Available online at www.ijpsdr.com
International Journal of Pharmaceutical Sciences and Drug Research 2010; 2(3): 213-215

Research Article ISSN 0975-248X

Evaluation of Analgesic & Anti-inflammatory activity of Hydro-alcoholic Extract of *Desmostachya bipinnata* (L.) Stapf root on Experimental Animals

Vinod Kumar*, Rajeev Kumar, Sanjay Yadav, Satyawan Singh, Surendra Nath Pandeya

Department of Pharmacy, Saroj Institute of Technology and Management, Sultanpur Road, Lucknow, Pin-226002, Uttar Pradesh, India

ABSTRACT

Desmostachia bipinnata (L.) Stapf (Gramineae) occurs widely in Egypt. This plant used traditionally as analgesic, antipyretic, anti-inflammatory, asthma, thirst, jaundice, vaginal discharges, vesicle calculi, diseases of bladder, skin eruptions, vomiting, and sedative to pregnant uterus. So, in the present study, *Desmostachia bipinnata* roots were explored for their anti-inflammatory (carrageenan induced paw oedema) and analgesic potential (Hot plate method) on experimental model and compared to standard drugs (Indomethacin for anti-inflammatory activity, Analgin for analgesic activity). In the carrageenan-induced rat paw edema test for acute inflammation, the extract of *Desmostachya bipinnata* in dose of 400 mg/kg body weight showed 62.5% inhibition of edema, at the end of 3h. Phytochemical analysis revealed the presence of tannins, saponins, alkaloids, sterols and flavonoids. The pharmacological activities of medicinal plants are usually due to their secondary metabolites. Some of the constituents of the extract have been documented to possess analgesic and anti-inflammatory activities. The results of this study provide evidence for the analgesic and anti- inflammatory activity of the hydro-alcoholic extract of *Desmostachya bipinnata* root thus supporting its traditional use in painful inflammatory conditions.

Keywords: Carrageenan, Analgesic, Anti-Inflammatory, *Desmostachya bipinnata,* Indomethacin.

INTRODUCTION

Analgesia is the inability to feel pain while still conscious. From the Greek an-, without + algesis, sense of pain. Salicylates are the class of compounds that are widely valued for their analgesic, antipyretic and anti-inflammatory properties. [1-2] The most commonly known and used salicylates are salicylic acid (also called 2-hydroxybenzoic acid), aspirin (acetylsalicylic acid -ASA) and sodium salicylates. They are used extensively for the relief of headache, inflammation, arthritis pain, and some are employed in the treatment of heart attacks and strokes in the elderly. [3] Their mode of action is the inhibition of the synthesis of prostaglandin and its derivatives that cause inflammation, pain, rise in temperature and related diseases. [1-4]

Despite the progress made in medical research during the past decades, the treatment of many serious diseases is still problematic. Chronic inflammatory diseases remain one of the world's major health problems. [5-7] Inflammation is the response of living tissues to injury. It involves a complex array of enzyme activation, mediator release, extravasations of fluid, cell migration, tissue breakdown and repair. [8-9] Inflammation has become the focus of global scientific research because of its implication in virtually all human and animal diseases. The conventional drugs used to ameliorate this phenomenon are either too expensive, side effects or toxic and not commonly available to the rural folks that constitute the major populace of the world. [7, 10-11] Therefore, development of newer and more powerful anti-inflammatory drugs with lesser side effects is necessary. [12] This study therefore seeks to examine *Desmostachya bipinnata* Stapf for anti-inflammatory activity and analgesic effects since pain is one of the cardinal signs of inflammation. *Desmostachya bipinnata* is an erect, tall, branched from the base perennial herb 30-90 cm height found in forest undergrowth, sandy areas almost throughout India. The plant used traditionally as analgesic, antipyretic, anti-inflammatory, asthma, thirst, jaundice, vaginal discharges, vesicle calculi, diseases of bladder, skin eruptions, vomiting, and sedative to pregnant uterus.

***Corresponding author: Mr. Vinod Kumar,**
Department of Pharmacy, Saroj Institute of Technology and Management, Sultanpur Road, Lucknow, Pin-226002, Uttar Pradesh, India; Tel.: +91-9450793106;
E-mail: vinodsitm09@gmail.com

MATERIALS AND METHODS

Collection of plant material

The roots of *D. bipinnata* were collected from wild sources surrounding Lucknow, U.P., India and authenticated by Dr. Tariq Hussain, scientist Head of department herbarium, N. B. R. I., Lucknow, Uttar Pradesh, India, Voucher no. 96520.

Preparation of plant material

250 g of air dried powder was extracted with water: ethanol (50:50) by cold maceration in a specified strength in a closed flask for twenty-four hours, shaking frequently during six hours and allowing standing for eighteen hours. The extract was filtered, concentrated under reduced temperature using rotary evaporator and dried extract was subjected for phytochemical and biological activity on experimental animal model.

Animal selection

Healthy adult Wister albino rats were used for all the experimental study of various parameters of inflammatory activity and Analgesic activity. The body weight of these animals varied from 160 to 190 g and the age from 4-6 months. Male or female rats were selected to anti-Inflammatory, Analgesic activity. Animals were housed individually with free access to food and water under standard condition and the basal food intake, body weights to the nearest gram were noted. The animals were starved 18 h prior to starting biological activity.

Experimental procedures

Phytochemical screening

The hydro-alcoholic extract of the root of *D. bipinnata* was evaluated for the presence of flavonoids, tannins, alkaloids, saponins, glycosides and sterols/triterpenes. [13]

Drugs and chemicals

Analgin, Indomethacin (standard drugs), Carrageenan, and Tween 80.

Biological activity

Anti-inflammatory activity

Carrageenan-induced paw oedema

Healthy albino rats of the either sex (160-190 g) were divided into 5 groups of 6 animals each. They were fasted for 18 h prior to the test, with free access to water. Group I received the vehicle (0.9% normal saline in 0.1 ml of 2% Tween 80) and served as the control group. Acute inflammation was produced by the sub-plantar administration of 0.1 ml of 1% carrageenan in normal saline that contained Tween 80 in the right paw of rats. [14] Groups II, III, IV and V were treated with standard drug (Indomethacin 5mg mg/kg body weight) [15], hydro-alcoholic extract (200, 300 mg & 400 mg/kg), respectively. All drugs/vehicle were administered orally (p. o.). Thirty minutes after the drug treatment, each rat was administered 0.1 ml of 2% Tween-80 orally and housed separately in metabolic cages, with special provision to examine. The paw edema episodes were observed for 4 h. The cumulative height in plethysmometer was noted (in ml.) at the end of the 4th h. Percentage inhibition of inflammation was calculated using the mean height of mercury. Anti-inflammatory activity was determined term of percentage of oedema the control group using the formula

$$\% \text{ inhibition} = (1\text{-}Vt/\ Vc)\ 100,$$

Vt & Vc are the mean relative changes in the paw volume of the test & control group respectively. [16]

Data were analyzed by student's t-test & the level of significant was set at $p<0.001$.

Table 1: Schedule for screening of hydro-alcoholic extracts for anti-inflammatory activity

Groups	Dose (p.o)
Control	0.1ml 2% Tween 80 Saline.
Standard	5 mg/kg. b.w. of Indomethacin in 2%Tween80 Saline.
Test extract-1	200mg/kg.b.w.in 2%Tween80 Saline.
Test extract-2	300mg/kg.b.w.in2%Tween80 Saline.
Test extract-3	400mg/kg.b.w.in 2%Tween80 Saline

Hot plate method for analgesia

Healthy albino rats of the either sex (160-190 g) were divided into 5 groups of 6 animals each. They were fasted for 18 h prior to the test, with free access to water. Group I received the vehicle (0.1 ml of 2% Tween 80, orally, and served as the control group. The groups II, rats were administrated with standard drug Analgin at dose 150 mg/kg body weight, Group III, IV, V and VI were treated with, hydro-alcoholic extract (200, 300 mg & 400 mg/kg), respectively. All drugs/vehicle were administered orally (p. o.). The reaction time was noted at 30, 60, 90, 120, & 180 min. of the interval of the drug administration. Percent protection against paw licking was calculated using the following formula,

$$\% \text{ inhibition} = (Wt\text{-}Wc/20\text{-}Wc)\ 100,$$

Wt & Wc are the mean value of the test & control groups, respectively. [16] The data were analyzed by student; s t-test& the level of the significance was set at $p<.001$.

Table 2: Schedule for screening of hydro-alcoholic extracts for analgesic activity

Groups	Dose (p.o)
Control	0.1ml/kg b.w. 2%Tween80 Saline.
Standard	150 mg//kg. b.w. of Analgin in 2% Tween80 Saline.
Test extract-1	200 mg/kg.b.w.in 2%Tween80 Saline.
Test extract-2	300mg/kg.b.w.in 2%Tween80 Saline.
Test extract-3	400 mg/kg. b.w. in 2% Tween80 Saline.

Statistical analysis

The data were expressed as mean ± SD. Where applicable the difference in response to test drugs was determined by student's t-test. $P<0.05$ was considered significant.

RESULTS AND DISCUSSION

The anti-inflammatory and analgesic effects of standard drug and hydro-alcoholic extracts of *D. bipinnata* roots are shown in Tables 3 and 4 respectively. In the carrageenan-induced rat paw edema test (Table 3) for acute inflammation, the extract of *D. bipinnata* in doses of 200 mg, 300 mg and 400 mg/kg body weight showed 46%, 33.3% and 62.5% inhibition of edema, respectively, at the end of 3h.

The carrageenan-induced rat paw edema is a biphasic process. [17] The release of histamine or serotonin occurs in the first phase and the second phase is associated with the production of bradykinin, protease, prostaglandin, and lysosome. [18] Therefore, the inhibition of carrageenan-induced inflammation by the hydro-alcoholic extract of *D. bipinnata* root could be due to the inhibition of the enzyme cyclooxygenase and subsequent inhibition of prostaglandin synthesis.

Phytochemical analysis revealed the presence of tannins, saponins, alkaloids, sterols and flavonoids. The pharmacological activities of medicinal plants are usually due to their secondary metabolites. Some of the constituents of the extract have been documented to possess analgesic and anti-inflammatory activities. [19-20]

Volatile oils, resin, flavonoids and terpenoids isolated from

Table 3: Anti-inflammatory effects of hydro-alcoholic extract of *D. bipinnata* root on carrageenan-induced rat paw edema

Treatment	Dose (mg/kg b.w.)	0 min	60 min	120 min	180 min	% Inhibition, Paw vol.
Control+ carrageenan	0.1 ml/mg	0.2±0.00	0.38±0.037	0.94±0.098	1.38±0.049	---------
Indomethacin	0.5	0.22±0.44	0.28±0.020	0.40±0.032	0.54±0.024	64%
Test extract-1	200	0.22±0.44[b]	0.28±0.20	0.42±0.020[b]	0.76±0.040	46%
Test extract-2	300	0.20±00	0.56±0.024[a]	0.64±0.040	0.70±0.032[d]	33.3%
Test extract-3	400	0.20±00[a]	0.26±0.024	0.46±0.024[c]	0.58±0.020	62.5%

Values are significant at P< 0.05, n=5, [a]p>0.001, [d]p>0.001 Values are Significant, [b]p<0.001, [c]p<0.001 values are not Significant

Table 4: Analgesic effect of hydro-alcoholic extract of *D. bipinnata* root in hot plate method albino rats

Treatment	Dose (mg/kg b.w.)	0 min	30 min	60 min	90 min	120 min	180 min	% Analgesia at 90 min.
Control	0.1ml/mg	2.62±0.20	3.13±0.13	2.5±0.20	2.4±0.24	2.75±0.32	2.62±0.38	------
Analgin	150	3.5±0.20	9.5±0.29	14.5±0.29	10.2±0.32	4.87±0.31	4.37±0.24	53%
Test extract-1	200	2.62±0.45	5.0±0.20	5.37±0.24	6.36±0.24[b]	5.0±0.35	5.00±0.35	24%
Test extract-2	300	3.13±0.24[a]	7.7±0.32	11±0.35	9.25±0.14	4.5±0.29	4.50±0.29[d]	42%
Test extract-3	400	2.75±0.32	5.63±0.24	7.7±0.32	7.75±0.32[c]	4.5±0.29[c]	2.70±1.3	25%

Values are significant at P< 0.05, n=5, [a]p>0.001, [d]p>0.001 Values are Significant, [b]p<0.001, [c]p<0.001 values are not Significant

plant extracts are known to produce analgesic and anti-inflammatory effects. [21]

Tannins are important compounds known to be potent cyclooxygenase-1 inhibitors and with anti-phlogistic activity. [22- 23] The mechanisms of anti-inflammatory activity may be related to the antiphlogistic action of the tannins. Non-steroidal anti-inflammatory drugs (NSAID) such as indomethacin used in this study are known to inhibit cyclooxygenase enzymes I and II which are implicated in the production of inflammation- mediating agent prostaglandin E_2 (PGE_2) from arachidonic acid. [24-26] The pattern of anti-inflammatory and analgesic activities exhibited by this extract was similar to that of indomethacin which suggests that the plant's activity may be mediated by cyclooxygenase I and II inhibition. The anti-inflammatory effect of the extract and the reference drug increased with time.

The analgesic effect of the extract (300 mg/kg) was comparable to that produced by 150 mg/kg of analgin (Table 4).

The present study on hydro-alcoholic extract of *Desmostachya bipinnata* (L.) Stapf root has demonstrated that this plant has comparable analgesic and anti-inflammatory properties, and it justifies the traditional use of this plant in the treatment of various types of pains and inflammation. It is also suggested that the mechanism of action of *Desmostachya bipinnata* might be associated with the inhibition of prostaglandin synthesis, as observed for most non-steroidal drugs.

ACKNOWLEDGEMENT

Authors are thankful to Mrs. Vandana Gurnani, H.O.D., Department of Pharmacy, Saroj Institute of Technology and Management, Sultanpur Road, Lucknow, Pin-226002, U.P., India for their support.

REFERENCES

1. Moncada S, Vane JR. Mode of action of aspirin-like drugs. Adv. Inter. Med. 1979; 24: 1-22.
2. Insel PA. Analgesic, antipyretics and anti-inflammatory agents: drugs employed in treatment of rheumatic arthritis and gout. In: Goodman, AG. and Gilman, AG.(Eds), The Pharmacological Basis of Therapeutics. 9th Edn, Pergamon Press, Oxford, 1991; 638-681.
3. Rainsford, KD. Aspirin and the Salicyclates. Butterworth, London, 1984.
4. Meade EA, Smith WL, DeWitt DL. Differential inhibition of prostaglandin endoperoxide synthetase (cyclooxygenase) isoenzymes by aspirin and other non-steroidal anti-inflammatory drugs. J. Biol. Chem. 1993; 268: 6610-6614.
5. Bohlin L. Structure-activity studies of natural products with anti-inflammatory effects. 1995; 137-161.
6. Yesilada E, Ustun O, Sezik E, Takishi Y, Ono Y, Honda G. Inhibitory effects of Turkish folk remedies on inflammatory cytokines: interleukin-1α, interleukin-1ß and tumor necrosis factor-α. J. Ethnopharmacol. 1997; 58: 59-73.
7. Li RW, Myers SP, Leach DN, Lin GD, & Leach G. A cross-cultural study: anti-inflammatory activity of Australian and Chinese plants. J. Ethnopharmacol. 2003; 85: 25-32.
8. Vane JR, Bolting RM. New insights into the mode of action of anti-inflammatory drugs. Inflammation Res. 1995; 44: 1-10.
9. Perianayagam JB, Sharma SK, Pillai KK. Antiinflammatory activity of *Trichodesma indicum* root extract in experimental animals. J. Ethnopharmacol. 2006; 104: 410-414.
10. Kumara NKVMR. Identification of strategies to improve research on medicinal plants used in Sri Lanka, In WHO Symposium. University of Ruhuna, Galle, Lanka, 2001; 12-14.
11. Dharmasiri JR, Jayakody AC, Galhena G, Liyanage SSP, & Ratnasooriya WD. Antiinflammatory and analgesic activities of mature fresh leaves of *Vitex negundo*. J. Ethnopharmacol. 2003; 87: 199-206.
12. Panda BB, Gaur K, Kori ML, Tyagi LK, Nema RK, Sharma CS, Jain AK. Anti-Inflammatory and Analgesic Activity of *Jatropha gossypifolia* in Experimental Animal Models. Global Journal of Pharmacology. 2009; 3 (1): 01-05.
13. Evans WC. Pharmacognosy, 14th edition, 1997.
14. Winter CA, Risley EA, Nuss GW. Carrageenin induced oedema in hind paw of the rat as an assay for anti-inflammatory drugs. Proc. Soc. Exp. Biol., 1962; 111: 533-547.
15. Adeolu AA, Margaret OS, Anthony JA. Anti-inflammatory and analgesic activities of the aqueous extracts of *Margaritaria discoidea* (Euphorbiaceae) stem bark in experimental animal models. Rev. Biol. Trop. 2009; 57 (4): 1193-1200.
16. Vogel HG. Drug, Discovery & Evaluation Pharmacological Assays. Springer Publications, New York, 2002; 692-769.
17. Vinegar R, Schreiber W, Hugo R. Biphasic development of carrageenan edema in rats. J. Pharmacol. Exp. Ther.1969; 166: 96-103.
18. Crunkhorn P, and Meacock SC. Mediators of the inflammation induced in the rat paw by carrageenin. J. Pharmacol. 1971; 42: 392-402.
19. Okoli CO, Akah PA. Analgesic activity of leaf extract of *Culcasia scandens* P. Beauv. Ind. J. Exp. Bio. 2006; 44: 422-424.
20. Park EH, Kahng JH, Lee SH, Shin KH. Anti-inflammatory princip les from cactus. Fitoterapia. 2001; 72: 288-290.
21. Atta AH, Alkofahi A. Anti-nociceptive and anti-inflammatory effects of some Jordanian medicinal plant extracts. J Ethnopharmacol.1998; 60:117-124.
22. Wagner, H. Search for new plant constituents with potential anti-phlogistic and anti-allergic activity. Planta Medica. 1989; 55: 235-241.
23. Xu, GJ. The Chinese Materia Medica. Volume 1. Chinese Medicine and Technology. Beijing, China,1996.
24. Dhara AK, Suba V, Sen T, Pal S, Chaudhuri AKN. Preliminary studies on the anti-inflammatory and analgesic activity of the methanol fraction of the root extract of *Tragia involucrata* Linn. J. Ethnopharmacol. 2000; 72: 265-268.
25. Moody JO, Robert VA, Connolly JD, Houghton PJ. Anti-inflammatory activities of the methanol extracts and an isolated furanoditerpene constituent of *Sphenocentrum jollyanum* Pierre (Menispermaceae). J. Ethnopharmacol. 2006; 104: 87-91.
26. Wu KK. Aspirin and other cyclooxygenase inhibitors: new therapeutic insights. Semin.Vasc. Med. 2003; 3: 107-112.