J.D.R.A.S. Vol. XXXIII-XXXIV, No.1-4, (2012-13) pp.31-42

ANALGESIC, ANTIPYRETIC AND ANTI-INFLAMMATORY EFFECTS OF *SISYMBRIUM IRIO.* LINN. SEEDS

Y. R. Sanjaya Kumar[1], N. Jaya[2] and C.K.Sasikala[3]

Abstract

Sisymbrium irio Linn. an annual herb belongs to Cruciferae family and is found in parts of northern India. As per classic literature, its seeds were said to relieve fever and inflammation. In the present study decoction and aqueous extracts of Sisymbrium irio seeds were screened for analgesic, antipyretic and anti-inflammatory properties in experimental animals. The test drug showed significant analgesic activity by hot plate method and radiant heat method. It also showed moderate decrease in number of stretching episodes against acetic acid induced writhing in mice. The test drug showed slight anti-inflammatory activity as evidenced by mild suppression of carrageenan hind paw oedema and granulation tissue formation in cotton pellet granuloma method. A mild antipyretic activity has also been observed at 3rd and 4th hour post test drug administration. Decoction of Sisymbrium irio seeds was found to be more effective than aqueous extract.

Keywords: *Sisymbrium irio, Analgesic, Anti-inflammatory, Antipyretic Experimental animals.*

Introduction

Sisymbrium irio. Linn. (Cruciferae) is an annual herb, 90 cm tall found in Kashmir, Punjab and parts of Haryana and from Rajasthan to Uttar Pradesh mostly on moist soils. Its seeds are ovoid, reddish /yellow brown, 1.3 mm, mucilaginous when wet, with hot and sharp taste. It possesses rubifacient, expectorant, restorative and febrifugal properties and is mainly used in asthma and as poultice [1]. Its seeds are especially helpful in fever [2].

*Corresponding Author

1. Assistant Director (Pharmacology), National Research Institute for Panchakarma, Cheruthuruthy (P.O.), Thrissur (Dist.), Kerala, 679 531.

2. Deputy Director in charge 3.Laboratory Technician, National Research Institute for Panchakarma, Cheruthuruthy (P.O.), Thrissur (Dist.) Kerala 679 531

Materials and methods

The sample of *Sisymbrium irio* seeds procured from CRI (Ay.), Patiala was coarsely powdered and used in the preparation of extracts. Decoction and Aqueous extracts were prepared as per classics. Wistar albino rats (120- 150 grams) and Swiss albino mice (20-30 grams) bred in the Institute animal house were used in the experiment. The experiment consisted of four groups viz., control, test groups (2) and standard control, consisting of 6 animals each of either sex. The test extracts (Decoction and aqueous extracts) were screened at dose of 1250 mg (lower dose) and 2500 mg (Higher dose) per kg body weight.

Acute toxicity studies

Decoction and aqueous extracts of the test drug were administered in graded doses of 1250 mg to 5000 mg per kg body weight and were observed for next seventy two hours for toxic effect and death [3].

Analgesic activity

This was evaluated with 3 experimental models i.e., hot plate method and acetic acid induced writhing method (mice) and radiant heat method (rat).

(a). Hot plate method

This was carried out as per the method of Eddy and Leimbach [4]. In this method heat is used as a source of pain. Animals were individually placed on a hot plate maintained at constant temperature (55^0 C) and the time taken by the animals for showing reactions such as paw licking and jump response is noted. The reaction time in test group was compared with that of control and standard groups.

(b). Acetic acid writhing

The painful reaction characterized as writhing response in animals is produced by intra peritoneal injection of chemicals is taken as criteria to assess analgesic activity of test drugs [5]. 3% acetic acid was used to induce writhing in mice. Ibuprofen (100 mg per kg body weight) administered group served as reference standard.

(c). Radiant Heat Method

This test makes use of tail flicking (tail withdrawal from the heat source) response to the thermal stimuli generated by radiant heat source using analgesiometer [6]. The basal reaction time prior to drug administration was compared with that of post drug administration in rats between control, test groups and standard groups (Ibuprofen).

Anti inflammatory activity

Decoction and aqueous extract of the test drug were evaluated for anti-inflammatory effect by carrageenan hind paw oedema method [7] and Cotton pellet granuloma method [8] in rats.

(a). Effect on carrageenan hind paw oedema

Hind paw oedema was induced by injecting 1% carrageenan in normal saline beneath plantar aponeurosism of right hind paw of rats. Control, test drug and standard drugs (Ibuprofen 100 mg per kg. body weight) were administered 45 minutes prior to Carrageenan injection were injected. The increase in paw volume was noted 3 hours post carrageenan administration by fluid displacement method using plethysmometer [9] and compared between the groups.

(b). Cotton pellet granuloma method

Effect of test drug on cotton pellet granuloma formation was studied [8]. Cotton pellets weighing 10 mg were sterilized and implanted subcutaneously in both axillae and groin under ether anesthesia. Test drug, standard drug and distilled water were administered for six consecutive days after the implantation of cotton pellets. Rats were sacrificed on 7th day and granulation tissue around cotton pellet were excised and dried in hot air oven for 7 hours at 60-70^{0}C. Weight of each pellet was recorded. Effect of test drug and control on body weight and weights of liver, spleen and adrenal were recorded. Ibuprofen treated group served as reference standard.

Antipyretic activity

20% yeast in normal saline at the dose of 1 ml per 100 gram was injected subcutaneously to induce fever in rats [3]. Test drug, reference drug were administered at peak temperatures and rectal temperature was recorded at hourly intervals for 4 hours.

Results

Acute Toxicity.

Neither decoction nor aqueous extract produced apparent toxicity or death up to the dose of 5000 mg per kg body weight in both rats and mice.

Analgesic activity

(a). Hot plate method

Both decoction and aqueous extract of the test drug increased the mean licking response and jump response time in mice significantly at different time intervals (Table I and II).

(b). Acetic acid induce writhing

The test drug showed mild increase in stretching episodes. Both the extracts failed to completely protect the mice against acetic acid induced writhing, but a mild decrease ($P< 0.1$) in stretching episodes was noted (Tables III and IV).

(c). Radiant heat method

Statistically significant increase in the threshold tail flick response was observed with respect to both extracts of the test drug in rats at different time intervals (Tables V and VI).

Anti- inflammatory activity

(a). Carrageenan paw oedema

Both decoction and aqueous extract showed slight ($P<0.1$) decrease in the paw volume against carrageenan induced hind paw oedema (Table VII and VIII).

(b). Cotton pellet granuloma

The test drugs (decoction and aqueous extract) showed mild ($P<0.1$) anti-inflammatory effect and slight inhibition of granuloma formation. The inhibitory effect was mild when compared to that of Ibuprofen. None of the extracts affected the body weight and internal organ weight at the test dose level (Tables IX and X).

Antipyretic activity

Both the extracts showed mild ($P<0.1$) antipyretic activity in rats against yeast induced pyrexia in rats at 3^{rd} and 4^{th} hour post test drug administration (Table XI and XII).

Discussion

It was evident from the results of the experiment that *Sisymbrium irio* seeds possess significant analgesic activity. Both decoction and aqueous extract of the test drug significantly increased the jump and lick response time at different time levels. Both lick and jump response times were significantly ($P<0.01$) increased at 3^{rd} hour post test drug decoction administration at both dose levels, whereas the same level of significance was achieved at higher dose level (2500 mg per kg body weight) of aqueous extract. The threshold tail flick response was significantly ($P<0.01$) higher at 180 minutes post test drug administration at higher dose, while at lower dose level moderate analgesia ($P<0.05$) could be achieved. None of the extracts could prevent the mice against acetic acid induced writhing and showed mild analgesia ($P<0.1$) in comparison with the distilled water control group.

Both the extracts showed the mild (P<0.1) anti-inflammatory activity in drugs. While the decoction showed 11.11% reduction of paw oedema at lower dose, aqueous extract showed 16.66% reduction at the dose level in comparison with the control group. At higher dose decoction and aqueous extract showed 27.77% and 23.33% anti-inflammatory activity respectively. It showed mild (P<0.1) inhibition of granulation tissue formation in rats at either doses. The active principle in the seed is flavonoid, iso rhamnetin [10] and flavonoids have been shown to possess anti-inflammatory activity [11]. Compounds with anti-inflammatory activity also possess antinociceptive activity[12] and antipyretic activity [13]. *Sisymbrium irio* seeds also contain essential fatty acids such as gamma linolenic acid (GLA), oleic acid and erucic acid [10] and GLA upon oxidative metabolism is known to produce anti-inflammatory eicosanoids in the body [14]. Neither extracts showed significant effect on body weight gain and weight of internal organs at both dose levels. Mild antipyretic activity was observed with respect to both the extracts of *Sisymbrium irio* seeds against yeast induced pyrexia in rats at 3rd and 4th hour post test drug administration and inhibition of prostaglandin synthesis could be the mechanism of antipyretic action.

Table-I

Effect of *S. irio* seeds decoction on hot plate analgesia in mice.

Group	Reaction time in various time intervals							
	Initial		1 hour		2 hour		3 hour	
	LR	JR	LR	JR	LR	JR	LR	JR
Distilled water (Control)	6.83 ±0.4	8 ±0.57	7.16 ± 0.30	8.33 ±0.49	7.16 ±0.47	7.66 ±0.42	7.5± 0.49	7.66 ±0.49
S.irio 1250 mg per kg	6 ±0.25	8.5 ±0.22	6.5* ±0.22	9* ±0.25	7.83*** ±0.30	9.83*** ±0.40	7.83*** ±0.30	10.5*** ±0.42
S.irio 2500 mg per kg	5.83 ±0.4	8 ±0.25	6.83** ±0.3	8.66** ±0.33	7.83*** ±0.16	9.5*** ±0.22	8.33*** ±0.71	9.16*** ±0.40
Ibuprofen 100 mg per kg	4.5 ±0.22	6.66 ±0.42	7.83**** ±0.30	10.33**** ±0.42	8.33**** ±0.21	11.5**** ±0.34	8.83**** ±0.40	13**** ± 0.36

* P<0.1, ** P<0.05, *** P<0.01, **** P<0.001 LR-Licking Response, JR-Jumping Response

Table-II
Effect of *S. irio* seeds aqueous extract on hot plate analgesia in mice

Group	Reaction time in various time intervals							
	Initial		1 hour		2 hour		3 hour	
	LR	JR	LR	JR	LR	JR	LR	JR
Distilled water (Control)	6.16 ±0.40	7.83 ±0.16	6.5 ±0.34	8.16 ±0.47	6 ±0.57	7.5 ±0.22	6.33 ±0.42	7.5 ±0.22
S.irio 1250 mg per kg	6.16 ±0.30	7 ±0.36	6.33 ±0.21	6.83 ±0.40	6.66* ±0.33	7.5* ±0.22	7** ±0.36	8** ±0.36
S.irio 2500 mg per kg	5.16 ±0.31	6.66 ±0.21	5.83** ±0.47	7.16** ±0.30	5.83** ±0.47	7.33** ±0.33	6.66*** ±0.49	8*** ±0.25
Ibuprofen 100 mg per kg	4.66 ±0.21	6.33 ±0.33	7.33**** ±0.21	8.33**** ±0.21	8.33**** ±0.33	9.83**** ±0.30	9.33**** ±0.33	12.33**** ±0.42

* $P<0.1$, ** $P<0.05$, *** $P<0.01$, **** $P<0.001$ LR-Licking Response, JR-Jumping Response

Table-III
Effect of *S.irio* seeds decoction on acetic acid induce writhing in mice

Group	No. of mice protected	No. of stretching episodes during 30 minutes after acetic acid injection Mean ± SEM	'P' value
Distilled water (Control)	0/6	51.16 ± 1.27	-
S.irio 1250 mg ml per kg	0/6	53.5 ± 1.74	NS
S.irio 2500 mg per kg	0/6	48 ± 1.21	0.1
Ibuprofen 100 mg per kg	0/6	46.66 ± 1.68	0.01

Table-IV

Effect of *S.irio* seeds aqueous extract on acetic acid induce writhing in mice

Group	No. of mice protected	No. of stretching episodes during 30 minutes after acetic acid injection Mean ± SEM	'P' value
Distilled water (Control)	0/6	54.1 ± 1.73	-
S.irio 1250 mg per kg	0/6	53.5 ± 1.87	NS
S.irio 2500 mg per kg	0/6	53.33 ± 1.45	0.1
Ibuprofen 100 mg per kg	0/6	52.1 ± 1.84	0.01

Table-V

Effect of *S. irio* seeds decoction on latency of tail flick response in rats.

Group	Reaction time in Sec ± SEM					
	Recording at various time intervals (Min. after drug admn.)					
	Initial	30	60	120	180	240
Distilled water (Control)	3.33 ±0.61	3.16 ±0.33	3 ±0.28	3 ±0.28	2.83 ±0.18	2.83 ±0.18
S.irio 1250 mg per kg	2.5 ±0.22	2.83 ±0.30	3.16** ±0.16	3.16** ±0.16	3.66** ±0.21	3.66** ±0.21
S.irio 2500 mg per kg	3.16 ±0.16	3.83 ±0.30	3.83** ±0.30	4.16** ±0.31	4.5*** ±0.22	5*** ±0.36
Ibuprofen 100 mg per kg	3.33 ±0.21	4**	4.33*** ±0.21	4.66** ±0.21	5.5**** ±0.22	5.5**** ±0.22

** P<0.05, *** P<0.01, **** P<0.001

Table-VI
Effect of *S. irio* seeds aqueous extract on latency of tail flick response in rats.

Group	Reaction time in Sec ± SEM					
	Recording at various time intervals (Min. after drug admn.)					
	Initial	30	60	120	180	240
Distilled water (Control)	3.83 ±0.30	3.33 ±0.21	3.16 ±0.16	3.16 ±0.16	3.33 ±0.42	3.33 ±0.42
S.irio 1250 mg per kg	3.83 ±0.30	3.66 ±0.33	3.58 ±0.27	3.33 ±0.33	4.5** ±0.34	4.5** ±0.34
S.irio 2500 mg per kg	3.33 ±0.21	3.66 ±0.42	3.83* ±0.40	4** ±0.34	4.5** ±0.34	4.66*** ±0.33
Ibuprofen 100 mg per kg	3 ±0.25	3.66** ±0.21	3.83*** ±0.16	4.5*** ±0.22	4.66*** ±0.21	5.56**** ±0.30

* $P<0.1$, ** $P<0.05$, *** $P<0.01$, **** $P<0.001$

Table-VII
Effect of *S.irio* seeds decoction on Carrageenan induced hind paw oedema in rats

Group	Volume of paw oedema in ml. Mean ± SEM	Percentage reduction	'P' value
Distilled water (Control)	0.18 ± 0.03	-	-
S.irio 1250 mg per kg	0.16 ± 0.01	11.11	0.1
S.irio 2500 mg per kg	0.13 ± 0.03	27.77	0.1
Ibuprofen 100 mg per kg	0.08 ± 0.02	55.55	0.01

Table-VIII
Effect of *S.irio* seeds aqueous extract on Carrageenan induced hind paw oedema in rats

Group	Volume of paw oedema in ml. Mean ± SEM	Percentage reduction	'P' value
Distilled water (Control)	0.3 ± 0.06	-	-
S.irio 1250 mg per kg	0.25 ± 0 025	16.66	0.1
S.irio 2500 mg per kg	0.23 ± 0.042	23.33	0.1
Ibuprofen 100 mg per kg	0.13 ± 0.03	56.66	0.01

Table-IX
Effect of *S.irio* seeds decoction on granulation tissue formation in cotton pellet granuloma in rats.

Group	Weight of granulation (mg. per 100 g. body weight) Mean SEM	Percentage Inhibition	'P' value
Distilled water (Control)	58.5 ± 1.96	-	-
S.irio 1250 mg per kg	54.50 ± 1.80	6.83	0.1
S.irio 2500 mg per kg	49.33 ± 1.60	15.67	0.1
Ibuprofen 100 mg per kg	33.5 ± 1.91	42.73	0.01

Table-X
Effect of *S.irio* seeds aqueous extract on granulation tissue formation in cotton pellet granuloma in rats.

Group	Weight of granulation (mg. per 100 g. body weight) Mean SEM	Percentage Inhibition	'P' value
Distilled water (Control)	54.44 ± 2.01	-	-
S.irio 1250 mg per k g	48.85 ± 1.38	11	0.1
S.irio 2500 mg per kg	48.08 ± 2.81	11.68	0.1
Ibuprofen 100 mg per kg	27.01 ± 2.28	50.38	0.01

Table-XI
Effect of *Sisymbrium irio* seeds decoction on Yeast induced pyrexia in rats.

Group	Rectal temperature(^{0}C) at various time intervals after drug administration				
	0 hr	1 hr	2 hr	3 hr	4 hr
Distilled water (Control)	38.43 ±0.14	38.5 ±0.14	38.5 ±0.17	38.56 ±0.19	38.53 ±0.07
S.irio 1250 mg per kg	38.66 ±0.22	38.6 ±0.22	38.6 ±0.25	38.23* ±0.35	38.2* ±0.37
S.irio 2500 mg per kg	38.46 ±0.27	38.46 ±0.27	38.4 ±0.26	38.23* ±0.27	38* ±0.30
Ibuprofen 100 mg per kg	38.46 ±0.15	37.85** ±0.27	37.4*** ±0.26	37.06*** ±0.24	36.71**** ±0.25

* P<0.1, ** P<0.05, *** P<0.01, **** P<0.001

Table-XII

Effect of *Sisymbrium irio* seeds aqueous extract on Yeast induced pyrexia in rats.

Group	Rectal temperature(°C) at various time intervals after drug administration				
	0 hr	**1 hr**	**2 hr**	**3 hr**	**4 hr**
Distilled water (Control)	38.8 ±0.19	38.86 ±0.24	38.9 ±0.22	38.8 ±0.24	38.8 ±0.24
S.irio 1250 mg per kg	38.86 ±0.17	38.83 ±0.2	38.8 ±0.2	38.6* ±0.26	38.53* ±0.29
S.irio 2500 mg per kg	38.83 ±0.14	38.83 ±0.14	38.76 ±0.15	38.63* ±0.19	38.3* ±0.29
Ibuprofen 100 mg per kg	38.46 ±0.3	38* ±0.13	37.66** ±0.26	37.45*** ±0.26	36.98**** ±0.18

* P<0.1, ** P<0.05, *** P<0.01, **** P<0.001

References

1. Anonymous. Wealth of India: Publication and Information Directorate, CSIR, New Delhi. 1972; Vol. IX, p. 361.

2. Sharma PV. Dravyaguna Vijnana, Vol. II, Chowkamba Bharati Academy, Varanasi. 1998; p. 271.

3. Turner RA. Screening methods of Pharmacology. Vol. I, Academic press, New York. 1965; p. 26-34, 298.

4. Eddy NB, Leimbach DJ. Hand book of experimental Pharmacology Ed. By S.K. Kulkarni (1999) Vallabh Prakashan Delhi. 1953; p.125.

5. Witkin LB, Huebner CF, Caldi F, Keefe E, Spitaletta P, Plummer AJ. Pharmacology of 2-aminoindane hydrochloride (Su.89 -29), a potent no narcotic analgesic. Journal of Pharmacology and Experimental Therapeutics.1961;133: 400.

6. Gujral MC, Khanna BK. Comparative evaluation of some narcotic analgesics. Journal of Scientific &Industrial Research. 1956; Vol. 168: 11-13.

7. Winter CA, Risley EA, Nuss, G.W. Carrageenan induced oedema in hind paw of the rat as an assay of anti-inflammatory drugs. Proceeding of the Society for Experimental Biology and Medicine. 1962; Vol. III.:544-547.

8. Winter CA, Portar C.C. Effect of alteration inside chain upon anti-inflammatory and liver glycogen activities of hydrocortisone esters. Journal of American Pharmaceutical Association. 1957; 46:515-520.

9. Singh H, Ghosh MN. Modified plethysmometer for measuring foot volume of anaesthetized rats. Journal of Pharmacy and Pharmacology. 1968; 21:136.

10. Khare CP. Indian Herbal remedies, Springer, India. 2004; p. 427.

11. Kim HP, Son KH, Chang HW, Kang SS. Antiinflammatory plant flavinoids and cellular action mechanisms. Journal of Pharmacological Science. 2004; 96: 229-245.

12. Attaway DH, Zaborsky OR. Marine Biotechnology: Pharmaceutical and bioactive natural products. Plenum Press, New York. 1993; Vol. 1, p.1-23.

13. Periyanayagam JP, Sharma SK, Joseph A, Christina AJM. Evaluation of antipyretic and analgesic activity of *Emblica officinalis* Gaertn. Journal of Ethnopharmacology. 2004; 95: 83-85.

14. Kapoor R, Huang YS. Gamma Linolenic acid: an anti-inflammatory omega -6 fatty acid. Current Pharmaceutical Biotechnology. 2006; 7 (6):531-534.

सारांश

वाई आर संजय कुमार, एन जया एवं सी के सशिकला

प्रस्तुत लेख मे जंगली सरसों (सिसिम्ब्रियम इरियों. लिन.) बीज का क्वाथ और जलविलय (एक्यूस) सात्व पर कतिपय भेषजगुण कर्म विज्ञानीय अध्ययन प्रस्तुत किया गया है। क्वाथ और जलविलय सत्वों का चूहों में, तीव्र वेदनाहर, अल्प शोथहर और ज्वरहर प्रभाव है। इस अध्ययन से यह भी पता चला कि क्वाथ का प्रभाव जलविलय सात्व से अधिक है।

(Approved on 02/05/2013)