

Evaluation of Anti-inflammatory Activity of *Asparagus racemosus* Willd. (Liliaceae) root extract

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Abstract – The methanol extract of *Asparagus racemosus* root was evaluated for its anti-inflammatory activity on carrageenin and serotonin-induced rat hind paw oedema models. The extract (200 and 400 mg/kg) showed maximum inhibition of oedema of 18.6% and 33.7% at 3 h with carrageenin and 22.2% and 40.5% at 5 h with serotonin-induced rat paw oedema respectively. The experimental models tested, where the effect produced by the extract was compared to that of phenylbutazone, a prototype non-steroidal anti-inflammatory drug.

Key words – *Asparagus racemosus* root extract, anti-inflammatory activity, carrageenin and serotonin-induced, phenylbutazone.

Introduction

Asparagus racemosus Willd. (Liliaceae) commonly known as Satamuli (Bengali), Shatavari (Hindi and Sanskrit) is a tall climber under-shrub, found all over the India. All parts of this plant are used in Indian traditional (Ayurvedic and Yunani) system of medicine for the treatment of various ailment in human being. The roots are useful in rheumatism, dysentery, tumour, billiousness, epilepsy and in inflammation (Kirtikar and Basu, 1975; Nadkarni *et al.*, 1976). Presently it has come to our notice that tribal people of Bankura, West Bengal, India use the juice of this root of *A. racemosus* against inflammatory enlargement and get cure of such. The anti-inflammatory activity of *Leucas lavandulaefolia* Rees. extract has been performed in the same laboratory (Saha *et al.*, 1996).

In the light of above information, the present study was undertaken to evaluate the anti-inflammatory activity of the extract of this plant and is being reported in the present communication.

Experimental

Plant material – The roots of *A. racemosus* were collected in the month of October from Hetyasole, Bankura district of West Bengal, India and identified by Central National Herbarium, Botanical Survey of India, Shibpur, Howrah. A voucher specimen (DP-DPT/HS-1/92) has been kept in our laboratory for future references. The roots were cut into small pieces, dried under shade, pulverised by a mechanical grinder and stored in closed container for further use.

Preparation of extract – The powdered roots (500 g) were extracted with methanol (S.D. Fine Chemical, Bombay, India) in a

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soxhlet extractor. The extract was then distilled under reduced pressure, a reddish-brown coloured semisolid mass was obtained (yield 28.17% w/w with respect to dry powdered material) and used for evaluation of anti-inflammatory activity by suspending with Tween 80 in different doses.

Phytochemical screening – Freshly prepared extract of *A. racemosus* showed the positive Liebermann-Burchard reaction for steroid (Liebermann, 1885) and a positive Noller test for triterpenoid (Noller *et al.*, 1942), which were confirmed by thin layer chromatography with the solvent system benzene:methanol (9:5) over silica gel G (Stahl, 1969).

Animals used – Male albino Wister rats weighing 200-250 g supplied by M/s. B. N. Ghosh & Co., Calcutta, India, were placed in cages with wire-net floors in a controlled room temperature $22 \pm 2^\circ\text{C}$, relative humidity 60-70% and provided with food and water *ad libitum*. The animals were deprived of food for 24 h before experimentation but allowed free access to tap water throughout. All studies were carried out by using six rats in each group.

Carrageenin-induced rat paw oedema – Oedema was induced by subplanter injection of 0.1 ml of a 1% freshly prepared suspension of carrageenin (Sigma Chemical Co., USA) in normal saline into the right hind paws of the rats, according to the method of Winter *et al.*, 1962. The extract (200 and 400 mg/kg), standard drug phenylbutazone (100 mg/kg) were given (intraperitoneally) 30 min before the stimulus (carrageenin) injections. Control animals received an equivalent volume of 0.9% saline.

Serotonin-induced inflammation – In this model oedema of right hind paws of the rats was induced by subplanter injection of 0.05 ml of 1% solution of serotonin in 0.9% saline water. *A. racemosus* root extract (200 and 400 mg/kg), phenylbutazone, a standard anti-inflammatory drug (100 mg/kg) were

given (i.p.) 30 min before carrageenin injection. Control animals were treated like before.

The volume of the paws was measured by a plethysmometer (Ugo, Basili, Italy) before and 1, 2, 3, 4, and 5 h after carrageenin and serotonin injection. The oedema rate and inhibition rate were calculated as follows (Lin *et al.*, 1995).

$$\text{Oedema rate (E)\%} = \frac{V_r}{V_c} \times 100$$

$$\text{Inhibition rate (I)\%} = \frac{E_c - E_t}{E_c} \times 100$$

Where V_c is the contralateral paw volume of the rat (left hind paw without carrageenin) at t-hour, V_r is the right hind paw volume of rats (with carrageenin) at t-hour, E_c is the oedema rate of the control group and E_t is the oedema rate of the treated group.

Statistical analysis – The experimental results are expressed as the mean \pm SEM and the statistical significance was evaluated by student's t-test (Woodson, 1987).

Results and Discussion

The anti-inflammatory evaluation of extract of *A. racemosus* against acute pedal oedema is showing significant anti-inflammatory activity comparable to that of phenylbutazone, a prototype non-steroidal anti-inflammatory agent. The extract showed maximum inhibition of inflammation 18.6 and 33.7% at 3 h when inflammation is induced by carrageenin and maximum inhibition of inflammation 22.2 and 40.5% at 5 h, inflammation-induced by serotonin, at a doses of extract 200 and 400 mg/kg respectively for both the models (Table 1, 2 and Fig. 1).

Carrageenin induced oedema is commonly used as an experimental animal model of acute inflammation and is believed to be biphasic. The first phase is due to release of histamine and serotonin, the second phase

Table 1. Effect of extract of *A. racemosus* root on carrageenin-induced rat paw oedema

Treatment	Dose (mg/kg)	Oedema rate percentage (Mean±SEM)				
		1h	2h	3h	4h	5h
Control	-	30.4±1.1	36.7±1.0	42.1±1.5	44.3±1.1	48.2±1.2
Extract	200	28.3±1.3 (7.7)	31.8±1.1 ^b (13.2)	34.2±1.2 ^b (18.6)	38.2±1.2 ^b (13.7)	42.1±1.0 ^b (12.5)
Extract	400	26.7±1.2 ^c (12.1)	28.7±1.3 ^a (21.7)	27.9±1.4 ^a (33.7)	33.0±1.3 ^a (25.3)	36.5±1.3 ^a (24.1)
Phenylbutazone	100	24.8±1.0 ^b (18.2)	28.2±1.2 ^a (23.1)	25.6±1.2 ^a (39.1)	30.4±1.1 ^a (31.2)	32.0±1.1 ^a (33.5)

N=6 animals per experiment, ^a $p < 0.001$, ^b $p < 0.01$, ^c $p < 0.05$ by student's *t*-test. Figures in parenthesis represent oedema inhibition percentage.

Table 2. Effect of *A. racemosus* root extract on serotonin-induced rat paw oedema

Treatment	Dose (mg/kg)	Oedema rate percentage (Mean±SEM)				
		1h	2h	3h	4h	5h
Control	-	28.3±1.2	35.7±1.3	41.5±1.4	45.2±1.2	48.8±1.1
Extract	200	25.4±1.1 (10.2)	29.8±1.0 ^b (16.5)	33.3±1.0 ^a (19.7)	36.1±1.1 ^a (20.1)	37.9±1.2 ^a (22.2)
Extract	400	23.5±1.3 ^c (16.8)	25.5±1.1 ^a (28.5)	26.1±1.3 ^a (37.1)	27.5±1.2 ^a (39.0)	29.0±1.4 ^a (40.5)
Phenylbutazone	100	22.5±1.0 ^b (20.2)	24.5±1.2 ^a (31.3)	24.8±1.2 ^a (40.2)	25.7±1.3 ^a (43.1)	24.7±1.2 ^a (49.3)

N=6 animals per experiment, ^a $p < 0.001$, ^b $p < 0.01$, ^c $p < 0.05$ by student's *t*-test. Figures in parenthesis represent oedema inhibition percentage.

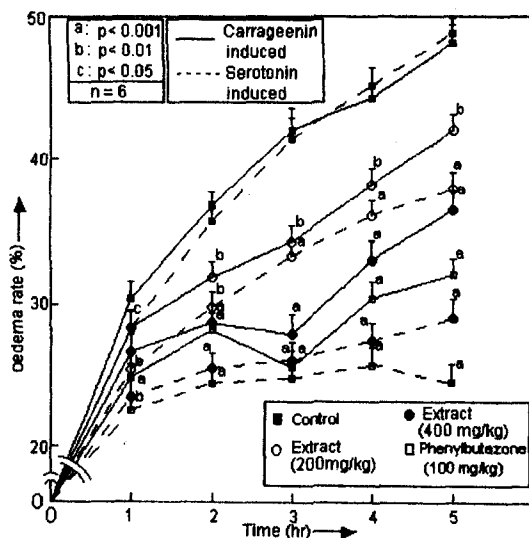


Fig. 1. Antiinflammatory activity of *A. racemosus* root on Carrageenin and Serotonin induced paw oedema in rats.

is caused by the release of bradykinin, protease, prostaglandin and lysosome (Castro *et*

al., 1968). It has been reported that the second phase of oedema is sensitive to most clinically effective antiinflammatory agents (Smucker *et al.*, 1967).

Subplanter injection of carrageenin and serotonin in rats caused a time dependent increase in paw volume. Carrageenin rat paw oedema is a suitable method for evaluating anti-inflammatory drugs which has been frequently used to assess the anti-oedematous effect of natural products (Della Loggia *et al.*, 1986; Alcaraz *et al.*, 1988). The effect of *A. racemosus* root extract and the inflammation process induced by serotonin indicates that they act by effecting a time delayed system in a similar fashion to glucocorticoids (Mukherjee *et al.*, 1997). This suggests that the active principles (steroid and triterpenoid) of extract having some degree of affinity for the glucocorticoid receptors.

Steroid and triterpenoid has been reported for its antiinflammatory activity (Mukherjee *et al.*, 1997). Since *A. racemosus* root extract also contain steroid and triterpenoid showing the similar activity.

So, the methanol extract of *A. racemosus* root has a good anti-inflammatory potential and supports the claims in folklore uses (Kirtikar and Basu, 1975; Nadkarni *et al.*, 1976). Further work relating to the isolation of the active constituents and allied approaches are in process in our laboratory.

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References

- Alcaraz, M. J. and Jimenez, M. J., Flavonoids as anti-inflammatory agents. *Fitoterapia* **59**, 25-38 (1988).
- Castro, J., Sasame, H., Sussman, H. and Bullette, P., Diverse effects of SKF₅₂ and antioxidants on CCl₄ induced changes in liver microsomal P-450 content and ethymorphine metabolism. *Life Sci.* **7**, 129-136 (1968).
- Della Loggia A., Tubaro, A., Dri, P., Zilli, C. and Del Negro, P., The role of flavonoids in the anti-inflammatory activity of *Chamomilla recutita*. *Clin. Biol. Res.* **213**, 481-488 (1986).
- Kirtikar, K. R. and Basu, B. D. *Indian Medicinal Plants.* **3**, Bishen Singh and Mahendra Pal Singh, Dehra Dun, 1975, pp. 2499-2501.
- Liebermann, C., *Ber.* 1803 (1885).
- Lin, C. C., Lin, W. C., Chang, C. H. and Namba, T., Anti-inflammatory and hepatoprotective effects of *Ventilago leiocarpa*. *Phytother. Res.* **9**, 11-15 (1995).
- Mukherjee, P. K., Saha, K., Das, J., Pal, M. and Saha, B. P., Studies on the antiinflammatory activity of rhizomes of *Nelumbo nucifera*. *Planta Medica* **63**, 387-389 (1997).
- Nadkarni, K. M., Nadkarni, A. K. and Chopra, R. N., *Indian Material Medica* **1**, Popular Prakashan, Bombay, 1976, pp. 153-155.
- Noller, C. R., Smith, R. A., Harris, G. R. and Walker, J. W., Saponins and Sapogenins XX. Some color reactions of triterpenoid sapogenins. *J. Amer. Chem. Soc.* **64**, 3047-3049 (1942).
- Saha, K., Mukherjee, P. K., Das, J., Mandal, S. C., Pal, M., and Saha, B. P., Antiinflammatory evaluation of *Leucas lavandulaefolia* Rees. extract. *Nat. Prod. Sci.* **2**(2), 119-122 (1996).
- Smucker, E., Arrhenins, E. and Eultin, T. Alteration in microsomal electron transport, oxidative N-demethylation and azo-dye cleavage in CCl₄ and dimethyl nitrosamine induced liver injury. *Biochem. J.* **103**, 55-64 (1967).
- Stahl, E., *Thin-layer chromatography*, Springer-Verlag, New York, 1969, pp. 329-334.
- Winter, C. A., Risley, E. A. and Nuss, G. W., Carrageenin induced oedema in hind paw of the rat as assay for anti-inflammatory drugs. *Proc. Soc. Exp. Biol. Med.* **111**, 544-547 (1962).
- Woodson, R. F., *Statistical methods for the analysis of biochemical data*, Series in probability and mathematical statistics. Wiley, New York, 1987, pp. 315-316.

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