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PRELIMINARY STUDIES ON ANTI-INFLAMMATORY ACTIVITIES OF *DIPLAZIUM ESCULENTUM* IN EXPERIMENTAL ANIMAL MODELS

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Asst. Professor (Medicinal chemistry), School of Pharmacy, ACHS, Asmara, Eritrea To study the anti-inflammatory effect of different extract of leaves of *Diplazium esculentum* in rats. Anti-inflammatory activity was screened by measuring the reduction in carrageenan induced hind paw edema. The potency of various extracts of leaves was compared with each other and standard Ibuprofen (100mg/kg) for anti-inflammatory activity. The results indicated that all the extracts of *Diplazium esculentum* showed anti-inflammatory activity. Percentage inhibition of inflammation observed in chloroform extract group was more than other treated groups.

ABSTRACT

INTRODUCTION: Higher plants have served humankind as sources of medicinal agents since its earliest beginnings. Infact, natural products once served as the source of all drugs. Today, natural products still represent over 50% of all drug in clinical use, with higher plant-derived natural products representing 25% of the total. On numerous occasions, the folklore records of many different cultures have provided leads to plants with useful medicinal properties ¹. The plant Diplazium esculentum (Dryopteridacea), commonly called "Latawar" is considered being of high economic value amongst the natives of Tehri hills and adjoining areas in Uttar Pradesh, India. The plant contains steroids, triterpenoids, phenols, flavones and flavonoids². The leaves are used as vegetables and the dried rhizomes used as insecticides. The decoction is used for cure of haemoptysis and cough ³. The vegetable shoots posses antioxidant activity ⁴. The crude ethanol extract of the plant showed marginal anti cancer activity ⁵. Keeping these points in mind the aim of the study was design to screen the leaves of Diplazium esculentum for anti-inflammatory effect in order to establish its folklore use in Uttaranchal India.

MATERIALS AND METHODS:

Collection of the plant: The plant material *Diplazium esculentum* was collected from Dehradun, Uttaranchal, India, and Identified by Dr. H.C. Pandey, Scientist, Botanical Survey of India (BSI), Dehradun, Uttarakhand, India.

Preparation of the plant Extract: The plant material was dried in shade, powdered and passed through 40-mesh sieve. Dried powder 200g was taken and subjected to successive extraction with petroleum ether (40-60), chloroform, acetone and methanol in a soxhlet apparatus using hot continuous percolation method. The extracts were concentrated under reduced pressure in rotary evaporator. The yields of dry extracts were (4.0, 3.5, 2.5 and 5.0 % w/w respectively) recorded and stored in a clean glass bottles for biological activity.

Animals: Albino rats of Wistar strain (150-200gm) either sex was procured from central animal house of the institute. They were housed in standard polypropylene cages and kept under controlled room temperature ($24\pm2^{\circ}$ C: relative humidity 60-70%) in a 12 h light-dark cycle. The rats were given a standard laboratory diet and water *ad libitum*. Food was withdrawn 12h before and during the experimental hours. The institutional animal ethics committee approved all experimental protocols.

Anti-inflammatory activity: The petroleum ether (E-1), chloroform (E-2), acetone (E-3) and methanol (E-4) extracts were evaluated for its anti-inflammatory activity by carrageenan induced rat paw edema method 6 .

The animals were divided into six groups comprising six animals each. First group marked as control & received normal saline (10ml/kg p.o.), second, third, fourth and fifth group (test groups) received E-1, E-2, E-3 and E-4 extracts of Diplazium esculentum at a dose level of 100mg/kg p.o. respectively. The sixth group served as positive control and received ibuprofen (100mg/kg body weight). The drugs were given orally with the help of an oral catheter. After one hour, a sub plantar injection of 0.1 ml of 1% solution of carrageenan was administered in the right hind paw of each rat. The paw volume was measured plethysmograph (UGO, Basile, Italy) at '0' and '3' hours after the carrageenan injection. The difference between the two readings was taken as the volume of edema and the percentage anti-inflammatory activity was calculated.

Statistical analysis: The data were expressed as mean \pm SD and statistically assessed by one-way analysis of variance (ANOVA)⁷. The difference between drug treated animals and controls were evaluated by Dunnett's t-test. Values of p <0.001 were considered statistically significant.

RESULTS: The results of the animal experiment are shown in **table 1**.

Treatment	Dose (mg/kg p.o.)	Mean Paw volume (ml) <u>+</u> SEM 3 hr	Inhibition (%) (After 3hr)
Control	10 ml/kg	0.206 <u>+</u> 0.088	-
E-1	100	0.17 <u>+</u> 0.11	17.47%
E-2	100	0.074 <u>+</u> 0.067*	64.07%
E-3	100	0.10 <u>+</u> 0.093*	51.45%
E-4	100	0.106 <u>+</u> 0.13	48.54%
Ibuprofen	100	0.064+ 0.020*	68.93 %

TABLE 1: ANTI-INFLAMMATORY ACTIVITY OF DIFFERENT EXTRACTS OF *DIPLAZIUM ESCULENTUM* ON CARRAGEENAN INDUCED RAT HIND PAW EDEMA

Values are mean \pm S.E. (n=5) *p < 0.001 vs. Control

In the acute inflammation model, the chloroform and acetone extracts of *Diplazium esculentum* in a dose level of 100mg/kg. p.o. showed significant reduction in the edema volume which is comparable to the standard drug ibuprofen.

DISCUSSION: Carrageenan induced hind paw edema is the standard experimental model was used for acute inflammation. Carrageenan is the phlogistic agent of choice for testing anti-inflammatory drugs as it is not known to be antigenic and is devoid of apparent systemic effect. Moreover the experimental models exhibit a high degree of reproducibility. Carrageenaninduced edema is a biphasic response. The first phase is mediated through the release of histamine, serotonin and kinins whereas the second phase is related to the release of prostaglandin and slow reacting substances which peak at 3 hrs.⁸ .The significant anti-inflammatory effect of E-2 and E-3 extracts of Diplazium esculentum appears to be similar to that of ibuprofen, which could be related to its histaminic, kinin and prostaglandin inhibitory activity.

CONCLUSION: The present study concludes that the plant *Diplazium esculentum* selected for anti-

inflammatory activity has shown appreciable results which supports the claim of local people and much work in this direction has to be done to confirm its utility in higher models.

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