloroform extract from leaves is highly efficient in scavenging nitric oxide.²⁰ The chloroform extract from leaves, with an IC₅₀ value of 6.26 µg/ml, showed greater

effectiveness compared to the methanol and ethanol extracts. Additionally, among the rhizome extracts, the chloroform extract demonstrated the highest potency with the IC₅₀ value of 19.38 µg/ml (Table 2 and Figure 1A).

Hydrogen peroxide scavenging activity

Hydrogen peroxide (H₂O₂) is a significant oxygen metabolite produced by activated phagocytes and oxidase enzymes.²¹ The test to measure its antioxidant potency is simple, accurate, and sensitive. All samples exhibited lower antioxidant potency against H₂O₂ compared to the standard. The most potent leaf extract was methanol with an IC₅₀ value of 1.99 µg/mL, and the strongest rhizome extract was chloroform with an IC₅₀ value of 1.81 µg/mL. Further comparison between leaf and rhizome extracts revealed that methanol and ethanol rhizome extracts had a better inhibition capacity than leaf extracts, while chloroform leaf extracts had a better inhibition capacity than chloroform extracts of the rhizome.

DPPH free radical scavenging activity

DPPH is an artificial free radical used for testing antioxidant activity.²¹ All samples, except for the ethanol extract of leaves, demonstrated greater antioxidant properties than the standard. The chloroform extract of rhizomes showed the highest antioxidant property against DPPH free radicals, while the chloroform leaf extract was the most efficient. The ethanol and chloroform extracts of rhizomes showed greater free radical inhibition capacity compared to the leaf extracts. Additionally, methanol leaf extracts showed better free radical inhibition capacity than methanol rhizome extracts (Table 2 and Figure 1C). The sequence of inhibition percentage of all extracts of leaf and methanol including reference standards are as follows:

Ascorbic Acid > Ethanol Leaf Extracts > Methanol Leaf Extracts > Chloroform

Leaf Extracts - Ascorbic Acid > Ethanol Rhizome Extracts > Chloroform Rhizome Extracts > Methanol Rhizome extracts

Cupric-reducing antioxidant capacity

The cupric reducing antioxidant assay measures antioxidant activity by the increase in absorbance of the reaction mixtures.²¹ In the study, L-ascorbic acid showed the highest reducing capacity. Methanol leaf and rhizome extracts had the most significant cupric-reducing antioxidant capacity at 200 µg/ml, while ethanol and chloroform leaf extracts as well as methanol rhizome extract, showed moderate significance at 100 µg/ml. Ethanol extracts of the rhizome showed the highest cupric reducing capacity compared to methanol and chloroform extracts (Table 3 and Figure 1E).

Reducing power capacity

The Ferric to Ferrous reduction procedure, as determined spectrophotometrically from the production of Perl's Prussian blue color complex, was used to evaluate the reducing power of the extracts.²² The methanol extract exhibited the highest reducing power among leaf extracts (46.25%), which is higher than the standard, L-ascorbic acid (42.53%). Among rhizome extracts, the methanol extract showed the highest activity (50.02%), which is likewise higher than the standard (Table 3 and Figure 1D). Further investigations revealed that methanol and ethanol extracts of both leaf and rhizome exhibit better reducing capacity than those of leaf but chloroform leaf extracts show higher reducing properties than chloroform rhizome extracts. The reducing capacity of both leaf and rhizome tends to decrease with an increase in concentration. The reducing power was found to increase in the following order:

-Leaf: Chloroform extract > Ethanol extract > Methanol Extract

-Rhizome: Methanol Extract > Ethanol Extract > Chloroform extract

This result indicates that the extracts may consist of polyphenolic compounds that usually show great reducing power.

Total phenolic content

The Folin-Ciocalteu Reagent (FCR) was used to determine the total phenolic content of plant extracts, measured in gallic acid equivalents (GAE) per gram of extract (Table 3). The leaf methanol extract had the highest amount of phenolic content among all samples at 1.49 mg/g.

Table 1: Phytochemical constituents of various solvent extracts of leaves and rhizomes of *S. dichotomus*

| Tests | Leaf | | | Rhizome | | |
|---------------|-------------|-------------|--------------|-------------|-------------|--------------|
| | ME Extracts | EE Extracts | ChE Extracts | ME Extracts | EE Extracts | ChE Extracts |
| | - | + | + | + | + | - |
| | - | + | - | + | + | - |
| Alkaloids | - | - | - | - | - | - |
| | - | - | - | + | + | - |
| | - | - | - | - | - | - |
| Carbohydrates | + | - | - | - | - | + |
| | - | - | - | - | - | - |
| | - | - | - | + | + | - |
| Tannins | + | + | - | - | - | - |
| | - | - | - | + | + | + |
| Steroids | + | + | - | + | + | - |
| Saponins | + | - | - | + | + | - |
| Phenol | + | + | - | + | + | - |
| Glycosides | + | + | - | + | + | - |
| Flavonoids | + | - | - | + | - | - |

ME = Methanol Extract, EE=Ethanol Extract, ChE=Chloroform Extract; + = Presence of phytochemicals, - = Absence of phytochemicals.

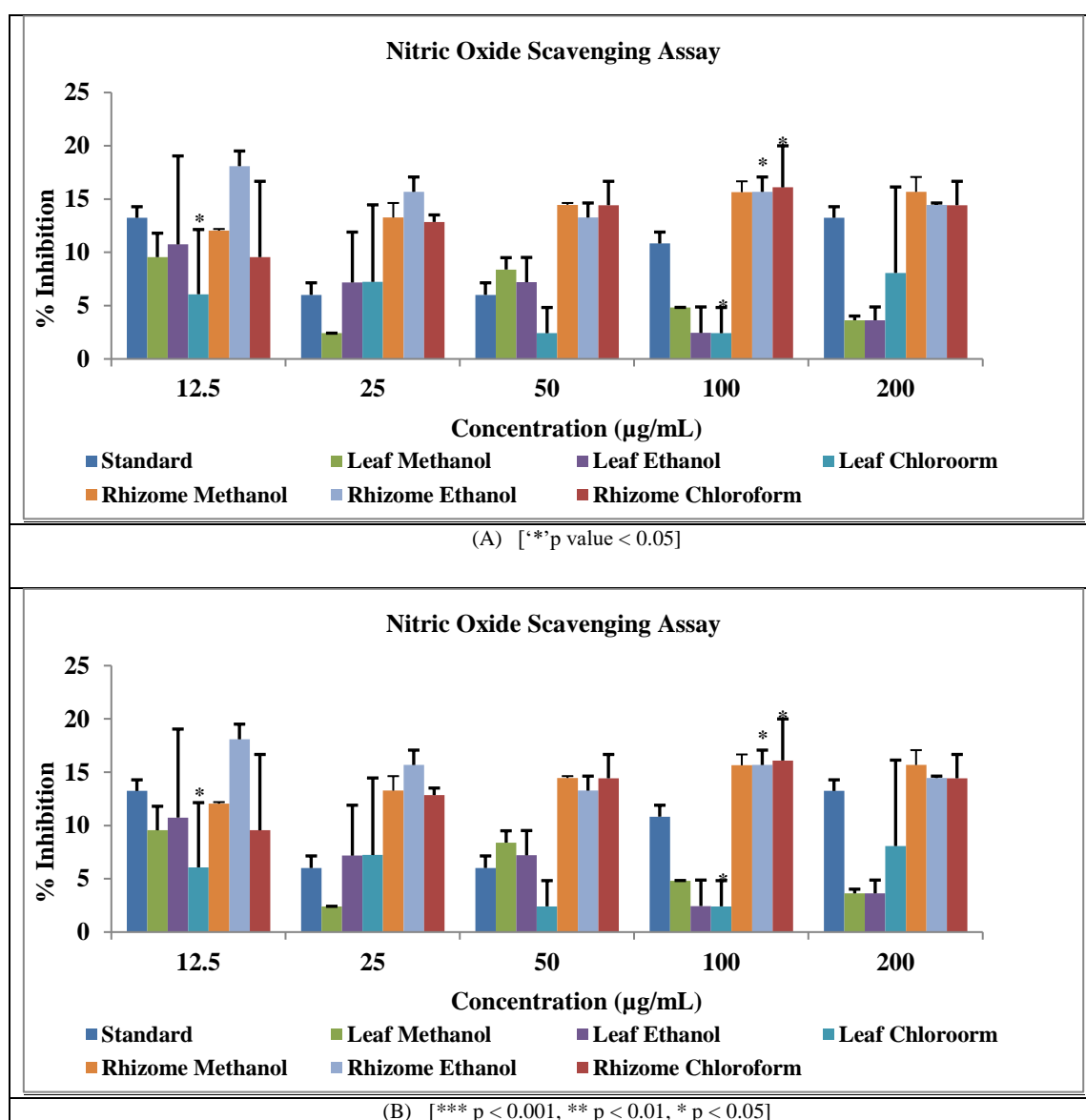
Table 2: IC₅₀ values of the respective extracts in different *in vitro* models

| Sample | Nitric Oxide Scavenging Assay | H ₂ O ₂ Scavenging Activity | DPPH Scavenging Activity |
|--------------------|--------------------------------|---|--------------------------------|
| | IC ₅₀ value (µg/mL) | IC ₅₀ value (µg/mL) | IC ₅₀ value (µg/mL) |
| L -Ascorbic Acid | 43.5 | 1.69 | 19.51 |
| Leaf Methanol | 6.58 | 1.99 | 17.21 |
| Leaf Ethanol | 6.58 | 2.16 | 22.54 |
| Leaf Chloroform | 6.26 | 2.03 | 12.86 |
| Rhizome Methanol | 41.67 | 1.85 | 11.59 |
| Rhizome Ethanol | 42.58 | 1.85 | 2.11 |
| Rhizome Chloroform | 19.38 | 1.81 | 1.84 |

Table 3: Reducing power capacity, total phenolic content, total antioxidant capacity and total flavonoid content of different extracts of *S. dichotomus*.

| Sample | Reducing power capacity % Inhibition | Total phenolic content (mg GAE/g) | Total flavonoid content (mg QE/g) | Total antioxidant capacity (mg AAE/g) |
|--------------------|---|--------------------------------------|--------------------------------------|--|
| L-Ascorbic Acid | 42.53 ± 14.57 | | | |
| Leaf Methanol | 46.25 ± 20.26 | 1.49 ± 0.24 | 11.36 ± 3.69 | 3.60 ± 0.081 |
| Leaf Ethanol | 44.47 ± 19.81 | 1.37 ± 0.04 | 8.77 ± 1.48 | 3.59 ± 0.059 |
| Leaf Chloroform | 30.93 ± 15.57 | 1.48 ± 0.02 | 8.21 ± 0.54 | 3.57 ± 0.05 |
| Rhizome Methanol | 50.02 ± 19.54 | 0.51 ± 0.003 | 11.35 ± 3.69 | 3.51 ± 0.04 |
| Rhizome Ethanol | 47.96 ± 23.23 | 0.52 ± 0.004 | 7.77 ± 1.48 | 3.5 ± .07 |
| Rhizome Chloroform | 29.86 ± 20.27 | 0.52 ± 0.003 | 8.11 ± 0.54 | 3.54 ± .06 |

Values represent Mean ± SEM

**Figure 1:** Percent inhibition of Nitric oxide (A), and hydrogen peroxide (B) free radicals

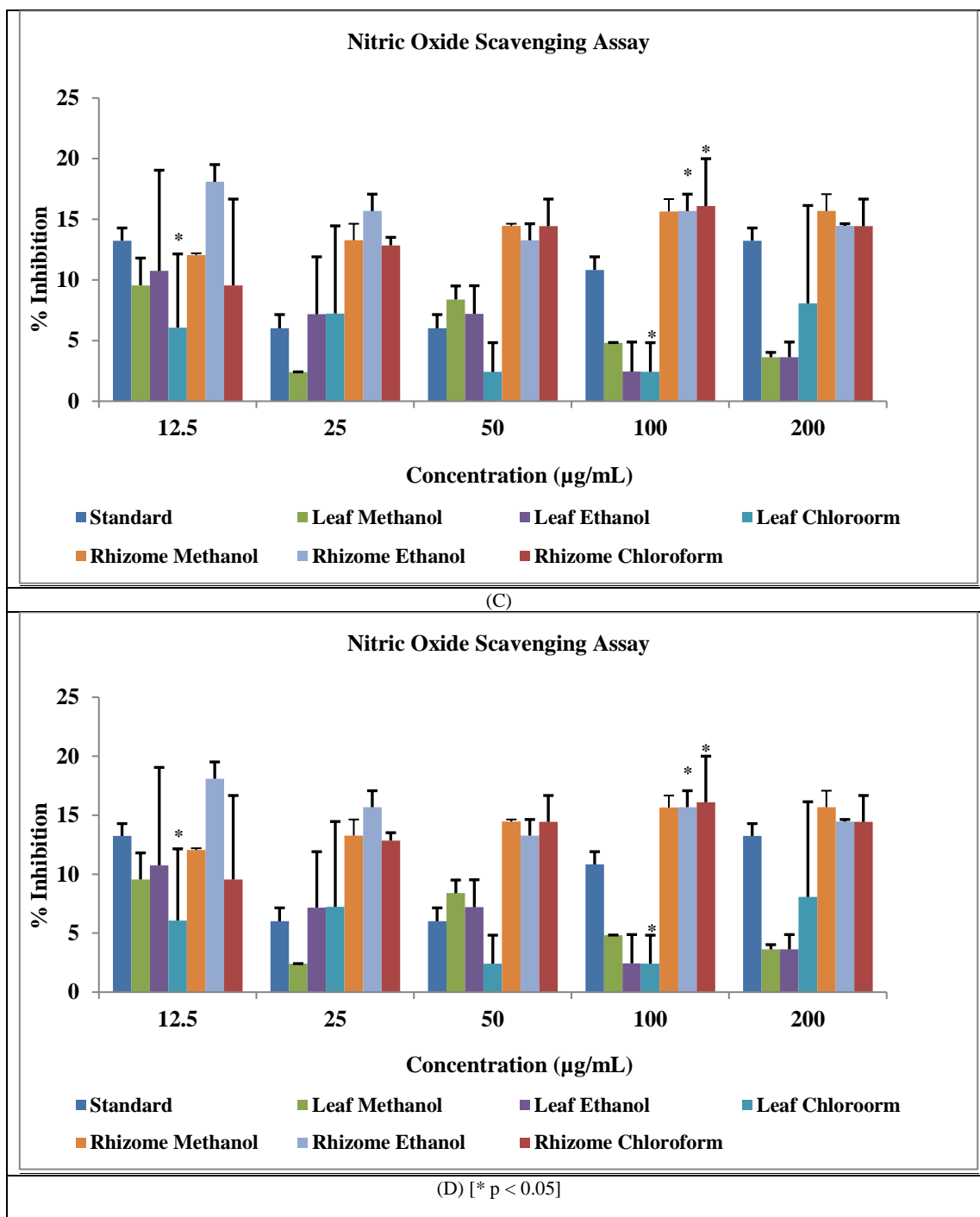


Figure 1 (Continue): Percent inhibition of DPPH free radical (C); reducing power capacity (D)

Phenolic compounds and flavonoids are responsible for the antioxidant capacity of the extracts exhibiting antioxidant activity in biological systems, by acting as scavengers of singlet oxygen and free radicals.²³ A comparison between leaf and rhizome extracts shows that leaf extracts contain more phenolic content than rhizome extracts.

Total flavonoid content

The study aimed to measure the total flavonoid contents of different *S. dichotomus* leaf and rhizome extracts using the aluminum chloride colorimetric method with quercetin as the standard. The results showed that among the methanol, ethanol, and chloroform extracts of leaves, the methanol extract of the plant's leaves contained the highest amount of flavonoid content (11.36 mg QE/g). On the other hand,

the methanol extracts of the rhizome exhibited the highest amount of antioxidant capacity (11.35 mg QE/g) compared to the chloroform and ethanol extracts of the same. (Table 3).²⁴

Total antioxidant capacity

The antioxidant capacity of various extracts was determined by using the extract to reduce Mo (VI) to Mo (V) and create a green phosphate/Mo (V) complex at acidic pH. The results were reported as L-ascorbic acid equivalents (AAE) per gram of plant extracts. The methanol extract from the plant's leaves exhibited the highest antioxidant capacity at 3.60 mg AAE/g. Among the rhizome extracts, the chloroform extract had the highest antioxidant capacity at 3.54 mg AAE/g (Table 3).^{25, 26}

Table 4: The effect of different extracts of leaf and rhizome of *S. dichotomus* on hypotonic solution and heat induced hemolysis of

| Samples | Concentration (mg/mL) | % Inhibition of hemolysis | |
|--------------------|--------------------------|---------------------------|---------------|
| | | Hypotonic induced | Heat induced |
| Control | – | – | – |
| Standard | 0.1 | 40.80 ± 6.94 | 50.91 ± 12.30 |
| Methanol Leaf | 1.0 | 40.14 ± 4.85 | 28.10 ± 7.60 |
| Ethanol Leaf | 1.0 | 38.40 ± 7.78 | 35.78 ± 16.87 |
| Chloroform Leaf | 1.0 | 44.50 ± 7.05 | 46.69 ± 16.33 |
| Methanol Rhizome | 1.0 | 47.10 ± 2.75 | 54.44 ± 10.95 |
| Ethanol Rhizome | 1.0 | 34.82 ± 1.33 | 47.15 ± 10.56 |
| Chloroform Rhizome | 1.0 | 40.80 ± 5.23 | 44.62 ± 9.07 |

erythrocyte

[Values represent Mean ± SEM]

Chloroform Rhizome 22.98 ± 1.68

[Values represent Mean ± SEM]

***In vitro* membrane stabilizing activity**

The study examined the effects of *S. dichotomus* leaf and rhizome extracts on the stabilization of human erythrocyte membrane²⁷. Since the lysosomal membrane is similar to the erythrocyte membrane, so the study extrapolated lysosomal membrane stabilization from the membrane stabilization activity of extracts on erythrocytes. The study compared the effects of the extracts to those of acetylsalicylic acid (ASA) at a dosage of 0.1 mg/mL. Standard ASA demonstrated a 40.80% inhibition of RBC hemolysis in hypotonic solution-induced hemolysis, while the chloroform extract showed the maximum inhibition (Table 4).

***In vitro* thrombolytic activity**

Thrombolytic medications unblock arteries, reduce mortality rates, and prevent damage to tissues like the brain, heart, and limbs. They're used for treating strokes, deep vein thrombosis, pulmonary embolism, and heart attacks.²⁸ A quick and easy clot lysis method was used to test the thrombolytic activity of *S. dichotomus* leaf and rhizome extracts. The standard (streptokinase) exhibited a clot lysis rate of 24.14%. The two extracts that demonstrated the closest thrombolytic activities to the standard were the methanol leaf extract and the ethanol rhizome extract (Table 5).

Table 5: Percent clot lysis of leaf and rhizome extracts of *S. dichotomus*

| Sample | % Clot lysis |
|------------------|---------------|
| Control | 8.68 ± 1.77 |
| Standard | 24.14 ± 9.5 |
| Methanol Leaf | 23.74 ± 10.71 |
| Ethanol Leaf | 20.82 ± 8.63 |
| Chloroform Leaf | 15.4 ± 4.98 |
| Methanol Rhizome | 20.75 ± 5.42 |
| Ethanol Rhizome | 23.56 ± 1.65 |

Conclusion

The plant under investigation contains a variety of chemical compounds. *In vitro* studies support the plant's significant pharmacological potential. Several solvent extracts of *S. dichotomus* show strong antioxidant properties, with chloroform extracts showing the highest activity. The chloroform leaf extract and methanol rhizome extract of the plant exhibit the highest *in vitro* membrane stabilizing activity. The methanol leaf extract and ethanol rhizome extract show nearly identical *in vitro* thrombolytic activity. Further research is needed to identify potent bioactive molecules.

Conflict of Interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

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