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ANTI-INFLAMMATORY AND ANALGESIC ACTIVITIES OF ETHYL ACETATE EXTRACT OF ROOTS OF *EUGENIA JAMBOLANA*

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ABSTRACT

The present study was carried out to evaluate the anti-inflammatory and analgesic activities of the ethyl acetate extract of roots of *Eugenia jambolana*. The ethyl acetate extract of *Eugenia jambolana* in gum acacia was administered orally at 150 and 300mg/kg. The results of the present study revealed that the treatment groups showed a significant reduction in paw volume in a dose dependent manner indicating their anti-inflammatory action, which had provided a proof for the scientific validation of their ethno pharmacological property.

Keywords: Anti-inflammatory, Analgesic, Eugenia jambolana, Paw

INTRODUCTION

Inflammation is the response of living tissues to injury. It involves a complex array of enzyme activation, mediator release, cell migration, tissue breakdown and repair. ^[1, 2] Drugs that are currently used for the management of pain are opioids or nonopioids and that for inflammatory conditions are non-steroidal anti-inflammatory drugs (NSAIDs) and corticosteroids. All these drugs carry potential toxic effects. One study suggests that risk of gastrointestinal bleeding was significantly associated with acute use of non-steroidal anti-inflammatory drugs (NSAIDs) like regular-dose aspirin, diclofenac, ketorolac, naproxen or nimesulide. Piroxicam increased the risk of bleeding in both acute and chronic therapy.^[3] Opioids are the commonly used drugs for the management of acute postoperative pain. ^[4]

On the contrary many medicines of plant origin had been used since ages without any adverse effects. It is therefore essential that efforts should be made to introduce new medicinal plants to develop more effective and cheaper drugs. Plants represent a large natural source of useful compounds that might serve as lead for the development of novel drugs. ^[5] In the present study, *Eugenia jambolana* was selected because it is one of the medicinal plants commonly used in the ayurvedic system of medicine. *Eugenia jambolana* (*E. jambolana*), belonging to the family Myrtaceae. ^[6] It is also known in Hindi as jamun, jambo, jambul, jamhool, in English as black plum, purpleplum, black berry, nerudu in Telugu. ^[7] It has been reported to have hypoglycemic, neuropsychopharmacological, antibacterial, anti-HIV and antidiarrheal effects. The bark is astringent and is used in sore throats, bronchitis, asthma, ulcers and dysentery. The seeds are used in the treatment of diabetes. ^[8] The present study was conducted to evaluate the antiinflammatory and analgesic activities of the ethyl acetate extract of roots of *Eugenia jambolana*.

MATERIALS AND METHODS

Plant material: The roots of *Eugenia jambolana* used in this study were obtained from the Yamunanagar, Haryana and were identified based on its physical characteristics. The roots were dried and crushed to small pieces using pestle and mortar and powered in an electric grinder.

Preparation of Ethyl acetate Extracts: The shade dried roots were crushed into small pieces and

powdered. The powder was subjected to maceration for about 25–30 h with ethyl acetate. After maceration the solvent was distilled off and the extract was concentrated under reduced pressure on a water bath at a temperature below 50°C to a syrupy consistency. Then it was dried in the dessicator.

Selection of animals, caring and handling: In the present study wistar rats (150-200g) and albino mice (25-30g) of either sex were used. All the animals were procured from the disease-free animal house of CCS Haryana Agriculture University, Hisar, India. The animals had free access to food and drinking water as per CPCSEA dietary norms. They were subjected to natural light-dark cycle (12 hours each). The animals were acclimatized for at least 5 days to the laboratory conditions prior to experimentation. Experiments pertaining to this research work were carried out between 0900-1800h. The experimental protocol was approved by the Institutional Animal Ethics Committee wide its letter No-LSCP/2010/463 dated 13-04-2011. The care of the animals was taken as per the guidelines of CPCSEA, Ministry of Forests & Environment, Government of India.

Experimental Design: The ethyl acetate extract of the plant were assessed for anti-inflammatory and analgesic activities by using carrageenan induced paw edema in rats and acetic acid induced writhing in mice respectively.

Anti-inflammatory activity (Carrageenan induced paw edema in rats): Carrageenan-induced paw edema model was used for evaluating potential of ethyl acetate extract of the plant on inflammation. In this method, acute inflammation was produced by the administration of 0.1 mL of 1% (w/v) of carrageenan (Sigma Co.) in the sub plantar region of left hind paw of rat. The standard drug and extract were administered 30 min before the carrageenan injection. In this study diclofenac was taken as standard anti-inflammatory agent. ^[9] The experimental protocol comprises as follows:

Group 1- served as control group and was treated with vehicle alone in the sub plantar region of left hind paw of rats.

Group 2- was treated with diclofenac (50mg/kg; orally) used as standard drug for the evaluation of anti-inflammatory activity.

Groups 3 and 4- served as test group and were used for observing the influence of ethyl acetate extract (150 and 300mg/kg body weight) on carrageenan induced edema on rat hind paw.

The volume of the paw was measured immediately and also at the end of 2 hours and 4 hours after the Analgesic Activity (Acetic acid-induced writhing in *mice*): Acetic acid-induced writhing model was used for evaluating the potential of ethyl acetate extract of the plant on pain. In this method, pain was produced by the administration of 1% v/v of acetic acid (1mL/100g body weight of mice). The mice were placed in separate boxes under observation immediately after acetic acid injection and numbers of abdominal constrictions were counted over a period of 20 min. ^[11-12] The experimental protocol comprises as follows:

Group 1- served as control group and was treated with 2% w/v gum acacia solution (1mL/100g body weight of animals).

Group 2- was treated with diclofenac (25mg/kg, orally).

Groups 3- served as test group and were used for observing the influence of ethyl acetate extract (150mg/kg body weight) on 1% v/v of acetic acid-induced writhing in mice.

Statistical analysis: All the values were expressed as mean \pm SEM. The data was analyzed using Dunett t-test. In all tests, the criterion for statistical significance was p < 0.01.

RESULTS AND DISCUSSION

Anti-inflammatory Activity: The results of antiinflammatory activity after oral administration of *Eugenia jambolana* ethyl acetate extract are given in Table 1. Statistical analysis showed that the oral administration of *Eugenia jambolana* ethyl acetate extract has showed significant inhibition of carrageenan induced rat paw edema when compare control group at all the tested concentrations and the activity is dose-dependent. The results showed that the anti-inflammatory effect of the *Eugenia jambolana* ethyl acetate extract was similar to the effect of Diclofenac used as Standard.

Analgesic activity: The effects of oral administration of *Eugenia jambolana* ethyl acetate extract on acetic acid-induced writhing in mice are shown in Table 2. In the acetic acid –induced writhing in mice, the ethyl acetate extract at a dose of 150mg/kg body weight showed a significant reduction in pain in mice. The maximum percentage protection in writhing induced by acetic acid was 51.28% by ethyl acetate extract at 150mg/kg body weight. However, standard (Diclofenac, 25mg/kg body weight) showed highly significant inhibition at the same time.

On the basis of these findings, it may be inferred that ethyl acetate extract of Eugenia jambolana has analgesic and anti-inflammatory activities. These activities were related to the dose and these results corroborate the potential traditional use of the plant in folk medicine

CONCLUSION

From these overall results, we can conclude that the oral administration of Eugenia jambolana ethyl

possesses acetate extract significant antiinflammatory and analgesic effect, which can be useful for the treatment of acute pain and local inflammation.

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Table 1: Anti-inflammatory activity of the ethyl acetate extract of Eugenia jambolana roots and Diclofenac on carrageenan-induced edema in right hind-limb of rats

Drug/Extract Group Dose No. (mg/kg)			Increas	Increase in paw volume (mm)		inflammatory ctivity
			2hr	4hr	2hr	4hr
Control	Ι		1.04±0.12	1.02±0.04		
Diclofenac	II	50	$0.89{\pm}0.14^{**}$	$0.48{\pm}0.10^{**}$	34.67	54.38
Ethyl acetate	III	150	$0.78 \pm 0.06^{**}$	$0.56{\pm}0.08^{**}$	26.83	38.91
extract of root	IV	300	0.73±0.08**	$0.50{\pm}0.06^{**}$	32.18	52.80

Values of paw thickness are mean \pm SEM from 6 animals in each group.

Statistical analysis was done by dunett t-test

No.

p<0.05, p<0.01, p<0.01, p<0.001, compared with control: Dose = 50 mg/kg

Table 2: Analgesic activity of the ethyl acetate extract of Eugenia jambolana roots and Diclofenac on acetic acid-induced writhing in mice Drug/Extract Group Dose Mean no. of writhing±SEM %Protection (mg/kg)

Control	Ι		37.33±2.62		
Diclofenac	II	50	20.33±0.76**	55.76	
Ethyl acetate	III	150	24.50±2.20**	51.28	
extract of root					

Numbers of writhing are mean \pm SEM from 6 animals in each group.

Statistical analysis was done by dunett t-test

*p<0.05, **p<0.01, *** p<0.001, compared with control; Dose = 25 mg/kg

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