AMERICAN JOURNAL OF PHARMTECH RESEARCH

Journal home page: http://www.ajptr.com/

Anti-Inflammatory Activity of Root Extract of *Leucas Aspera* and *Cassia Tora*

Sandip Shelke[*1], Chandanam Sreedhar[2], K.B. Chandrasekhar[3]

1. Department of Pharmaceutical Sciences, Jawaharlal Nehru Technological University, Anantapur, A.P., India.

2. HOD of Pharmaceutical Analysis, Karnataka College of Pharmacy, Bangalore, India.

3. Professor of chemistry, Director of OTRI, Jawaharlal Nehru Technological University, Anantapur, A.P., India.

ABSTRACT

Inflammation plays key role in various diseases such as asthma, atherosclerosis and rheumatoid arthritis. Inflammation is defined as the local response of living mammalian tissues to tissue injury due to any agent. It is a body defense reaction in order to eliminate or limit the spread of injurious agent as well as to remove the consequent necrosed cells and tissues. *Leucas aspera* is a used orally as stimulant, anthelmintic, laxative, diaphoretic, for the treatment of headache, asthma, and bronchitis. The decoction of roots, stem of *Leucas aspera* is used orally for high fevers, for influenza, and for malarial fevers. *Cassia tora* commonly is used as tonic, carminative and stimulant. According to Ayurveda the leaves and seeds are acrid, laxative, antiperiodic, anthelmintic, ophthalmic, liver tonic, cardiotonic and expectorant. This study was aimed to evaluation of anti-inflammatory activity of the roots of *Leucas aspera* and *Cassia tora*.

Keywords: *Leucas aspera*, *Cassia tora*, anti-inflammatory

*Corresponding Author Email: shelkesandip83@rediffmail.com

Received 25 December 2014, Accepted 03 January 2015

Please cite this article as: Shelke S *et al*., Anti-Inflammatory Activity of Root Extract of *Leucas Aspera* and *Cassia Tora*. American Journal of PharmTech Research 2015.

INTRODUCTION

Inflammation is defined as the local response of living mammalian tissues to tissue injury due to any agent. It is a body defense reaction in order to eliminate or limit the spread of injurious agent as well as to remove the consequent necrosed cells and tissues. There are two types of Inflammation, acute inflammation and chronic inflammation. Acute inflammation is of short duration and represents the early body reaction. Chronic inflammation is defined as prolonged process in which tissue destruction and inflammation occur at the same time. Inflammation plays key role in various diseases such as asthma, atherosclerosis and rheumatoid arthritis[1, 2]. India has one of the richest plants medical traditions in the world. Medicinal plants play an important role in the development of potent therapeutic agents. Plant based drugs provide outstanding contribution to modern therapeutics[3]. *Leucas Aspera* commonly known as Thumbai (Family- *Lamiaceae) is distributed throughout India*[4, 5].

Leucas aspera is a used orally as stimulant, anthelmintic, laxative, and diaphoretic. It is also used orally for the treatment of headache, asthma, and bronchitis. The decoction of roots, stem of *Leucas aspera* is used orally for high fevers, for influenza, and for malarial fevers[6-14]. *Cassia tora* commonly known as wild senna (Family- *Caesalpiniaceae) is wild crop and grown in most parts of India as a weed*[15]. Traditional it is used as tonic, carminative and stimulant. Its leaves, seeds, and roots are used medicinally, primarily in Asia. According to Ayurveda the leaves and seeds are acrid, laxative, antiperiodic, anthelmintic, ophthalmic, liver tonic, cardiotonic and expectorant. The leaves and seeds are useful in leprosy, ringworm, flatulence, colic, dyspepsia, constipation, cough, bronchitis, cardiac disorders[16]. However the roots of Leucas aspera and Cassia tora have not been evaluated for anti-inflammatory activity. This study was aimed to evaluation of anti-inflammatory activity of the roots of *Leucas aspera* and *Cassia tora*.

MATERIALS AND METHOD

Collection of plant materials

The roots of *Leucas aspera* and *Cassia tora* were collected from Ananthagiri forest region, Vishakapatnam District, Andhra Pradesh, India. The plant species were authenticated by Dr. K. Madhava Chetty, Department of Botany, Shri Venkateshwara University, Tirupati, India.

Extraction of plant materials[17-18, 19]

The fresh roots were cleaned and shade dried at room temperature and was chopped into small pieces. Dried plant were powdered and packed in air tight container. The coarse powders of both

plant materials were packed in soxhlet column for 6 hr successively with methanol and petroleum ether. Thereafter, the extracts were concentrated using rotary flash evaporator (50^{o}C).

Experimental Animals

Albino wistar rats weighing 150-220g were maintained under controlled condition of temperature at 27^{o} ± 2^{o} C and 12-h light-dark cycles and relative humidity of 50 ± 15%). All the studies conducted were approved by the Institutional Animal Ethical Committee (IAEC) of according to prescribed guidelines of Committee for the Purpose of Control and Supervision of Experiments on Animals, Govt. of India. (Regd no. 769/2011/CPCSEA) They were housed in polypropylene cages and had a free access to standard pellets (Supplied by Ratan Brothers) and water *ad libitum*.

Acute Toxicity Studies (LD_{50})[20]

Acute toxicity study was performed according to the OECD guideline no. 425.

***IN VIVO* ANTI-INFLAMMATORY ACTIVITY**

Acute Anti-inflammatory Activity[21]

Albino wistar rats with body weight between 150-220g were used. The animals were starved overnight and fasted for 18 hours prior to the experiment. The animals were dived into eight groups of six animals each. Acute inflammation was induce by injecting formalin (0.1 ml of 1% suspension in 0.9 % saline) in sub-plantar region and paw volume was measured 0, 1, 2, 3, 4 and 5 hour, with the help of Plethysmometer.

All the treatment compounds compound were administered 30 minutes, prior to formalin. Acute inflammation was induced in right hind paw. The initial reading was taken at 0 hour, i.e., immediately after injecting formalin and the procedure was repeated at 1, 2, 3, 4 and 5 hour after formalin injection. The difference between 0 hour reading and one of the subsequent readings provides the actual oedema volume at the time. The mean paw volume at different times was calculated and compared with the control and the results are tabulated by percentage of inhibition.

Chronic Anti-inflammatory Activity[22]

Albino wistar rats weighing 170-230 mg/kg were divided into eight groups of six in each group. All these animals were fasted for 18 hours before the beginning of the experiment and water was given ad libitum. In animals of all the groups chronic inflammation was produced by sub plantar injection of 20µl of freshly prepared 2% suspension of formalin in normal saline in right hind paw of rat was used as the oedematogenic agent. Animals were treated with drugs for 6 consecutive days. The paw volume was measured using a plethysmometer before and 6 days after formalin challenge in each group. The increase in paw volume and percent of inhibition was calculated.

Group-I: Distilled water will be supplied and served as control.

Group-II: Animals received a dose of 100 mg/kg of Diclofenac sodium i.p. and served as standard

Group-III: Animals received a dose of 200 mg/kg of MELA and MECT p.o.

Group-IV: Animals received a dose of 400 mg/kg of MELA and MECT p.o.

Group-V: Animals received a dose of 600 mg/kg of MELA and MECT p.o.

Group-VI: Animals received a dose of 200 mg/kg of, PELA and PECT p.o.

Group-VII: Animals received a dose of 400 mg/kg of PELA and PECT p.o.

Group-VIII: Animals received a dose of 600 mg/kg of PELA and PECT p.o.

Statistical Analysis

The values are expressed as Mean ± SEM. The data was analysed by using one way ANOVA followed by Tukey multiple comparison tests using Graph pad prism software. Statistical significance was set at $P \leq 0.05$.

RESULT AND DISCUSSION

Acute Anti-inflammatory Activity

All the test compounds were tested with the diclofenac sodium as a standard drug in the dose of 10 mg/kg for the anti-inflammatory activity. Presently diclofenac showed significant 86.23 % inhibition of inflammation at 5^{th} hour (0.19 ± 0.02) when compared with control (1.34 ± 0.03) respectively. The test compounds showed maximum percentage of inhibition of oedema at 5^{th} hour significantly in respective dose level i.e., at 200, 400 and 600 mg/kg. The test compounds MELA and PELA showed 68.84%, 77.53 %, 84.05% and 59.42%, 71.73%, 78.26%. The values are tabulated in the Table 1.

Results with *Leucas aspera* and *Cassia tora* of MELA, PELA, MECT and PECT (600mg/kg, p.o.) are showed quite compatible with those of the standard drug diclofenac sodium. Therefore, the drug appears to be effective against formalin-induced arthritis.

Chronic Anti-inflammatory Activity

Formalin induced paw oedema is one of the most suitable test procedure to screen chronic anti-inflammatory agents. The results obtained as mean increase in paw volume (ml) and % inhibition are represented in Table 2. The mean response of standard was 85.02% inhibition of increase in paw thickness after 6 days respectively. In this model at 200, 400 and 600 mg/kg dose level of MELA and PELA extracts showed 36.43%, 59.91%, 72.46%, and 29.55%, 40.89%, 66.39% inhibition of increase in paw thickness after 6 days. All the results were compared with solvent control and diclofenac sodium reference drug control. Formalin induced paw oedema is one of the most suitable test procedure to screen chronic anti-inflammatory agents. The mean response of

standard 85.02% was inhibition of increase in paw thickness after 6 days respectively. In this model at 200, 400 and 600 mg/kg dose level of MELA, PELA, MECT and PECT extracts showed significantly inhibition of increase in paw thickness after 6 days.

Table 1: Effect of MELA and PELA on Formalin-induced paw Oedema in Rats

Groups	Treatment	Paw Oedema Volume (hr)						
		0 hr	1hr	2hr	3hr	4 hr	5 hr	% Inhibition
Group-I	Saline	0.17 ±0.02	0.81 ± 0.05	1.13 ± 0.04	1.23 ± 0.07	1.29 ± 0.03	1.34 ± 0.03	-
Group-II	Diclofenac sodium (10mg/kg i.p.)	0.16 ± 0.03	0.39 ± 0.04***	0.61 ± 0.04***	0.34 ± 0.02***	0.27 ± 0.05***	0.19 ± 0.02***	86.23 %
Group-III	MELA (200mg/kg p.o.)	0.18 ± 0.02	0.68 ± 0.05*	0.99 ± 0.03**	1.13 ± 0.05***	0.83 ± 0.06***	0.43 ± 0.03***	68.84%
Group-IV	MELA (400mg/kg p.o.)	0.17 ± 0.03	0.61 ± 0.04**	0.86 ± 0.06***	0.58 ± 0.03***	0.42 ± 0.04***	0.31 ± 0.05 ***	77.53 %
Group-V	MELA (600mg/kg p.o.)	0.16 ± 0.02	0.52 ± 0.03***	0.74 ± 0.05***	0.42 ± 0.03***	0.34 ± 0.04***	0.22 ±0.03***	84.05%
Group-VI	PELA (200mg/kg p.o.)	0.17 ± 0.02	0.74 ± 0.03^{ns}	1.02 ± 0.05*	1.150 ± 0.04**	0.98 ± 0.04***	0.56 ± 0.016***	59.42%
Group-VII	PELA (400mg/kg p.o.)	0.15 ± 0.01	0.63 ± 0.04**	0.89 ± 0.05***	0.62 ± 0.03***	0.50 ± 0.02***	0.39 ±0.04***	71.73%
Group-VIII	PELA (600mg/kg p.o.)	0.17 ± 0.02	0.58 ± 0.03***	0.84 ± 0.05***	0.51 ± 0.03***	0.41 ± 0.02***	0.30 ± 0.015***	78.26%

Values are Mean ± SEM (n=6) one way ANOVA followed by Tukey-Karmer's test. Where, *** P<0.001, ** P<0.01, * P<0.05 and ns represents Not significant. MELA- Methanolic extract of *Leucas aspera*, PELA: Petroleum ether extract of *Leucas aspera*

Table 2: Effect of MELA and PELA on Formalin-induced paw Oedema in Rats (Chronic Anti-inflammatory Activity)

Groups	Treatment	Initial Paw Volume	Paw Volume After 6 Days	Increase in Paw Volume	% of Inhibition
Group-I	Saline	1.27 ± 0.05	3.74 ± 0.12	2.47 ± 0.06	-
Group-II	Diclofenac sodium (100 mg/kg i.p.)	1.24 ± 0.09	1.61 ± 0.09	0.37 ± 0.07	85.02%
Group-III	MELA (200mg/kg p.o.)	1.29 ± 0.06	2.86 ± 0.29	1.57 ± 0.08	36.43%
Group-IV	MELA (400mg/kg p.o.)	1.22 ± 0.07	2.21 ± 0.22	0.99 ± 0.16	59.91%
Group-V	MELA (600mg/kg p.o.)	1.26 ± 0.03	1.94 ± 0.16	0.68 ± 0.12	72.46%
Group-VI	PELA (200mg/kg p.o.)	1.32 ± 0.05	3.06 ± 0.27	1.74 ± 0.14	29.55%
Group-VII	PELA (400mg/kg p.o.)	1.30 ± 0.04	2.76 ± 0.20	1.46 ± 0.14	40.89%
Group-VIII	PELA (600mg/kg p.o.)	1.29 ± 0.03	2.12 ± 0.18	0.83 ± 0.12	66.39%

Results are expressed on mean + SEM from four observations Paw Volume was measured after 6 days. MELA- Methanolic extract of *Leucas aspera*, PELA: Petroleum ether extract of *Leucas aspera*.

CONCLUSION

Anti-inflammatory activity evaluated was acute and chronic models of inflammation. The acute inflammatory property the formalin induced paw oedema in rats was employed and for acute anti-inflammatory activity, formalin-induced paw oedema in rats was used. The result of study showed that methanolic and petroleum ether extracts of Leucas aspera and Cassia tora roots has showed significantly acute anti-inflammatory activity. It also showed significantly chronic anti-inflammatory activity.

REFERENCES

1. Mohan H. Text book of pathology, 3 ed., New Delhi: Jaypee publisher; 1998: 92-130.
2. Naik SR. Inflammatory process and screening methods for anti-inflammatory agents- a review. Jr. of postgraduate Medicine. 1976; 22(1): 05-21.
3. Cooper EL. Drug discovery and natural products. Evidence Based Complementary Alternative Medicine. 2004; (1): 215–7.
4. R. Srinivasan, B. Ravali, P. Suvarchala, A. Honey, A. Tejaswini, P. Neeraja. Leucas Aspera- Medicinal Plant: A Review. International Journal of Pharma and Bio Sciences, Apr-June 2011; 2(1): 153-159.
5. M.S.Prajapati, J.B. Patel, K.Modi; M.B.Shah. Leucas Aspera: A Review. Pharmacogn Rev., 2010; 4(7): 85-87.
6. Balunas M.J., Kinghorn A.D. Drug discovery from medicinal plants. Life Sci., 2005; 78: 431–41.
7. Hedge I.C., Nasir Y.J. Flora of Pakistan, Karachi, University of Karachi, Department of Botany; 1990: 192.
8. Pushpangadan P., Atal C.K. Ethno-medico-botanical investigations in Kerala I. Some primitive tribals of western ghats and their herbal medicine. J Ethnopharmacol,1984; 11: 59–77.
9. Girach R.D., Aminuddin., Siddioui P.A., Khan S.A. Traditional plant remedies among the Kondh of district Dhenkanal (Orissa)Int. J Pharm., 1994; 32: 274–83.
10. Bhandary M.J., Chandrasekhar K.R., Kaveriappa K.M. Medical ethnobotany of the Siddis of Uttara Kannada district, Karnataka, India. J. Ethnopharmacol, 1995; 47: 149–58.
11. Selvanayahgam Z.E., Gnanevendhan S.G., Balakrishna K., Rao R.B. Antisnake venom botanicals from ethnomedicine, J Herbs Spices Med Plants, 1994; 2: 45–100.
12. Khaleque A., Huq M.E., Huq M.S., Mansoor M.H. Chemical investigations on *leucas aspera*. I. Isolation of compound a, beta-sitosterol and alpha-sitosterol from the aerial parts. Sci Res. (DACCA, PAK), 1970; 7: 125–7.

13. Kapur S.K., Sarin Y.K. Medico-botanic survey of medicinal and aromatic plants of katra valley (J.& K.STATE) India. Indian Drugs, 1984; 2: 4–10.
14. John D.; One hundred useful raw drugs of the kani tribes of Trivandrum Forest Division, Kerala, India. Int. J. Crude Drug Res., 1984; 22: 17–39.
15. International Legume Database & Information Service. Botanic Garden and Botanical Museum Berlin. Dahlem, 2006.
16. Smita Jain., Patil U.K. Phytochemical and pharmacological profile of Cassia tora Linn.- An Overview, Indian J. of Natural Products and Resources, 2010; 1(4): 430-437.
17. Kirtikar K.R.; Basu B.D. Indian Medicinal Plants. 3rd Edition, Bombay: Popular Prakashan; 1998: 87.
18. Anonymous. Quality standards of Indian medicinal plants. New Delhi : Periodical Experts; 2008: 265-274.
19. Dr. Pulok Mukherjee, Quality Control of Herbal Drugs. First edition, New Delhi: Horizons Pharmaceutical Publisher; 2002: 112-515.
20. OECD Guidelines for the Testing of Chemical. Acute Oral Toxicity – Up and Down Procedure (UDP) [Internet]. 2008 Available from: http://iccvam.niehs.nih.gov/SuppDocs/FedDocs/OECD/OEC Dtg425.pdf.
21. Yogesh Baravalia., Yogeshkumar Vaghasiya., Sumitra Chanda. Brine Shrimp Cytotoxicity, Anti-inflammatory and Analgesic Properties of *Woodfordia fruticosa* Kurz Flowers, Iranian J Pharm Res., 2012; 11(3): 851-861.
22. Bikash Kumar Nanda., Jyotirmoyee Jena.; et al. Anti inflammatory activity of whole parts of *Sphaeranthus indicus* Linn, Der. Pharmacia. Lettre, 2010; 2(1): 181-188.