

## Research Article

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## Oral *Prosopis juliflora* treatment ameliorates inflammatory responses against carrageenan induced paw edema in rats

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### Abstract

*Prosopis juliflora* (Leguminosaceae) is a shrub which is traditionally used to treat, colds, diarrhea, dysentery, inflammation, itching, measles, sore throat, and wounds. The plant also possesses antiemetic, antibacterial, expectorant and antiseptic activities. In lieu of the same, the present study was designed to phytochemically screen and evaluate the anti-inflammatory potency of the ethanolic extract of leaves of *Prosopis juliflora* (100, 200, and 400 mg/kg) against carrageenan induced paw edema in rats. The preliminary phytochemical screening revealed the presence of flavonoids, saponins, carbohydrates, cardiac glycosides, tannins, and alkaloids in ethanolic extract of the leaves of *Prosopis juliflora*. The oral median lethal dose was found to be 3807.9 mg/kg in mice and > 5000 mg/kg in rats. Our result depicted that the extract significantly attenuated the paw edema with the highest activity observed at dose of 400 mg/kg. This study supports the folkloric claim of the use of *Prosopis juliflora* in the management of inflammations.

**Keywords:** *Prosopis juliflora*, Rat paw oedema, Carrageenan, Anti-inflammatory.

### Introduction

In Indo-Pak subcontinent *Prosopis juliflora* is called 'vilayati kikkar', 'kibuli kikkar', 'vilayati babul', 'vilayati khejra'. The history of the first introduction of *Prosopis juliflora* into India is about 130 years old. It is found especially in areas with 150-750 mm mean annual rainfall and maximum shade temperatures of 40-45°C. The domain of the species in arid and semi-arid tropical regions is mostly in plains and valleys but in many places it grows at altitudes of up to 1200 m above mean sea level.<sup>1,2</sup> *P. juliflora* is used to treat colds, diarrhea, dysentery, inflammation, itch, measles, sore throat, wounds and sexually transmitted diseases. The plant also possessed antiemetic, antibacterial, expectorant and antiseptic activities. Its juice is used in folk remedies for the cancerous condition.<sup>3</sup> Pain and inflammation are common complaints in many patients suffering from acute conditions.<sup>4, 5</sup> Anti-inflammatory agents inhibit the synthesis of prostaglandin synthesis which is one of the most important mediators of inflammation. Other mechanism of anti inflammatory activity the stabilization of lysosomal membrane in leucocytes (lysosomal enzymes destroy cartilage and other issues and perpetuate inflammation) and antagonism of certain actions of bradykinin. Accordingly, anti-inflammatory tests have to be divided into those measuring acute inflammation, sub acute inflammation and chronic repair processes. In some cases, the screening is directed to test compounds for local application.

Predominantly however, these studies are aimed to find new drugs against polyarthritis and other rheumatic diseases. Since the etiology of polyarthritis is considered to be largely immunological, special tests have been developed to investigate various immunological and allergic factors.<sup>6</sup> Paw oedema in rat, is based upon the ability of such agents to inhibit the edema produced in the right hind paw of the rat after injection of phlogistic agent. Many phlogistic agents (irritants) have been used, such as Brewer's yeast, formaldehyde, dextran, egg albumin, kaolin, aerosols sulphated polysaccharide like carrageenan or naphthoyiheparamine.<sup>7, 8</sup> The objective of this investigation was to ascertain the scientific basis of its use in treatment of inflammation, on which there is no previous data available. Hence, the present study was designed to investigate the anti-inflammatory activity of ethanolic extract of leaves of *Prosopis juliflora*.

## Material and methods

### Collection and identification of plant material

*Prosopis juliflora* was collected from nearby area of Bilara, district Jodhpur (Rajasthan). The taxonomic identification of the plant was done by, Botanical Survey of India, Arid Zone Circle, Jodhpur (Rajasthan) and a specimen is kept in the laboratory for future references. *Prosopis juliflora* leaves were removed from plants and were open dried in room temperature for 10 days. After drying, the dried leaves were grinded to a coarse powder with the help of suitable grinder. The powdered leaves were kept in airtight polythene bags and stored in cool and dark place to discourage deterioration by elevated temperature, light and moisture.

### Drug and chemicals

Carrageenan was procured from S.D Fine Chemical and other chemicals were purchased from local commercial sources and were of analytical grade.

### Preparation of extract

Dried plant material was abridged to a fine powder with a mechanical grinder. The powdered leaves were extracted with petroleum ether (60-80°C) and then defatted powder was refluxed with ethanol. The ethanolic extract of leaves of *Prosopis juliflora* (EELPJ) was concentrated to dryness and stored.

### Animals

Wistar rats of either sex, weighing 150-200 gm, were used for the study. All animal experimental at protocols were approved by Institutional Animal Ethics Committee (IAEC) of Lachoo Memorial College of Science and Technology, Jodhpur (Reg. No. 0541/02/C/CPCSCEA). The animals were fed with standard diet and water ad-libitum. They were housed in poly propylene cages maintained under standard conditions (12 hour light / 12 hour dark cycle; 22±2°C). The animals were deprived of food for 24 hour before experiment but allowed free access to drinking water throughout.

### Acute toxicity study

The acute toxicity study was performed as per the method described by Litchfield and Wilcoxon, and LD<sub>50</sub> was calculated accordingly. EELPJ in the dosage range of 100-2000 mg/kg was administered orally to different groups of rats (n=6). The animals were examined at every 1, 3 and 6 h. periods; finally 24 h mortality was recorded. All the animals were found to be safe. The dosing schedule was followed as per guideline 423. The anti-inflammatory activity was performed on experimental rats at three dose levels 100, 200 and 400 mg/kg. of body weight.<sup>9-11</sup>

### Induction of inflammation

The EELPJ was evaluated for its anti-inflammatory activity by carrageenan induced paw edema method. The experimental animals were divided into 5 groups (n=6). A mark was made on left hind paw of each rat just beyond tibio-tarsal junction. Group-I served as control and was administered 5% acacia solution in a volume of 1 ml/100 kg body weight, per oral. Group-II served as standard control and was administered Indomethacin (10 mg/kg body weight, orally). Group-III, IV & V served as test, was administered EELPJ in the doses of 100, 200 and 400 mg/kg body weight, orally. One hour after the oral administration of control, standard and test drug, 0.1 ml of 1% carrageenan in normal saline was injected into the plantar aponeurosis of the left hind paw of each rat. The volume of paw was measured by using a vernier caliper at in 1 and 3 hour after carrageenan suspension injection and the percentage increase in paw volume was compared with the increase in paw volume of animals of control group.<sup>12, 13</sup>

### Statistical evaluation

The results were expressed as mean (± SEM), and were compared to control by using one way analysis of variance (ANOVA) test, Followed by Dunnett's 't' test and a value

of at  $p \leq 0.05$  were considered to be significant. Percentage inhibition =  $1 - \left[ \frac{\text{paw diameter in treated}}{\text{paw diameter in control}} \right] \times 100$ .

### Results and discussion

Subplater injection of carrageenan in rats (control) showed time dependent significant increases in paw thickness when compared to normal and group. There were severely red and oedematous swelling of paw and this trend was gradually culminated in a deformed joint, swelling and oedematous paw (Figure 1).



**Figure 1:** Effect of *Prosopis juliflora* on joint of experimental rats

At 1 hr there is a significant ( $p \leq 0.05$ ) increase in paw volume and it sustained through the study when compare to the normal groups. At 1h, 3h there, is significant ( $p \leq 0.05$ ) increase in paw volume in Indomethacin, 100 mg/ kg and 200 mg/kg, 400 mg/kg of EELPJ. The percentage of inhibition of paw volume for 100 mg/kg is 8.5%, 12.8%,

for 200 mg/kg, 16.5%, 31.4%, for 400 mg/kg, 26.3%, 48.1% and for Indomethacin treated groups is 36.8%, 75.7%. Hence the % of inhibition of paw volume by EELPJ is significant ( $p < 0.05$ ) as compared with Indomethacin.

**Table 1:** Effect of *Prosopis juliflora* on rat paw diameter

S.No	Treatment	Paw Diameter (mm)			
		1 Hr		3Hr	
		Diameter	% Inhibition	Diameter	% Inhibition
1.	Vehicle	1.63±0.094	-	2.1±0.01026	-
2.	Carrageenan + EELPJ (100 mg/kg/p.o)	1.49±0.0796	8.5	1.83±0.1100	12.8
3.	Carrageenan + EELPJ (200 mg/kg/p.o)	1.36±0.0761*	16.5	1.44±0.0819*	31.4
4.	Carrageenan + EELPJ (400 mg/kg/p.o)	1.20±0.0592*	26.3	1.09±0.0882*	48.1
5.	Indomethacin (10mg/kg)	1.30±0.0884	36.8	0.51±0.0691	75.7

All values are expressed as mean  $\pm$  SEM, n=6. The minimum value of ( $p \leq 0.05$ ) was considered as significant. \*( $p \leq 0.05$ ) as compared with normal group. (EELPJ=Ethanollic extract of leaves of *Prosopis juliflora*).

We determined anti-inflammatory activity in by using inhibition of carrageen induced inflammation which one of the most feasible method to screen anti-inflammatory activity, the development of the first phase is attributed to the releases of histamine, serotonin and kinin and the second phase is related to the prostaglandin and bradykinin 14,15, 16, 17, 18, 19. We observed that EELPJ showed significant inhibition against carrageenan induced paw edema in the dose dependent manner to possess the anti-inflammatory effect may be due to presence of alkaloids. This response tendency of the extract in carrageenan induces rat paw edema revealed good peripheral anti-inflammatory properties of the EELPJ. Thus, it is concluded that the extract of EELPJ produced significant in dose dependent manner.

### Conclusion

The finding of the present study has demonstrated that ethanolic extract of *Prosopis juliflora* has potent anti-inflammation property and it justifies the traditional use of this plant in treatment of various types of inflammation. Further chronic inflammatory studies will be performed with specific anti-inflammatory and pro-inflammatory markers to elucidate the anti-inflammatory mechanism of action of this wonder plant

### Conflict of Interest

The authors declare no conflict of interest for this article.

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