

ANTI-INFLAMMATORY ACTIVITY OF THE LEAF EXTRACTS/FRACTIONS OF *BRYOPHYLLUM PINNATUM* SALIV.SYN

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ABSTRACT

The various extracts/fractions of leaves of *Bryophyllum pinnatum* were investigated in chemically-induced inflammation rodents model. The extracts/fractions inhibited formaldehyde-induced paw edema in rats. These inhibitions were statistically significant ($p < 0.05-0.01, 0.001$) as compared to control. Methanolic extract showed highest activity.

Keywords: *Bryophyllum pinnatum*, anti-inflammatory activity, leaf extract.

INTRODUCTION

Bryophyllum pinnatum (Crassulaceae) is widely used medicinal plant in traditional system with a wide range of biological activities. It is a succulent glabrous herb 0.3-1.2 m high, stems obtusely 4-angled, the older light colored, the younger reddish speckled white, leaves variable, decussate, the lower usually simple or occasionally compound, the upper usually 3-5 or 7-folliolate, long-petiole, petiole united by a ridge round the stem. Leaflet ovate or elliptical, crenate or serrate rich in alkaloids, triterpenes, glycosides, flavanoids, steroids and lipids¹. The leaves contain a group of chemicals called bufadienolides which are very active similar in structure and activity as two other cardiac glycosides, digoxin and digitoxin². The leaves are used as astringent, refrigerant, emollient, mucilaginous, haemostatic, vulnerary, depurative, constipating, anodyne, carminative, disinfectant and tonic. They are useful in vitiated conditions pitta and vata, haematemesis, haemorrhoids, menorrhagia, cuts and wounds, discolourations of the skin, boils, sloughing ulcers, ophthalmia, burns, scalds, corn, diarrhea, dysentery, vomiting³. Formaldehyde will induce an inflammatory reaction at the site of injection, and this has been used for studying inflammation quantitatively in the rat foot⁴. Selye first described the effect of injected formaldehyde in the rat foot as an arthritic reaction⁵. The reaction of the rat foot to formaldehyde has been investigated pharmacologically by Parratt and West, who showed that the reaction was not antagonized by inhibitors of either histamine or 5-hydroxytryptamine⁶. Selye has shown that in the adrenalectomized rat both cortisone and adrenocorticotropic hormone are effective in antagonizing the reaction to formaldehyde. It has been reported by Setnikar, Salvaterra and Temelcou that the swelling of the rat foot caused by an injection of formaldehyde can be partially prevented by iproniazid, phenylbutazone and salicylamide⁷. Recently, Buch has shown that various phenylpropylcarbamates are effective in reducing the reaction to formaldehyde⁸.

This study has been undertaken to evaluate the anti-inflammatory activity of the water, petroleum ether,

chloroform, acetone and methanol leaf extracts and alkaloidal fraction, flavonoids fraction, phenolic acid fraction, alkaloidal anhydride fraction of *Bryophyllum pinnatum* using Formaldehyde induced paw edema model of inflammation.

MATERIALS AND METHODS

Collection and extraction of plant material

The leaves of *Bryophyllum pinnatum* were collected from the herbal garden of B.B.D.N.I.T.M, Lucknow, U.P., India, and were identified in the taxonomy division, N.B.R.I, Lucknow, U.P., India. The dried and powdered leaves were successively extracted with various solvents in Soxhlet apparatus. The solvent extracts were concentrated separately using Buchi rota evaporator and dried under vacuum. The dried extracts were preserved in desiccator until further use.

Animals

Sprague Dawley rats (120-150 g) and Swiss albino mice (40-50 g) were used in these experiments. They were housed in polypropylene cages and kept in room temperature maintained under controlled condition. All animals were fed with a standard diet ad libitum and had free access to drinking water. Animal study was performed in institutional animal house and according to CPCSEA norms.

Formaldehyde-induced rat hind paw edema

The effects of extracts/fractions and indomethacin on the acute phase of inflammation were investigated. Doses of extracts/fractions were administered orally once a day for a period of 2 days. An hour after the last dose was administered; 0.2 ml of formaldehyde (1%, w/v) was injected into the rat hind paw. Before formaldehyde injection, the paw volume for each rats were measured separately by means of Plethysmometer. Edema caused by formaldehyde was measured at 3, 6 and 24 h the first day, and measured once per day on the following days until inflammation disappeared. The anti-inflammatory potency of extracts/fractions was determined by comparing it with

a group in which a 10 mg/kg dose of indomethacin was administered orally. The ratio of the anti-inflammatory effect of extracts/fractions was calculated by the following equation:

$$\text{Anti-inflammatory activity (\%)} = (1-D/C) \times 100$$

Where D represents the percentage difference in paw volume after extracts/fractions were administered to the

rats and C represents the percentage difference of volume in the control group.

Statistical analysis

Body weight and volume of paw level was analyzed by using one way new man-Keulis ANOVA with repeated measures. The level of significance was set at $p < 0.05$.

RESULTS

Table 1: Effect of extracts/fractions on paw edema induced by formaldehyde in rats:

Groups	Dose (mg/kg)	Mean paw volume \pm SEM(ml) and % Inhibition (P.I)			
		Before formaldehyde	Time after formaldehyde injection		
			+3h	+6h	+24h
I.	-	0.81 \pm 0.03	1.42 \pm 0.32	1.67 \pm 0.89	-
II.	10	0.84 \pm 0.08	1.27 \pm 0.42 (57.30)***	1.10 \pm 0.21 (67.44)***	1.12 \pm 0.45 (74.15)***
III.	500	0.87 \pm 0.06	1.66 \pm 0.54 (9.19)	1.60 \pm 0.66 (15.11)	1.56 \pm 0.74 (20.68)
IV.	500	0.81 \pm 0.04	1.54 \pm 0.62 (9.87)	1.48 \pm 0.53 (22.09)	1.47 \pm 0.21 (21.99)
V.	500	0.80 \pm 0.67	1.64 \pm 0.65 (5.00)	1.59 \pm 0.42 (8.13)	1.58 \pm 0.76 (8.01)
VI.	500	0.89 \pm 0.42	1.20 \pm 0.41 (57.14)***	1.20 \pm 0.80 (63.95)***	1.10 \pm 0.43 (69.94)***
VII.	500	0.83 \pm 0.51	1.45 \pm 0.78 (25.30)	1.38 \pm 0.56 (36.00)	1.40 \pm 0.21 (34.76)
VIII.	500	0.82 \pm 0.69	1.18 \pm 0.45 (56.09)***	1.15 \pm 0.67 (63.05)***	1.06 \pm 0.54 (64.98)***
IX.	500	0.80 \pm 0.54	1.25 \pm 0.65 (43.75)**	1.20 \pm 0.78 (53.48)**	1.16 \pm 0.62 (51.78)**
X.	500	0.82 \pm 0.47	1.46 \pm 0.87 (21.95)	1.42 \pm 0.54 (30.23)	1.38 \pm 0.87 (28.98)
XI.	500	0.86 \pm 0.61	1.28 \pm 0.85 (51.52)**	1.24 \pm 0.47 (55.81)**	1.18 \pm 0.32 (53.74)**

Group-I: Diseased control,

Group-II: Diseased rat treated with standard indomethacine drug,

Group-III: Diseased rat treated with pet-ether extract,

Group-IV: Diseased rat treated with chloroform extract,

Group-V: Diseased rat treated with acetone extract,

Group-VI: Diseased rat treated with methanol extract,

Group-VII: Diseased rat treated with aqueous extract,

Group-VIII: Diseased rat treated with alkaloidal fraction,

Group-IX: Diseased rat treated with flavonoids fraction,

Group-X: Diseased rat treated with phenol and phenolic acid,

Group-XI: Diseased rat treated with alkaloidal anhydride, 0.05<p, 0.01<p**, 0.001<p*** all values in \pm SEM.

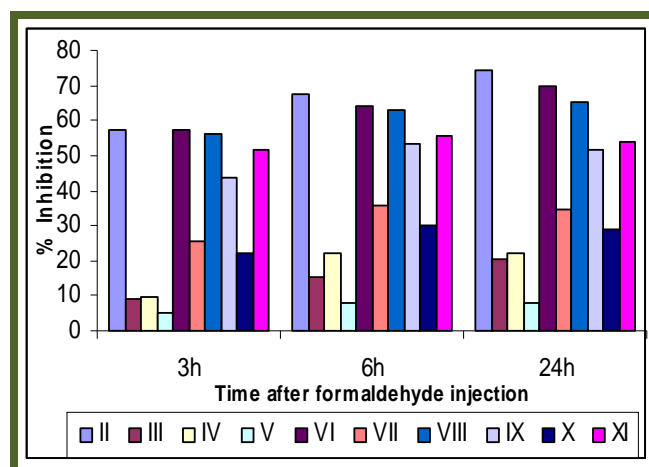


Figure 1: Percentage inhibition of formaldehyde induced edema in rats by various extracts/fractions.

DISCUSSION

Indomethacin showed more or less uniform inhibition of edema in early intermediate and later phases. Methanolic fraction showed also more or less significant inhibition of formaldehyde induced edema in early phases while significant inhibition at later phases. Out of Pet-ether, Chloroform, Acetone and Methanol fractions from *B. pinnatum* leaves, Methanol fraction was more significant than the other fractions in percentage inhibition of paw edema. Acute inflammation induced by formaldehyde results from cell damage, which provokes the production of endogenous mediators, such as, histamine, serotonin, prostaglandins, and bradykinin⁹. It is well known that inhibition of edema induced by formalin in rats is one of the most suitable test procedures to screen antiarthritic and anti-inflammatory agents as it closely resembles human arthritis. Arthritis induced by formalin is a model used for the evaluation of an agent with probable antiproliferative activity. As some of the above fractions significantly inhibited this model of inflammation they can be thought to possess antiproliferative and antiarthritic activities similar to indomethacin, a cyclooxygenase inhibitor.

REFERENCES

1. Kirtikar KR, Basu BD. *Indian Medicinal Plants*. 2nd edn. vol. 2. Allahabad :Oriental Enterprises 2000:1511.
2. Rastogi RP, Mehrotra BN. *Compendium of Indian Medicinal Plants*, vol. 5. New Delhi: Photocomposing-cum-printing unit 1990:141.
3. Khare CP, *Encyclopedia of Indian Medicinal Plants*, New York: Springer 2004:276.
4. Parrat JR, West GB, 5-Hydroxytryptamine and the anaphylacoid reaction in rat. *J. Physiol.*, 139:27-41.
5. Selye H, Further studies in the participation of the adrenal cortex in the pathogenesis of arthritis. *Brit.med.J.*2:1129-1135.
6. Parrat JR, West GB, Inhibition by various substances of edema formation in the hind paw of the rat induced by 5-Hydroxytryptamine, histamine, eggwhite, and compound 48/80. *Brit. J. Pharmacol.* 13:65-70.
7. Setnikar I, Salvaterra M, Temelcou O, Antiphlogistic activity of iproniazid, *Brit. J. Pharmacol.* 14:484-487.
8. Buch O, Anti-inflammatory properties of 3-phenylpropylcarbamate, *Arch. int. Pharmacodyn.* 123:140-147.
9. Yuh-Fung C, Huei-Yann T, Tian-Shung W. Anti-inflammatory and analgesic activities from roots of *Angelica pubescens*. *Planta Med.* Vol.61. 1995: 2-8.
