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Research Article

ANTI-INFLAMMATORY AND ANTI-ARTHRITIC ACTIVITIES OF AERIAL PARTS OF NARAVELIA ZEYLENICA (L) DC.

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ABSTRACT

Naravalia zeylanica (L) DC. (Ranunculaceae) is a climbing vine distributed in hilly areas used by the tribal to cure various ailments. The leaf and stem of this plant used in vitiated vata, pitta, inflammation, skin diseases, rheumatoid arthritis, arthritis, headache, colic, wounds and ulcers The plant material was collected from kolli hills, Tamilnadu, and authenticated. To create the scientific evidence for the folk claiming, the present work focused on evaluation of Anti-inflammatory and Anti-arthritic activity of aerial parts of *Naravalia zeylanica*. The plant material was extracted by successive solvent extraction by cold maceration method. From the preliminary phytochemical studies observed that the chloroform and ethanolic extracts contains phytoconstituents such as flavanoids, triterpenoids, alkaloids, polyphenols and saponins. From the acute oral toxicity studies noted that the chloroform and ethanolic extract did not showed any mortality and toxic reaction in the dose of 2000mg/kg. Anti inflammatory studies was carried out by Carrageenan induced edema method in rats. The percentage increase in paw edema reduced in both chloroform (20.70 ± 3.64) and ethanol extract (29.38 ± 2.19) treated animals when compared with standard Indomethacin (22.01 ± 4.08) and control (38.20 ± 1.63) animals. The results were statistically significant ($p<0.01$). The anti-arthritic activity was evaluated in Freud's adjuvant induced arthritis model in albino rats. The percentage increase in paw volume 7days and 21 days after the drug administration were noted. It was noted that a moderate reduction in paw volume in the right and left paw of rats treated with chloroform extract (69.86 ± 3.39 &50.88 ± 2.51), ethanolic extract (66.99 ± 3.85 & 49.040 ± 2.87) and Prednisolone 10 mg/kg p.o. (63.82 ± 1.86 & 34.90 ± 3.00) treated animals when compared with control (147.94 ± 5.84 & 111.97 ± 8.45) group animals. Both the extracts (200mg/kg) were statistically significant ($p<0.01$). The activity may be due to the presence of flavanoids, triterpenoids and phenolic compounds in the both extracts

Keywords: Naravalia zeylanica, Ant-arthritic activity, Anti- inflammatory activity,

INRODUCTION

Naravalia zeylanica DC (Ranunculaceae) is a woody climber with tuberous roots opposite, ovate, cordate leaflets, small flowers arranged in panicles and red coloured achenes along with long feathery styles, occurring in the hot to warm regions in India. From the literature survey and ethnomedical studies, observed that the aerial parts of Naravelia zeylanica traditionally used in vitiated vata, pitta, inflammation, skin diseases, rheumatoid arthritis, arthritis, headache, colic, wounds and ulcers[1]. Leaf paste is consumed to treat Chest pain. The vines when crushed give a pungent odour which is inhaled to cure cold, all type of headaches including migraine[2]. The leaf extract were already reported for anti-ulcer and anthelmintic activity[3-4]. Based upon the ethnomedical importance, present work focused on anti-inflammatory and anti-arthritic activity of aerial parts of Naravalia zeylanica to reveal its folk claiming.

MATERIALS AND METHODS

Collection of Plant Material

Naravalia zeylanica DC was collected from Kolli hills of Namakkal district, Tamilnadu, India. The plant was identified and authenticated by National Institute of Herbal Science, Chennai (RHT: 42669 & SHC: CM 1695)[5]. A voucher specimen has been maintained in our lab for future reference. The aerial part of the plants were collected in the month of August and shade dried.

Extraction of Plant Material

Coarse powder of the aerial parts of the plant material was extracted by cold maceration method using successive solvents such as petroleum ether, chloroform and ethanol in increasing polarity for 48 hours each. The extracts were concentrated by distillating the solvent and dried under reduced pressure.

Preliminary Phytochemical Tests

Petroleum ether, chloroform, ethanol and aqueous extracts were subjected to phytochemical chemical tests to identify the phytoconstituents using standard qualitative reagents[6-7]. From the phytochemical test, presence of triterpenoids, alkaloids, flavonoids and saponins were observed in chloroform and ethanolic extracts.

Acute Toxicity Studies

The animal studies were carried out after getting approval from the institutional animal ethical committee. Acute oral toxicity was performed as per organization for economic co-operation for development (OECD) guideline 423 methods. Healthy young adult Swiss albino mice, weighing about 25-30gm of either sex were divided into three groups (chloroform extract, ethanolic extract & control) of each nine animals. The chloroform and ethanolic extracts was administered in a single dose by gavage using specially designed mice oral needle. Animals were fasted 3 h prior to dosing (food was withheld for 3 h but not water). Following the period of fasting animals was administered orally at a single dose of 2000 mg/kg. After substance administration, food was withheld 2 h in mice. Animals were observed individually after atleast once during the first 30 minutes, periodically during the first 24 hrs, with special attention given during the first 4 hrs, and daily thereafter, for a total of 14 days. The direct observation parameters such as Tremors, convulsions, salivation, diarrhea, lethargy, sleep and coma were observed. There was no death other complications reported in the 14 days.From the lethal dose (LD_{50})2000mg/kg, the effective dose (ED_{50}) was calculated as 200mg/kg.

Anti inflammatory studies

Anti inflammatory studies was carried out by Carrageenan induced oedema method in rats (winter et,al 1962). Healthy young adult winstar albino rats, weighing about 150-170gms of either sex were divided into four groups each of five animals. The four groups were treated with ethanol and chloroform extracts (200mg/kg), indomethacin (10mg/kg) and control vehicle orally. After 30 minutes, the rats were challenged with subcutaneous injection of 0.1ml of 1%w/v solution of carrageenan into the sub planar region of left paw. The paw was immersed in mercury up to the mark. The paw volume was measured at 0,1,2,3 and 4 hr after carrageenan injection using a volume transducer attached with strain gauge coupler of student physiograph (model no.PG-02.INCO, Ambala, India).The difference between initial and subsequent reading gave the actual edema volume(Table no 1). The animals that received *Naravelia zeylanica* extracts and indomethacin

(10mg/kg) was compared with vehicle control groups[8].

Anti arthritic activity screening

Freund's adjuvant induced arthritis model was used to assess the anti-arthritic activity in albino rats. Healthy young adult Swiss albino rats, either sex weighing 200gm of animals were taken and divided into four groups of six animals each. Group I served as control, which received 5ml/kg saline, Group II & III received 200mg/kg body weight of Chloroform & ethanol extract and Group IV received prednisolone 10 mg/kg po., respectively. Arthritis was induced by injecting 0.05 ml of suspension of killed Mycobacterium tuberculosis bacteria (0.5% w/w) homogenized in liquid paraffin into the left hind paw. Drug treatment was started from the day of adjuvant injection (0 day-30 min before injection) and continued till 21st day. Paw volume was measured on 7th and 21st day with the help of a volume transducer attached with strain gage coupler of student physiograph (Model no.PG-02.INCO, Ambala, India). The percentage increase in paw edema with respect to initial paw volume, were calculated on respective days in percentage (Table no 2&3). The reduction in percentage of increased paw volume showed the higher protection activity[9].

Statistical Analysis

The experimental results were expressed as mean ± S.E.M. Data were assessed by the method of analysis of Oneway ANOVA followed by Dunnett's test. P value, $p<0.01$ & $p<0.05$was considered as statistically significant.

RESULT AND DISCUSSION

The coarse powder of aerial part of *Naravalia zeylanica* DC extracted with various solvents successively by cold maceration method and the percentage yield were such as petroleum extract 6.2%w/w, chloroform extract 10.6%w/w, ethanol extract 12.35%w/w and aqueous extract 19.28%w/w. The phytochemical tests revealed the presence of alkaloids (chloroform & alcohol extracts), triterpenoids (chloroform and ethanol extracts), saponins (alcohol & aqueous extracts), phenolic compounds (alcohol & aqueous extracts), and flavonoids (pet.ether, chloroform & alcohol extracts).

In Acute oral toxicity screening after administration of single oral dose of 2000 mg/kg, there was no death and other complications reported with in the 14 days. The anti-inflammatory studies revealed that the percentage increase in paw edema, reduced in both chloroform (20.70 ± 3.64%) and ethanol extract (29.38 ± 2.19%) treated animals when compared with standard Indomethacin (22.01 ± 4.08%) and control (38.20 ± 1.63%) animals. The results were statistically significant ($p<0.01$).

From the Freud's adjuvant induced arthritis models, the percentage increase in paw volume 7days and 21 days after the drug administration were noted. It was revealed that the reduction in paw volume in the right and left paw of rats treated with chloroform extract (69.86 ± 3.39% &50.88 ± 2.51%), ethanolic extract (66.99 ± 3.85% & 49.040 ± 2.87%) and Prednisolone 10 mg/kg p.o. (63.82 ± 1.86% & 34.90 ± 3.00%) were moderately reduced when compared with control (147.94 ± 5.84% & 111.97 ± 8.45%) group animals. Both the extracts were statistically significant ($p<0.01$).

SUMMARY

The chloroform extract of aerial part of *Naravalia zeylanica* DC possessed significant anti-inflammatory activity that may be due to its ability to prevent the production of some pro-inflammatory mediators. In the anti arthritic activity screening both chloroform and ethanolic extract showed significant activity when compared with the standard and control (Figure 1&2). The cordial signs of the chronic inflammatory reactions like redness, swelling, arthalgia and immobility of affected joints were significantly less in the drug treated animal than those of the control. The activity may due to the presence of triterpenoids and poly phenolic constituents. Polyphenols may inhibit NF-kB, a transcription factor which stimulates the enzyme which produces the inflammatory agent nitric oxide. Phenolics are antioxidants and free-radical scavengers, which may aid in suppressing reactive oxygen species (ROS) that stimulate inflammatory responses.

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Table 1: Effect of extract on carrageenan induced paw edema in rats after 4hrs

S. No.	Treatment	Dose	Percentage increase in paw volume (%)
1	Control		38.20 ± 1.63
2	Indomethacin	10mg/kg	22.01 ± 4.08**
3	Chloroform extract	200 mg/kg p.o.	20.70 ± 3.64**
4	Ethanolic extract	200 mg/kg p.o.	29.38 ± 2.19^{ns}

Values are expressed as mean ± SEM. **p<0.01, *p<0.05, ^{ns} p>0.05.
(Oneway ANOVA followed by Dunnett's test)

Table 2: Anti-arthritic activity on Freund's adjuvant induced arthritis in rats on 7th day

S. No.	Treatment	Dose	Percentage increase in paw volume on 7th day (%)	
			Right paw	Left paw
1	Control (saline)	5 ml/kg	147.94 ± 5.84	111.97 ± 8.45
2	Chloroform extract	200 mg/kg p.o.	69.86 ± 3.39 **	50.88 ± 2.51 **
3	Ethanolic extract	200 mg/kg p.o.	66.99 ± 3.85**	49.040 ± 2.87**
4	Prednisolone	10 mg/kg p.o.	63.82 ± 1.86**	34.90 ± 3.00**

Values are expressed as mean ± SEM. **p<0.01, *p<0.05, ^{ns} p>0.05.
(Oneway ANOVA followed by Dunnett's test)

Table 3: Anti-arthritic activity on Freund's adjuvant induced arthritis in rats on 21th day

S. No.	Treatment	Dose	Percentage increase in paw volume on 21th day (%)	
			Right paw	Left paw
1	Control (saline)	5 ml/kg	87.41 ± 3.91	53.97 ± 2.50
2	Chloroform extract	200 mg/kg p.o.	42.77 ± 2.08**	33.63 ± 3.02**
3	Ethanolic extract	200 mg/kg p.o.	41.13 ± 2.15**	32.70 ± 3.22**
4	Prednisolone	10 mg/kg p.o.	33.17 ± 1.37**	23.76 ± 2.50**

Values are expressed as mean ± SEM. **p<0.01, *p<0.05, ^{ns} p>0.05.
(Oneway ANOVA followed by Dunnett's test)

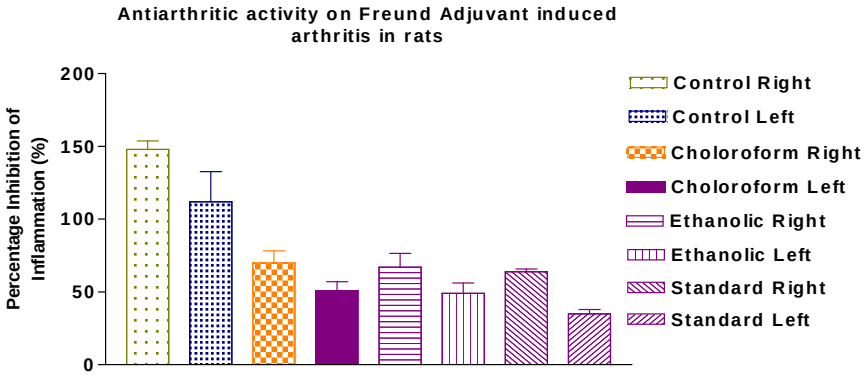

Fig. 1: 7th day

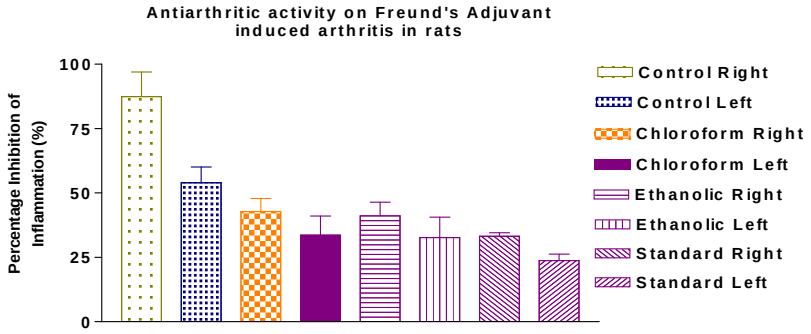

Fig. 2: 21st day

REFERENCES

1. Varriar PS. Indian Medicinal Plants. Orient Longman Ltd., Hyerabad, India. 1995;4:100-103.
2. Bhandary MJ, Chandrashekar KR and Kaveriappa. Medical Ethnobotany of the Siddis of Uttara Kannada District, Karnataka, India. J Ethnopharmacol. 1995;47:149-158.
3. Ashoka Shenoy M, Sridevi, Shastry CS and Gopkumar P. Anthelmintic activity of Naravelia zeylanica leaves extract. Pharmacologyonline. 2009;1: 1239-1245.
4. Ashoka Shenoy M, Shastry CS, Sridevi and Gopkumar P. Anti ulcer activity of Naravelia zeylanica leaves extract. Journal of Pharmacy Research. 2009;2(7):1218-1220.
5. Gamble JS. Flora of the Presidency of Madras. Bishen Singh & Mahendra Pal Singh, Dehradun. 1915;1:1-3.
6. Kokate CK. Practical Pharmacognosy. Vallabh Prakashan, Delhi. 2005;4th Edn:20-30.
7. Khandelwal KR. Practical Pharmacognosy Technique and Experiments. Nirali Prakashan, Pune. 2006;16th Edn: 15-29, 149-56.
8. Agarwal RB and Rangari VD. Anti-inflammatory and Anti-arthritic activities of Lupeol and 19a-H Lupeol isolated from Strobilanthus callosus and Strobilanthus ixiocephala roots. Indian Journal of Pharmacology. 2003;35:384-387.
9. Kamalutheen M, Gopalakrishnan S and Syed Ismail T. Anti-inflammatory and Anti-arthritic Activities of Merremia tridentata (L.) Hall. E-Journal of Chemistry. 2009;6(4):943-948.