RESEARCH ARTICLE

Phyto-therapeutic Potential of Aerial Part of *Sida rhombifolia* for Anti-Inflammatory, Antinociceptive, and Antioxidant Activity

Amit Kumar[1]*, P. Ganesh Mishra[2], S. Dimple Tomar[3], Manish Pathak[2], K. Rupesh Pandey[4], Lubhan Singh[4]

[1]*Department of Pharmacognosy, Kharvel Subharti College of Pharmacy, Swami Vivekanand Subharti University, Meerut, Uttar Pradesh, India*

[2]*Department of Pharmaceutical Chemistry, Kharvel Subharti College of Pharmacy, Swami Vivekanand Subharti University, Meerut, Uttar Pradesh, India*

[3]*Department of Pharmaceutics, Kharvel Subharti College of Pharmacy, Swami Vivekanand Subharti University, Meerut, Uttar Pradesh, India*

[4]*Department of Pharmacology, Kharvel Subharti College of Pharmacy, Swami Vivekanand Subharti University, Meerut, Uttar Pradesh, India*

Received: 24th December, 2022; Revised: 12th January, 2023; Accepted: 07th February, 2023; Available Online: 25th March, 2023

ABSTRACT

Background: To investigate the potential of anti-inflammatory, antinociceptive, and antioxidant activity of different extracts of *Sida rhombifolia.*

Materials and Methods: The successive extraction of dried aerial parts of *S. rhombifolia* was performed with the help of the soxhlet apparatus by pet. ether, chloroform, acetone, ethanol and water were used as a solvent. Anti-inflammatory activity was performed using the carrageenin-induced edema model in the rat paw method. Antinociceptive activity was performed using the mouse writhing and hot plate tests. Antioxidant activity was determined through the ability of hydrogen peroxide scavenging.

Result: The ethanolic extract (200 mg/kg.) oral showed maximum anti-inflammatory activity 51.42 (maximum, %inhibition) after 2 hours. The ethanolic extract (200 mg/kg.) oral showed maximum % inhibition of writhing 57.74 for writhing test and 2.15 ± 0.02 time (sec) of jumping for hot plate test. The ethanolic extract of the drug showed high scavenging (59.25%) of hydrogen peroxide.

Conclusion: On successive extraction process of aerial parts of *S. rhombifolia* reported that different ethanolic extracts are more effective as anti-inflammatory, antinociceptive, and antioxidant activity, respectively.

Keywords: Anti-inflammatory, Antinociceptive, Antioxidant, *Sida rhombifolia.*

International Journal of Pharmaceutical Quality Assurance (2023); DOI: 10.25258/ijpqa.14.1.16

How to cite this article: Kumar A, Mishra PG, Tomar SD, Pathak M, Pandey KR, Singh L. Phyto-therapeutic Potential of Aerial Part of *Sida rhombifolia* for Anti-Inflammatory, Antinociceptive, and Antioxidant Activity. International Journal of Pharmaceutical Quality Assurance. 2023;14(1):91-95.

Source of support: Nil.

Conflict of interest: None

INTRODUCTION

Arrowleaf Sida (*Sida rhombifolia*) is an annual or perennial shrub (family- Malvaceae) native to all tropical and subtropical countries.[1] Common names are countary mallow (English), chittamadi (Srilanka), atibala (Sanskrit), huang hua mu (china), bala, mahabala (Hindi), petoria bassie (Africa).[2] This plant contains different phytoconstituents like alkaloids (indole), flavonoids, steroids, phenolic compounds, ascorbic acid, beta-carotene, calcium, carbohydrates, ephedrine (pseudoephedrine), gums, saponin, mucilage, protein, saponin, tannins, triterpenoids, vascin, vasicine.

The habitual uses of *S. rhombifolia was* reported as antipyritic, ulcers, boils, urinary diseases, asthma[3], toothaches, as laxative, applied in the vagina as an antiseptic,[3] and treatment of gout.[4] The *S. rhombifolia* reported different pharmacological actions like hepatoprotctive, anticancer, antibacterial, antifungal, anti-gout, anthelmintic, and hypoglycemic activities.[5]

MATERIALS AND METHODS

Collection and Authentication of Plants Material

The aerial part of *S. rhombifolia* (Figure 1) was obtained from botanical garden of Forest Research Institutes, Dehradun,

**Author for Correspondence: a.k.punia85@gmail.com*

Figure 1: Flower of *S. rhombifolia*

UK, India. Dr. Shiddamallayya N (senior scientist) at N.A.D.I (Ay), Bangalore, India, authenticated plant material. Drug Authentication/SMPU/NADRI/BAG/2011-12/540.

Extraction

The successive extraction of dried aerial parts of *S. rhombifolia* were performed with the help of soxhlet apparatus by pet. ether, chloroform, acetone, ethanol & water were used as a solvents.[6-8]

Anti-inflammatory Activity

Anti-inflammatory activity was performed by using carrageenin-induced edema in the rat paw method. After 16 hours. fasting, 42 rates equally divided into seven groups. Group first received 0.5% carboxy methyl cellulose at a dose of 1-mL/100 gm, and served as a control group. Group second to six, animals received a suspension of 0.5% w/v CMC of pet ether, chloroform, acetone, ethanol, and water extract, respectively, with a 200 mg/kg dose orally. Group seven received a standard drug (indomethacin) orally with a dose of 10 mg/kg. After one hour of administration, 0.1 mL (l% w/v carrageenin in normal saline) was injected into the right hind paw to induced edema in animals. Plethymometer was used to measurement of the paw volume of animals.[9-11]

Detail Study Plan: For Anti-inflammatory Activity

The study was conducted consisting of 7 groups each containing six animals.

1st Group–0.5 w/v CMC, at a dose of 1-mL/100 gm.

2nd To 6th Group–Suspension of 0.5% w/v CMC of pet ether, chloroform, acetone, ethanol, and water extract, respectively, with 200 mg/kg dose, orally.

7th group–received the standard drug (indomethacin), with dose (10 mg/kg), orally.[12-16]

Antinociceptive Activity

- Mouse Writhing Test

The rates were divided into seven groups, every group containing eight animals. The doses (100 mg/kg) of *S. rhombifolia* were administered 1 to 5 groups, while groups 6 and 7 administered a 10 mL/kg dose of distilled water and 5 mg/kg dose of indomethacin, respectively. Following 30 minutes, 0.6% acetic acid solution in normal saline was injected with I.P (10 mL/kg). After acetic acid injection, the writhe numbers were counted for 15 minutes.

$$(N\text{-}Nt/N)100$$

Where,

Nt and N was an average number of writhes (test group) and average number of writhes (control group),[17-19] respectively.

Detail Study Plan: Mouse writhing test

The study was conducted consisting contain seven groups, each containing eight animals.

1st To 5th group- Administration of 200 mg/kg each group, Pet. ether, chloroform, acetone, ethanol, and water extract, respectively.

6th group - dis. water (10 mL/kg)

7th group- Indomethacin (5 mg/kg). respectively.

(Ahmed et al., 2001)[20]

- *Hot Plate Test*

The animals were positioned on a hot plate for a maximum time of 30 seconds, at a temperature of 55°C. Reaction time was measured on time i.e 30, 45, 60, and 90 minutes by animal licking paws and jumping responses after I.P administration of 200 mg/kg of different extracts to seven different groups. Reference drug morphine was used with a dose of 10 mg/kg. The study was conducted consisting contain seven groups, each containing eight animals.

1st group- Dis. water (10 mL/kg), treated as control.

2nd to 6th group- Administration of 200 mg/kg each group Pet. ether, chloroform, acetone, ethanol, and water extract, respectively.

7th group- Reference drug morphine was used at a dose of 10 mg/kg.[21]

Antioxidant Activity (Hydrogen Peroxide Scavenging)

The different extracts of *S. rhombifolia* were used to determine antioxidant activity on behalf of hydrogen peroxide scavenging capacity. The phosphate buffers (pH 7.4) were used to prepare the sample of different extracts, standard and ascorbic acid. The extracted sample with 0.6 mL hydrogen peroxide solution (2 mM hydrogen peroxide with buffers) was taken in different test tubes containing 0.5 mL standard. Phosphate buffers were taken in control group test tube. These solutions were standing for 10 minutes at room temperature. The absorbance of the different solutions was measured by ultraviolet-visible spectroscopy at 230 nm. The % inhibition was calculated by:

$$Control_{(Absorbance)} - Sample_{(Absorbance)} \times 100/Control_{(Absorbance)}$$

Where

Sample–Standard and extract solution.

Control–Hydrogen peroxide in phosphate buffer.[8]

The result of antioxidant activity was expressed as IC_{50}. (Narendhirakannan *et al.,* 2010).[22]

RESULT AND DISCUSSION

Anti-inflammatory Activity of Extracts

Animal activities were conducted as per CPCSEA guidelines.

Anti-inflammatory activity was performed by pet. ether, chloroform, acetone, ethanol and water extracts. The ethanolic

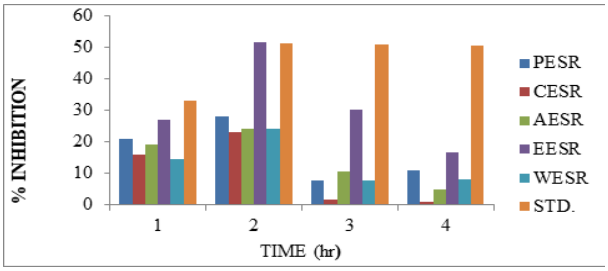

Figure 2: Graphical representation of % inhibition.

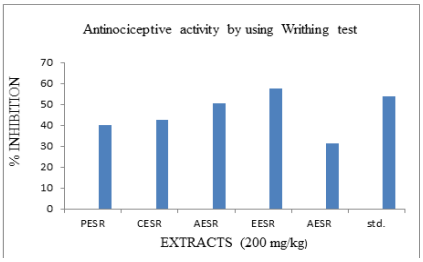

Figure 3: Graphical representation of %inhibition in mouse writhing test.

extract (200 mg/kg. oral) showed maximum %inhibition 51.42 after 2 hours while pet. ether, chloroform, acetone, and water extracts showed maximum %inhibition 28, 22.85, 24, and 24, respectively but the standard drug indomethacin (10 mg/kg) showed 51.14 (Table 1 and Figure 2). From the above result, we can conclude that ethanolic extract is more comparable to standard drug so ethanolic extract is more effective as an anti-inflammatory.

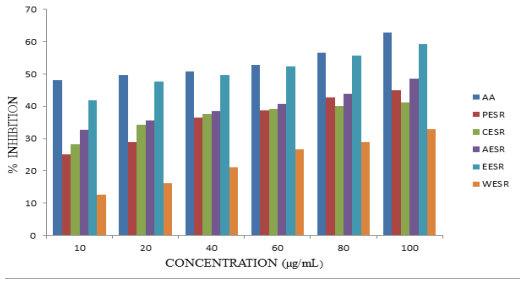

Figure 4: Antioxidant activity by using hydrogen peroxide-scavenging model.

Antinociceptive Activity of Extracts

Writhing Test

Antinociceptive activity was performed by using different extracts i.e. pet. ether, chloroform, acetone, ethanol, and water extracts. The ethanolic (200 mg/kg. oral) and acetone extract (200 mg/kg) showed maximum % inhibition of writhing 57.74 and 50.36 after 15 minutes while pet. ether, chloroform, and water extracts showed 40.14, 42.61, and 31.35, respectively in the Writhing test but the standard drug indomethacin (5 mg/kg) showed 53.97 (Table 2 and Figure 3), from the above result we can conclude that ethanolic and acetonic extracts are more comparable to the standard drug so they have more effectiveness as antinociceptive activity.

Hot Plate Test

Antinociceptive activity was performed by per. ether, chloroform, acetone, ethanol and water extracts with respect to standard drug. The ethanolic extract (200 mg/kg. oral) and

Table 1: Anti-inflammatory activity of *S. rhombifolia* by carrageenan-induced hind paw edema.

S. No.	*Groups*	*Dose*	*Paw volume (mL) ± S.E.M*			
			1 hour	*2 hour*	*3 hour*	*4 hour*
1.	Vehicle (control)	1mL/100gm	1.64 ± 0.02	1.75 ± 0.03	1.30 ± 0.01	1.21 ± 0.87
2.	Pet. Ether	200 mg/kg	1.30 ± 0.03	1.26 ± 0.02	1.20 ± 0.02	1.08 ± 0.04
3.	Chloroform	200 mg/kg	1.38 ± 0.04	1.35 ± 0.04	1.28 ± 0.03	1.23 ± 0.03
4.	Acetone	200 mg/kg	1.36 ± 0.04	1.33 ± 0.04	1.16 ± 0.02	1.15 ± 0.03
5.	Ethanol	200 mg/kg	1.25 ± 0.03	1.21 ± 0.03	1.10 ± 0.02	1.01 ± 0.02
6.	Water	200 mg/kg	1.40 ± 0.03	1.33 ± 0.02	1.20 ± 0.02	1.11 ± 0.16
7.	Standard (Indomethacin)	10 mg/kg	1.10 ± 0.12	0.85 ± 0.08	0.64 ± 0.05	0.60 ± 0.04

The volume of hind paw oedema was expressed as mean ± S.E.M, *Data differs significantly ($p \leq 0.01$) when compared against treated group (normal saline). Data differs significantly ($p \leq 0.05$) when compared Indomethacine treated group.

Table 2: Effect of different extracts of *S. rhombifolia* by writhing tests.

Animal Group	*Dose (mg/kg)*	*Writhes Response*	*% Inhibition*
Control		35.50 ± 0.86	
Pet. Ether extract	200	21.25 ± 0.59	40.14
Chloroform extract	200	20.37 ± 0.53	42.61
Acetone extract.	200	18.62 ± 0.32	50.36
Ethanol extract.	200	15.0 ± 0.42	57.74
Water	200	24.37 ± 0.56	31.35
Standard	5	16.34 ± 6.30	53.97

The writhing response was expressed as mean ± S.E.M, *Data differs significantly ($p \leq 0.01$) when compared against the normal saline. Data differ significantly ($p \leq 0.001$) when compared with Indomethacin treated group.

Table 3: Effect of different extract of *S. rhombifolia* by using hot plate.

Animal Group	*Dose (mg/kg)*	*Time (sec.)*			
		0	30	45	60
Control		3.14 ± 0.08	3.01 ± 0.10	2.68 ± 0.02	2.65 ± 0.07
Pet. Ether	200	2.0 ± 0.21	3.37 ± 0.37	2.0 ± 0.23	1.18 ± 0.09
Chloroform	200	3.75 ± 0.04	4.78 ± 0.03	4.47 ± 0.06	3.80 ± 0.04
Acetone	200	3.33 ± 0.10	4.30 ± 0.04	4.12 ± 0.02	3.63 ± 0.02
Ethanol	200	2.10 ± 0.06	4.02 ± 0.03	2.46 ± 0.07	2.15 ± 0.02
Water	200	2.45 ± 0.05	3.96 ± 0.03	2.0 ± 0.068	1.25 ± 0.09
Standard	10	2.25 ± 0.20	5.14 ± 0.08	3.04 ± 0.07	2.52 ± 0.80

The frequency of rat paw licking, jumping or shaking off from the surface was expressed as mean ± S.E.M, *Data differs significantly ($p \leq 0.01$) when compared against the normal saline with treated group. Data differs significantly ($p \leq 0.05$) when compared with Morphine treated group.

Table 4: Antioxidant activity of *S. rhombifolia* using hydrogen peroxide-scavenging model

Conc.(µg/mL)	*%antioxidant activity*					
	Ascorbic acid	*S. rhombifolia*				
		Pet. ether	*Chloroform*	*Acetone*	*Ethanol extract*	*Aqueous extract*
10	47.98	25.12	28.30	32.74	41.93	12.66
20	49.59	28.95	34.35	35.56	47.58	16.14
40	50.80	36.37	37.58	38.38	49.59	21.14
60	52.82	38.79	39.10	40.80	52.41	26.77
80	56.65	42.82	40	42.82	55.64	28.79
100	62.85	44.85	41.20	48.62	59.25	32.80

acetone extract showed 2.15 ± 0.02 and 3.63 ± 0.02 time (sec) for jumping after 60 minutes while pet. ether, chloroform, water extracts showed 1.18 ± 0.09, 3.80 ± 0.04, 1.25 ± 0.09, respectively in hot plate test but the standard drug morphine (10 mg/kg) showed 2.52 ± 0.80 sec (Table 3), from the above result we can conclude that ethanolic and acetonic extracts are more comparable to the standard drug so they have more effectiveness as antinociceptive activity

Antioxidant Activity of Extracts

Antioxidant activity was performed by using different extracts of *S. rhombifolia* with the help of the hydrogen peroxide-scavenging model. Ethanolic extract of the drug reported high scavenging (59.25%) of hydrogen peroxide. The aqueous extract showed the least scavenging (30.80%) while the standard (Ascorbic acid) showed 62.85 (Table 4 and Figure 4). After comparison, ethanolic extract showed the comparative result with the ascorbic acid. From the above result, it was observed that ethanolic extract of *S. rhombifolia* showed maximum scavenging activity.

CONCLUSION

In anti-inflammatory activity, the ethanolic extract (200 mg/kg, orally) showed maximum %inhibition of 51.51 after 2 hours while petroleum ether, chloroform, acetone, and water extracts showed maximum % inhibition of 23.18, 14.54, 24.18, and 19.60, respectively, whereas Indomethacin (10 mg/kg) showed 58.16%. From the above result, we can conclude that ethanolic extract is more effective when compared to standard drugs.

In antinociceptive activity, the ethanolic and acetonic extract (200 mg/kg, orally) showed maximum %inhibition of writhing 58.33 and 50 after 15 minutes. In contrast, petroleum ether, chloroform, and water extracts showed 41.16, 45.83, 33.33, respectively. Still, the standard drug indomethacin (5 mg/kg) showed 62.50 in the writing test while in hot plate test. The ethanolic and acetone extract (200 mg/kg, orally) showed more comparable results with standard Morphine (10 mg/kg). From the above result, we can conclude that ethanolic. Acetonic extracts are more comparable to the standard drug, so they are more effective as antinociceptive activity.

In antioxidant activity, the free radical scavenging activity was done by the H_2O_2 scavenging model. It was observed that the ethanolic extract (200 mg/kg) of *S. rhombifolia* showed the maximum scavenging (59.25%) of hydrogen peroxide and aqueous extract show the least scavenging (32.80%) while the standard ascorbic acid showed scavenging (62.85%).

After comparison with a standard, ethanolic extract of *S. rhombifolia* showed the comparative result so ethanolic extract is more effective as antioxidant activity.

Conclusively, it revealed from the present study that *S. rhombifolia* leaves and stems has anti-inflammatory, antinociceptive & antioxidant activities due to alkaloids, flavonoids and phenolic constituents in the plant.

REFERENCES

1. Asdaq SMB, Kubavat JB. Role of Sida cordifolia leaves on biochemical and antioxidant profile during myocardial injury. Journal of ethnopharmacology. 2009;124:162-165.
2. Jain A, Choube S, Singour P K, Rajak H, Pawar R S. S. cordifolia (Linn)- An overview. Journal of applied pharmaceutical science. 2011; 01 (02): 23-31.
3. Singh K K, Maheshwari J K. Traditional phytotherapy of some

medicinal plants by tharus of nanital dist, uttar Pradesh, india. Journal of ethnopharmacology.1994; 32, 51-58.
4. Antoniolli A R, Franzotti E M, Santos C V F, Rodrigues H M S L, Mourao, R H V, Andrade M.R. Anti-inflammatory, analgesic activity and acute toxicity of S. Cordifolia. Journal of Ethopharmacology. 2000; 72: 273-278.
5. Singh A, Dhariwal S, Navneet. Traditional uses, antimicrobial potential, pharmacological properties and phytochemistry of S. rhombifolia Linn. International Journal of Innovative Pharmaceutical Sciences and Research. 2018; 6(02), 54-68.
6. Franco C I F, Morasis L C S L, Quintas L J, Antoniolli A R. CNS Pharmacological effects of the hydroalcoholic extract of S. cordifolia leaves. Journal of Ethnopharmacology. 2005; 98:275-279.
7. Gupta S R, Nirmal S A, Patil R Y, Asana G S. Anti-arthritic activity of various extracts of S. rhombifolia aerial parts. Natural product research. 2009; 23(8):689-695.
8. Narendhirakannan R T, Limmy T P. In Vitro antioxidant studies on ethanolic extracts of leaf, stem and root of S. rhombifolia L. International Journal of Pharma and Bio sciences. 2010; 2:105-106.
9. Rao K S, Mishra S H. Anti-inflammatory and hepatoprotective activities of S. rhombifolia Linn. Indian Journal of Pharmacology. 1997; 29(1):110-116.
10. Shekhar T C, Bahuguna Y M, Vijender S. Anti-Inflammatory activity of ethanolic stem extracts of Rubi cardifolia Linn. in rats. IJRAP. 2010; 1:126-130.
11. Narendhirakannan R T, Limmy T P. Anti-inflammatory and antioxidant properties of S. rhombifolia stems and roots in adjuvant induced arthritic rats. Immunopharmacology and Immunotoxicology. 2011; 34 (2) 326-336.
12. Khalil M S, Sperotto S J, Manfron P M. Anti-inflammatory activity of the hydroalcoholic extract of leaves S. rhombifolia L. (malvaceae). Bonaerense. 2006; 25:260.
13. Sulaiman M R, Moin S, Alias A, Zakaria Z A. Antinociceptive and anti-inflammatory effects of S. rhombifolia L. in various animal models. Research Journal of pharmacology. 2008; 2:13-16.
14. Venkatesh S, Reedy Y S M, Suresh B, Reedy B M, Ramesh M. Antinociceptive and anti-inflammatory activity of S. rhombifolia. Journal of Ethopharmacology. 1999; 67 (29):279-232.
15. Zakaria Z S, Rahman N L A, Sulaiman, M.R. Antinociceptive and anti-inflammatory activities of the chloroform extract of Bauhinia purpurea L. in animal models. International Journal of TropicalMedicine. 2009; 4:140-145.
16. Franzotti E M, Santos C V F, Rodrigues H M S L, Mourao M R, Andrade A R, Antoniolli. Anti-inflammatory, analgesic activity and acute toxicity of S. cordifolia L. Journal of Ethnopharmacology. 2000; 72: 273-278.
17. Ahmed S, Sultana M, Hasan M M, Azhar, I. Analgesic and antiemetic activity of cleome viscose L. Pak. J. Bot. 2011; 43:119-122.
18. Antoniolli A R, Franzotti E M, Santos C V F, Rodrigues H M S L, Mourao, R H V, Andrade M.R. Anti-inflammatory, analgesic activity and acute toxicity of S. Cordifolia. Journal of Ethopharmacology. 2000; 72: 273-278.
19. Sulaiman M R, Moin S, Alias A, Zakaria Z A. Antinociceptive and anti-inflammatory effects of S. rhombifolia L. in various animal models. Research Journal of pharmacology. 2008; 2:13-16.
20. Rahman M A, Paul L C, Solaiman M, Rahman A A. Analgesic and cytotoxic activities of S. rhombifolia Linn. Pharmacology. 2011; 2:707-714.
21. Franzotti E M, Santos C V F, Rodrigues H M S L, Mourao M R, Andrade A R, Antoniolli. Anti-inflammatory, analgesic activity and acute toxicity of S. cordifolia L. Journal of Ethnopharmacology. 2000; 72: 273-278.
22. Pawa R S, Jain A, Sharma P, Chaurasiya P K, Singour P K. In vitro studies on S. cordifolia Linn for anthelmintic and antioxdant Properties. Chinese Medicine. 2011; 2:47-52.