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Anti-inflammatory, antipyretic and analgesic activities of ethanol extract of *Eugenia jambolana* Lam

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Abstract

The aim of present study is to determine anti-inflammatory, antipyretic and analgesic activities of ethanolic extract of *Eugenia jambolana* Lam. Plant of Eugenia is commonly used to treat different diseases like fever, pain and inflammation. A total of 144 albino rats with average weight of 200g were used in this study. Rats were divided into 4 groups each group contained 12 rats. Anti-inflammatory activity was evaluated by 0.1mL of 1% carrageenan extract that induce paw edema while analgesic activity was investigated by acetic acid that induce abdominal writhing response whereas, antipyretic activity was demonstrated by yeast that induce pyrexia in rats. In anti-inflammatory, antipyretic and analgesic activities, significant results were shown at 400mg/kg and 50mg/kg dose respectively in multiple comparisons. There was significant ($p<0.005$) decrease in inflammation, pyrexia and writhing. The stem extract of *E. jambolana* lam showed significant result as compare to leaf extract in anti-inflammatory and analgesic activity.

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Introduction

Eugenia jambolana Lam is commonly known as Jamun, Indian blue berry, java plum, Malabar plum, portuguese, doowet and faux pistachier etc. Eugenia leaf and stem are used for the maximum fall in acute inflammation (Parra *et al.*, 2019). A large evergreen tree mostly found in Indian subcontinent. Nowadays, tree of *Eugenia jambolana* Lam growing all over the Asian subcontinent, South America, Southeastern coast of Africa, Eastern Africa and naturalized Hawaii or Florida in the United State of America (Singh and Navneet, 2018). According to the medicinal system of Africa, species of Jamun are used for the treatment of multiple disorders (Sarkar *et al.*, 2017).

According to the World Health Organization (WHO), approximately three quarter of the population utilize herb in the form of ayurveda or traditional medicines to treat the diseases. Approximately 74% modern medicines are developed from pharmaceutical medicine (Verma *et al.,* 2018). As a result, it is said that the physiological action of Jamun that occur on human body is due to bioactive compound (Bayya *et al.*, 2018). Some bioactive compounds work as an agent in plants are responsible for the medicinal importance or activities. The remarkable active compounds are tannin, phenolic, flavonoid and alkaloids (Rafique *et al.*, 2018). Medicinal plant are free of side effects and less toxic (Bilal *et al.*, 2017).

The leaves of *E. jambolana* lam are 2 to 3 inches broad and 3 to 6 inches long. Phytochemicals in leaf extract of *E. jambolana* lam are myricitrin and have flavonol glycosides myricetin, betulinic-acid, n hepatcosane, maslinic etc. The phytochemicals present in stem extract of *E. jambolana* Lam are gallic acid, betulinic acid, beta sitosterol-D-glucoside, kaempferol, ellagic acid and myricetine etc. The astringent properties of stem bark are due to presence of gallo and ellagi-tannins (Kumawat *et al.*, 2018).

Stem of the tree of *E. jambolana* lam is very useful in digestive, anthelmintic, throat problem, spongy gums. It is used for the treatment of dyspepsia, diarrhea and dysentery. Dried seeds of *E. jambolana* lam are globally marketed for monotherapy.

The seed extract is effective against blood pressure, pharyngitis, splenopathy and ringworm infection (Vora *et al.*, 2018). The ethanolic extract of the leaf of *E. jambolana* lam is responsible for anti-inflammatory activities. Starch is converted in to energy with the help of leaves extract and maintain the blood sugar level in body (Bijauliya *et al.*, 2018). When extracts of *E. jambolana* lam were applied on animal models they show different kind of activities like anti-inflammatory, antipyretic diabetics and analgesic (Singh and Navneet, 2017).

The present study was designed to evaluate botanical uses of plant for the treatment of anti-inflammatory (inflammation), antipyretic (pyrexia) and analgesic (pain) activities in animal model.

Material and methods

Sample collection

The fully mature or fresh leaves and stem of *E. jambolana* Lam were directly collected as a sample from different botanical areas of Lahore. They were identified by botanist of University of the Punjab, Lahore, Pakistan.

Extract preparation

The stem and leaves were soaked in ethanol (semi polar solvent). All the mixtures were shaked gently for 2-3 minutes and placed for 15 days at room temperature (37°C). On 16th day, the mixtures were filtered with the help of whatsman filter paper.

Experimental rats

Albino rats of either female and male sex (160-200g) were purchased from University of Veterinary and Animal Sciences, Lahore. Rats were kept in animal house, University of Lahore in polypropylene cages. Before experimental work, rats were kept in fasting condition.

Drugs and chemicals

Normal saline, Diclofenac, *Eugenia jambolana* Lam leaf extract, Carrageenan, Yeast, Paracetamol, *Eugenia jambolana* Lam stem extract, Acetic acid and Distilled water.

Anti-inflammatory activity model

Group I: served as normal or control group in which rats were treated with normal saline.

Group II: served as standard in which rats were treated with diclofenac drug.

Group III: served as experimental design group in which rats were treated with aqueous extract of *E. jambolana* lam leaf.

Group IV: served as experimental design group in which rats were treated with *E. jambolana* lam stem.

Firstly, all groups of rats were treated with carrageenan in which 0.1 ml of 1% carrageenan injected into sub planter region of paw. Doses of carrageenan, leaf and stem extract are 50, 100, 200 and 400mg/kg respectively. The standard drug diclofenac 100mg/kg injected into rats. Anti-inflammatory activity was calculated by the given formula:

$$\% \text{ inhibition} = \frac{(Ct - Co)control - (Ct - Co)treated}{(Ct - Co)control} X 100$$

Where, C_o= Reading of paw before carrageenan, C_t = volume of hind paw of after carrageenan and (Ct-Co) = volume of hind paw of treated group after carrageenan injection.

Antipyretic activity model

In this model, rats were divided as discussed earlier in anti-inflammatory activity model. In standard group, rats were treated with paracetamol. In standard and experimental design groups, rats were treated with brewer's yeast with normal saline (mixture) which were injected below the nape of neck 50, 100, 200, 400mg/kg according to their body weight. After the interval of 20 hrs, pyrexia developed. The maximum rise in temperature was 38.3°C. Antipyretic activity was calculated by given formula:

$$\text{Percent reduction} = \frac{B - Cb}{B - A} X\ 100$$

Whereas, B = temperature after pyrexia induction, C_b =temperature after 1, 2 and 3 hour and A = normal body temperature.

Analgesic activity model

In this model, rats were divided as discussed earlier in anti-inflammatory activity model. Diclofenac used as standard drug. Acetic acid induce writing which was used to evaluate the potential of ethanolic extract of plant on pain. In standard and experimental design groups, all rats were treated with acetic acid 50, 100, 200 and 400mg/kg according to their body weight. The extract of leaf and stem were injected 1hr before, and standard drug diclofenac 12.5, 25, 50 and 100mg/kg were given 1/2hr before the administration of acetic acid. A number of abdominal constriction were counted in the period of 20 minutes. Analgesic activity was calculated by given formula:

$$\text{Analgesic activity} = \frac{Nc - Nt}{Nc} X\ 100$$

Where, Nc = control group writhe and Nt = treated group writhe.

Statistical analysis

Statistical analysis was performed by using ANOVA, the significance difference in activities were accepted (P<0.005) by multiple comparison. Whereas, P< 0.005 difference was considered to be significant.

Results

Carrageenan induce paw edema

In anti-inflammatory activity, the ethanolic extract of stem of *E. jambolana* lam at dose of 400mg/kg showed significant results (P<0.005) as compared to leaf of *E. jambolana* lam at doses of 50mg/kg, 100mg/kg, 200mg/kg and 400mg/kg. In inflammation assay, the maximum percentage inhibition of leaf showed 400mg/kg dose that is 96% as comparison to standard and control that was 100% (Table 1).

Antipyretic activity

In antipyretic activity, the ethanolic extract of stem and leaf extract of *E. Jambolana* lam showed results P>0.005, at doses of 50mg/kg, 100mg/kg, 200mg/kg and 400mg/kg as compared to standard drug (Table 2).

Analgesic activity

In analgesic activity, the ethanolic extract of stem of *E. jambolana* lam at dose of 400mg/kg showed significant results (P<0.005) in comparison with leaf of *E. jambolana* lam at doses of 50mg/kg, 100mg/kg, 200mg/kg and 400mg/kg. Dose of stem extract reduced abdominal writhing more significantly (P<0.005) in albino rats when compared to standard drug diclofenac (Table 3).

Table 1. Anti-inflammatory activity of *Eugenia jambolana* lam.

Group	Dose (mg/kg)	Paw size (mm) Mean ±S.D				% inhibition
		1hr	2hr	3hr	4hr	
Control	50mg/kg	1.450±0.636	1.200±0.424	0.710±0.975	0.005±0.007	0%
	100mg/kg	1.350±0.070	1.200±0.000	0.800±0.424	0.015±0.007	0%
	200mg/kg	1.300±0.000	1.050±0.070	0.800±0.282	0.300±0.282	0%
	400mg/kg	1.900±0.141	1.500±0.000	1.350±0.070	0.550±0.070	0%
Standard	50 mg/kg	1.500±0.000	1.350±0.070	0.600±0.141	0.400±0.141	76%
	100mg/kg	0.850±0.494	0.900±0.141	0.650±0.070	0.400±0.141	88%
	200mg/kg	1.600±0.565	1.050±0.070	0.850±0.212	0.550±0.070	90%
	400mg/kg	1.450±0.070	1.000±0.000	0.750±0.070	0.800±0.565	100%
Leaf	50 mg/kg	1.5000±0.000	1.350±0.070	0.600±0.141	0.400±0.141	76%
	100mg/kg	1.250±0.070	0.850±0.212	0.600+0.000	0.400+0.141	41%
	200mg/kg	1.250±0.070	1.050±0.070	0.900±0.000	0.900±0.282	81%
	400mg/kg	2.000±0.000	1.000±0.000	0.750±0.070	0.300±0.141	96%
Stem	50 mg/kg	1.300±0.141	1.100±0.000	1.000±0.000	0.750±0.212	76%
	100mg/kg	1.750±0.353	0.800±0.424	0.650±0.494	0.500±0.424	52%
	200mg/kg	1.650±0.212	1.500±0.141	1.100±0.141	0.800±0.282	83%
	400mg/kg	2.100±0.141	1.900±0.141	1.450±0.070	1.150±0.212	100%

Anti-inflammatory activity of *E. jambolana* lam leaf and stem

Values are mean ±S.D; n=4 in each group $P<0.005$ compare to control group.

Table 2. Antipyretic activity of *Eugenia jambolana* lam.

Group	Temperature	Dose (mg/kg)	Body temp (°c) Mean ±S.D				% inhibition
			0hr	0.5hr	1hr	2hr	
Control	37°C	50 mg/kg	37.50±0.14	37.35±0.07	37.20±0.00	37.05±0.07	0%
		100 mg/kg	37.65±0.07	37.45±0.07	37.20±0.00	37.00±0.00	0%
		200 mg/kg	37.75±0.07	37.55±0.21	37.35±0.21	37.15±0.07	0%
		400 mg/kg	37.90±0.00	37.75±0.07	37.55±0.07	37.25±0.07	0%
Standard	38.3°C	50 mg/kg	37.85±0.21	37.55±0.07	37.45±0.07	37.15±0.21	81%
		100 mg/kg	37.90±0.42	37.45±0.07	37.25±0.07	37.01±0.00	30%
		200 mg/kg	38.35±0.07	37.30±0.00	37.10±0.00	37.00±0.00	30%
		400 mg/kg	38.00±0.28	37.50±0.14	37.30±0.00	37.10±0.00	22%
Leaf	37.8°C	50 mg/kg	37.75±0.07	37.60±0.14	37.35±0.07	37.15±0.07	68%
		100 mg/kg	37.45±0.07	37.25±0.21	37.15±0.07	37.00±0.00	34%
		200 mg/kg	37.85±0.07	37.60±0.14	37.40±0.14	37.25±0.21	16%
		400 mg/kg	37.85±0.07	37.60±0.14	37.20±0.00	37.00±0.00	32%
Stem	37.7°C	50 mg/kg	37.75±0.07	37.70±0.28	37.30±0.14	37.05±0.07	63%
		100mg/kg	37.70±0.14	37.55±0.07	37.35±0.07	37.15±0.07	34%
		200 mg/kg	37.65±0.21	37.50±0.14	37.40±0.14	37.05±0.07	11%
		400 mg/kg	37.65±0.21	37.55±0.07	37.35±0.07	37.25±0.07	26%

Antipyretic activity of *E.jambolana* lam leaf and stem.

Values are mean ±S.D; n=4 in each group $P<0.005$ compare to control group.

Table 3. Analgesic activity of *Eugenia jambolana* lam.

Group	Dose (mg/kg)	Writhing Mean ±S.D	% inhibition
		20 (mints)	
Control	50 mg/kg	17.00±2.82	0%
	100 mg/kg	15.00±4.24	0%
	200 mg/kg	15.50±0.70	0%
	400 mg/kg	12.50±2.12	0%
Standard	50 mg/kg	13.50±2.12	20%
	100 mg/kg	13.50±0.70	10%
	200 mg/kg	17.00±1.41	9%
	400 mg/kg	18.50±0.70	48%
Leaf	50 mg/kg	11.00±1.41	68%
	100 mg/kg	16.00±1.41	34%
	200 mg/kg	16.00±2.82	16%
	400 mg/kg	18.5±0.70	32%
stem	50 mg/kg	11.50±2.12	63%
	100mg/kg	13.50±3.53	34%
	200 mg/kg	14.00±1.41	11%
	400 mg/kg	18.50±2.12	26%

Analgesic activity of *E. jambolana* lam leaf and stem

Values are mean ±S.D; n=4 in each group $P<0.005$ compare to control group.

Discussion

In our study, the ethanolic extract of *E. Jambolana* lam stem and leaf was used to evaluate anti-inflammatory, antipyretic and analgesic activities on albino rats. In analgesic activity, time difference occur due to time lag of drug entering into the body of rats (Parra *et al*., 2019). Anti-inflammatory activity induce edema in rat paw by carrageenan while antipyretic activity induce pyrexia in rats by brewer yeast where analgesic activity used to evaluate writhing test by acetic acid (Subedi *et al*., 2016). In inflammation process main symptoms are fever and pain (Deka *et al*., 2018). In our results, anti-inflammatory activity was dose dependent. Stem extract of *E. jambolana* lam showed similar effect as standard drug diclofenac for anti-inflammatory activity. Stem extract of *E. Jambolana* lam showed more significant results in anti-inflammatory activity as compared to leaf extract. When carrageenan injected into sub planter region of paw causes edema which is due to exudation of plasma protein, macrophages, leucocyte and neutrophils. Flavonoid are effective for acute inflammation because they target on prostaglandins that induce pain and edema (Safari *et al*., 2016).

When brewer's yeast is injected into nape of neck of albino rats, it induces pyrexia in body by increasing the synthesis of prostaglandins. Fever induced in body by brewer's yeast is called pathogenic fever etiologically cause the production of prostaglandins. Antipyretic action can be performed by synthesis of prostaglandins. Activity of cyclo-oxygenase enzyme is blocked by inhibition of prostaglandins and paracetamol action. Stem and leaves extract of *E. jambolana* lam injected intraperitoneally to reduce the fever (Muhammad *et al*., 2012). In our results, stem extract showed more significant results as compared to leaf extract and standard drug diclofenac in analgesic activity.

Abdominal and visceral pain in rats causes due to induction of acetic acid that lead pain in prostaglandins due to peritoneal fluid (Bairagi *et al*., 2017). Acetic acid is used as an agent for relief of endogenous substance responsible for pain which releases form nerve ending (Subedi *et al*., 2016).

According to our results, in analgesic activity, significant reduction in pain showed at the dose of 400mg/kg.

Conclusion

From above findings, the present study represent that ethanolic extract of *E. jambolana* lam used as a natural and safe remedy for the treatment of inflammation, acute pain, fever and writhing. From overall result, we have concluded that injection of *E. jambolana* lam extract in abdominal and intraperitoneal muscle possesses significant anti-inflammatory and analgesic activity due to phytoconstitute. In future prospective, secondary metabolites present in plant will help us to understand identification and isolation of compound that can be clinically used.

Reference

Bairagi SM, Inayat BP, Nitin N. 2017. Analgesic and anti-inflammatory activity of crude leaf and bark extract of *Lantana camara*. Marmara Pharmaceutical Journal **21(4),** 810-817.

Bayya MHRKG, Sachidananda AMN, Nagendra NIM, Asitava DR, Sampath KAK. 2018. Evaluation of reversal effect of *Eugenia jambolana* seed extracts against high fructose diet induced insulin resistance in albino rats. Asian Journal of Pharmaceutical and Clinical Research **11(12),** 572-576.

Bijauliya RK, Shashi A, Monika S, Dilip KC. 2018. *Syzygium cumini* (Linn.)- an overview on morphology, cultivation, traditional uses and pharmacology. International Journal of Pharmaceutical Science and Research **9(9),** 3608-3620.

Bilal R, Saba T, Maryam R, Waqas L, Saadia N. 2017. Comparison of Hepatoprotective activity of *Eugenia jambolana* fruit with *Punica granatum* fruit in diet induced hyperlipidemic rats. Journal of University Medical & Dental college **8(1),** 49-55.

Deka S, Ramakanta S, Mangala L. 2018. Analgesic anti-inflammatory and antipyretic activities of *Trichosanthes dioica* Roxb. Leaf extracts. International Journal of Pharmaceutical and Research **9(1),** 227-235.

Kumawat M, Jyoti D, Jaya K, Ayush KG, Chandan S. 2018. Pharmacological properties and therapeutic potential of *Syzygium cumini* (Jamun). World Journal of Pharmaceutical Research **7(3),** 312-322.

Muhammad N, Muhammad S, Haroon K. 2012. Antipyretic, analgesic and anti-inflammatory activity of Viola betonicifolia whole plant. BMC Complementary and Alternative Medicine **12(59),** 1-8.

Parra SA, Kalpesh G, Ashok D, Mohammad IR. 2019. Evaluation of anti-inflammatory and analgesic activity of methanolic extract of *Berberis lycium* Royle. Journal of Medicinal Plants Studies **7(1),** 39-45.

Rafique A, Inam-ur-Raheem M, Asghar A, Shafqat RM. 2018. Assessing the Therapeutic Potential of *Cinnamon zeylanicum* (Cinnamon) and *Syzygium cumini* (Jamun) Seeds on Hyperglycemia in Diabetic Rats. Journal of Nutrition Science Research **3(1),** 1-7.

Safari VZ, Kamau JK, Nthiga PM, Ngugi MP, Orinda G, Njagi EM. 2016. Antipyretic, Anti-inflammatory and Analgesic Activities of Aqueous Leaf Extract of *Aloe volkensii* in Albino Mice. Medicinal & Aromatic Plants **5(3),** 1-7.

Sarkar PK, Bishnoi SK, Shinde R. 2017. Under-Exploited Fruits of the Forests: Eastern Indian Plateau is a Potential Hub. Technology and Management Practices 261-278.

Singh A, Navneet. 2017. *Syzgium cumini* L.: its antimicrobal potential and pharmacological utilization in herbal drug. International Journal of Applied and Current Research **6(1),** 15-31**.**

Singh A, Navneet. 2018. Ethnobotanical uses, antimicrobial potential, pharmacological properties and phytochemistry of *Syzygium cumini* Linn syn. A review. International Journal of Innovative Pharmaceutical Sciences and Research **1(2),** 32-47.

Subedi NK, Rehman AMS, Mohammad AA. 2016. Analgesic and Antipyretic Activities of Methanol Extract and Its Fraction from the Root of *Schoenoplectus grossus*. Evidence-Based Complementary and Alternative Medicine 1-8.

Verma S, Madhu G, Harvinder P, Geeta A. 2018. Diabetes Mellitus Treatment using Herbal Drugs. International Journal of Phytomedicine **10(1),** 1-10.

Vora A, Varghese A, Kachwala Y, Bhaskar M, Laddha A, Jamal, Yadav P. 2018. *Eugenia jambolana* extract reduces the systemic exposure of aitagliptin and improves conditions associated with diabetes: A pharmacokinetic and pharmacodynamic herb drugs interaction study. Journal of Traditional and Complementary Medicine 1-8.