ANALGESIC AND ANTIPYRETIC ACTIVITY OF *ALBIZZIA LEBBECK*

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Summary

The alcoholic maceration extract from aerial parts of *Albizzia lebbeck* leaves (*Leguminosae*) was investigated for analgesic activity by tail flick model and anti-pyretic activity by Brewer's yeast-induced pyrexia in Wistar rats. The extract at 200 mg/kg and 400 mg/kg body weight doses was found to possess significant ($p<0.05$) and dose dependent analgesic and anti-pyretic activity in animal models. Further, the acute toxicity study with the extract showed no sign of toxicity up to a dose level of 2000-mg/kg. The potential to cause ulcer by extract was comparatively less than that of diclofenac sodium. Thus it could be concluded that *Albizzia lebbeck* alcoholic extract possess significant analgesic and anti-pyretic activity.

Key Words: Analgesic, Anti-Pyretic, *Albizzia lebbeck*

Introduction

Man has been using herbs and plants products for combating diseases since times immemorial. Indian systems of medicine have a deep root in our culture heritage and cater to the Medicare of large sections of our population. These systems mainly use herbs. If we dwell for a moment on our hoary past, the Rigveda, one of the oldest repositories of human knowledge, mentions the use of 67 plants for therapeutic use, the Yajurveda enlist 81 plants whereas the Atharveda written during 1200 BC describes 290 medicinal plants of medicinal value. Charak Samhita written during 990 BC describes 341 medicinal plants. The land mark in Ayurveda was Sushrut Samhita written during 600 BC mentioned 395 medicinal plants. Dhanwantari Nighantu mentions 750 medicinal plants, 450 are mentioned in the Bhavaprakash, 480 in Madanapala Nighantu and 450 in the Kaiyadeva Nighantu. India unquestionably occupies the top position in the use of herbal drugs. It is one of the foremost countries exporting plant drugs and their derivatives. It also excels in home consumption. It is not at all surprising that herbal drugs are so prevalent in India

given the great biodiversity and abundance of flora and the variety of geographical condition which allows the most exotic medicinal plants to be grown here. [1]

Albizzia lebbeck Benth. (Shirish, Family: *Leguminosae*) is a deciduous tree with compound leaves, flat oblong fruits, round cream colored seeds, grows wild. The plant is found throughout India, Bangladesh, tropical and subtropical Asia and Africa. [2] Barks are used in toothache and diseases of the gum. Decoction of the leaves and barks are protective against bronchial asthma and other allergic disorders. Barks and seeds are astringent and are given in piles and diarrhea. Ethanolic extract of pods possesses antiprotozoal, hypoglycemic and anticancer properties. The methanolic extract of the pod was investigated for antifertility activity. [3, 4] The plant extract also evaluated in allergic rhinitis [5] and memory and learning of mice. [6] Phytochemical investigations showed that the pod of the *Albizzia lebbeck* contains 3',5 Dihydroxy 4', 7 dimethoxy flavone, and N- Benzoyl L phenyl alaninol. [7] The beans of the plant contain albigenic acid-a new triterpenoid sapogenin. [8] The plant also contains saponins, [9, 10] macrocyclic alkaloids, [11] Tannins, [12] and flavonols. [13] The decoction of *Albizzia lebbeck* stem bark was found to be effective against bronchospasm induced by histaminic acid phosphate and shown to exert di-sodium chromoglycate like action on mast cells. [14] *Albizzia lebbeck* bark extract show the antimicrobial activity. The active constitute of bark extract is anthraquinone glycosides. The main constituent from bark is active against aerobes and mechanism of action is that glycosides cause the leakage of the cytoplasmic constituents. [15]

Two new tri-O-glycoside flavonols kaempferol and quercetin were identified from the leaves of *Albizzia lebbeck*. [16] Albizziahexoside a new hexaglycosylated saponin was isolated from leaves of *Albizzia lebbeck*. [17] Lignins Present in their cell walls have been oxidized with alkaline nitrobenzene. The phenolic acids were present in the range of 8.8-52.7 mg/g of cell wall. [18] The chloroform fraction of methanolic extract of *Albizzia lebbeck* leaves protected mice against maximal electroshocks. [19] Ethyl ether and alcoholic extracts of leaves of *Albizzia lebbeck* showed positive reaction against bacterial pathogens i.e. [*Staphylococcus aureus*] and [*Escherichia coli*] and fungal pathogen [*Candida albicans*]. Flavonoid contents like Quercetin and Kaempferol were isolated and identified form the leaves and Flavonoid was found contents (2.40 mg/g). [20] Methanolic extract of leaf and methanolic and water extracts of bark have shown in vitro mast cell stabilizing effect against compound 48/80. [21] The effect of saponin containing n-butanolic fraction (BF) extracted from dried leaves of *Albizzia lebbeck* on learning

and memory was studied in albino mice and significant improvement was observed in the retention ability of the normal and amnesic mice as compared to their respective controls. [22] Although *Albizzia lebbeck* leaves has traditionally been used in the treatment of many types of pain and fever conditions in India and no scientific report is available to date to validate these folkloric uses.

Materials and Methods

Plant Collection and Identification

The leaves of *Albizzia lebbeck* were collected from Jaipur in March 2009. A voucher specimen (Voucher No. RUBL 50033) was kept at the Department of Botany, University of Rajasthan after identification of the plant.

Extraction of Plant Material

Plant materials were washed with water and shade dried. The derided leaves were crushed to coarsely powdered by wood-grinder. The powdered material was defatted with petroleum ether (60-80 ºC) and then extracted with ethanol in the ratio of 1:10 of powdered drug and solvent by cold maceration method for 24 hrs. The extract was concentrated for further studies on water bath at 40 ºC.

Drugs and Chemicals

Diclofenac sodium (Voveran® Injection and Aspirin (Disprin® plus Tablets) were used in this study. Other chemicals used for extraction purpose were of laboratory grade.

Animals

Wistar rats of either sex of weighing between 100 – 150 gm of either sex were obtained from Seedling College, Jaipur National University, Jaipur. These animals were used for the acute toxicity, anti pyretic, analgesic activity. The animals were stabilized for 1 week. They were maintained in standard condition at room temp 60 ± 5% relative humidity and 12 h/12h light dark cycle. They had been given standard pellet diet and water ad-libitum throughout the course of the study. The animals were handled gently to avoid giving them too much stress, which could result in an increased adrenal out put.

Acute Toxicity Studies

The acute toxicity study was carried out in adult male albino rats by "fix dose" method of OECD (Organization for Economic Co-operation and Development) Guideline No. 420. Fixed dose method as in Annex 2d: Test procedure with a dose of 400 mg/Kg, 800 mg/Kg, and 2000 mg/Kg body weight was adopted. The animals were fasted overnight and next day extracts (suspended in 5% tween 80 solutions) were administered orally. Then the animals were observed continuously for three hours for general behavioral, neurological, autonomic profiles and then every 30 min. for next three hours and finally for mortality after 24 hours till 14 days. [23]

Pharmacological Studies

For the assessment of Anti pyretic, Analgesic activity, two dose level were chosen in such a way that, one dose was approximately one tenth of the maximum dose during acute toxicity studies and a high dose, which was twice that of one tenth dose (200 mg/kg, 400 mg/kg).rats were divided into 4 groups consisting of 6 animals each. Group I received vehicle, Group II, III, received per alcoholic extract of 200 and 400 mg/kg respectively whereas Group V served as positive control i.e. Diclofenac sodium (10 mg/kg, p. o.) and Aspirin (300 mg/kg, orally).

Analgesic Activity

The prescreened animals (reaction time: 6- 7 seconds) were divided into IV groups as described above. After administration of extract or vehicle or standard, the tail flick latency was assessed at 15, 30, 45, 60, and 120 minute by analgesiometer. The strength of current passing through naked nichrome wire was kept constant at 4 amps. The site of application of the radiant heat in the tail was maintained at 2.5 cm, measured from the root of the tail. The cut off time was fixed 15 seconds to avoid any tissue damage. [24, 25]

Antipyretic Activity

The antipyretic activity of plant extract was evaluated using Brewer's yeast-induced pyrexia in rats. Fever was induced by injecting 20 ml/kg (S.C.) of 20 per cent aqueous suspension of Brewer's yeast in normal saline below the nape of the neck and rectal temperature was recorded by digital thermometer immediately before (-18 h) and 18 h after (0 h) Brewer's yeast injection. After 30 min of treatment, all test drugs were given orally and the mean temperature is recorded at 15, 30, 45, 60, and 120 min after test, vehicle and drug administration per drug. The rectal temperature was measured at 1, 2, 3, 4 h. prior to the experiment, the rats were maintained in

separate cages for 7 days, and the animals with approximately constant rectal temperature were selected for the study. Aspirin (300 mg/kg) was used as standard drug for comparing the antipyretic action of plant extract. [26]

Statistical Analysis

The data were expressed as mean ± SEM. The data of analgesic activity and Antipyretic activity were analyzed by one way analysis of variance (ANOVA) followed by "Dunnett's test." P value less than 0.05 was considered as statistically significant.

Results

Acute Toxicity

Alcoholic extract did not show any toxicity and mortality up to maximum dose of 2000 mg/kg of body weight and weight of rat had a normal variation after 7 days of observations. Common side effects such as mild diarrhea, loss of weight and depression were not recorded.

Analgesic Activity

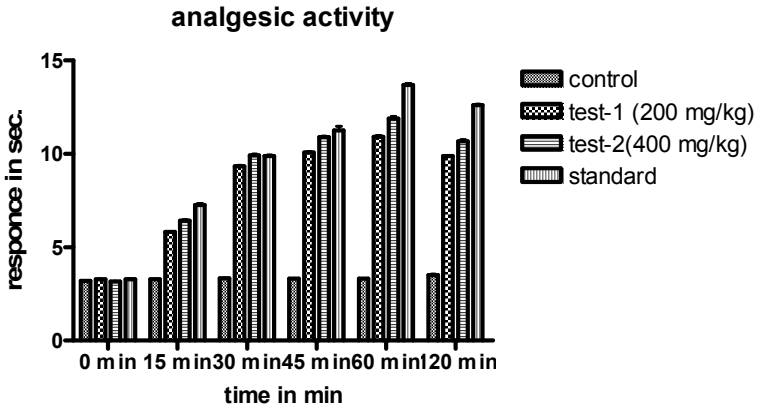
In radiant heat tail-flick the crude ethanolic extract produced 77.29% ($p<0.05$) and 95.80% ($p<0.05$) %elongation of Tail flicking time 15 minutes after oral doses of 200 and 400 mg/kg respectively (table 1, Graph 1). The crud extract is produced maximum analgesic effect 60 min. after oral doses 200 mg/kg and 400 mg/kg. The ethanolic maceration Extract with the oral doses of 200 mg/kg (test 1) and 400 mg/kg (test 2) show positive analgesic activity by tail flick method in albino rats method.

Table 1- Showing Effects of Extract on Radiant Heat Tail Flick Response in Rat

Time→ Group↓	Control	200 mg/kg	400 mg/kg	Standard
O min.	3.210 ± .005	3.290 ± .015	3.167 ± .008	3.280 ± 0.037
15 min.	3.286 ±.017	5.826 ± .020	6.417 ± .063	7.270 ± 0.090
30 min.	3.339 ± .020	9.330 ± .066	9.913 ± 0.063	9.898 ± 0.050
45 min.	3.313 ±.026	10.080 ± .040	10.893 ± 0.062	11.250 ± 0.231
60 min.	3.323 ± .026	10.923 ± .0592	11.900 ± 0.115	13.690 ± 0.091
120 min.	3.503 ±.049	9.897 ± .026	10.667 ± 0.084	12.600 ± 0.071

Values are means ±SEM (n=6) one way ANOVA, *p< 0.05 Compare to control

Graph 1: Analgesic Activity



Antipyretic Activity

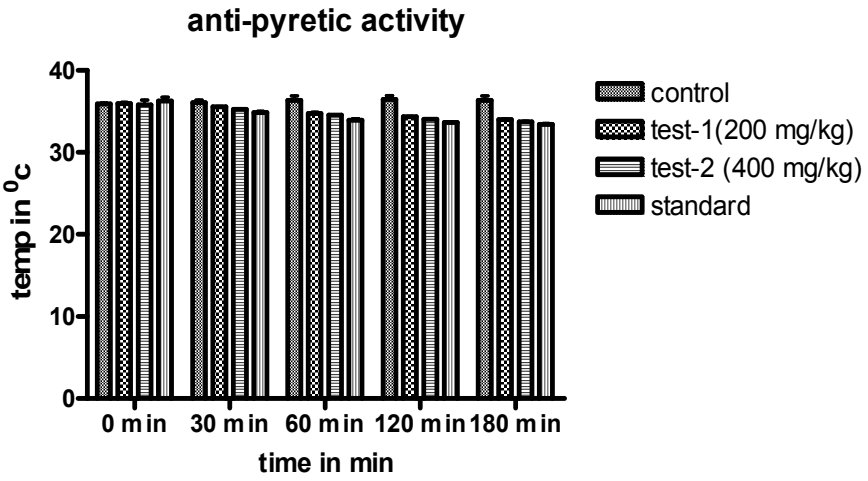
In Brewer's yeast-induced pyrexia method the crude Ethanolic extract reduced 6.46% ($p<0.05$) and 7.20% ($p<0.05$) of Temperature 180 minutes after oral doses of 200 and 400 mg/kg respectively (table 2, graph 2). The ethanolic maceration Extract with the oral doses of 200 (test 1) and 400 (test 2) mg/kg show positive antipyretic activity by Brewer's yeast-induced pyrexia in rats method.

Table 2- Showing Effects of Extract on Brewer's Yeast-Induced Pyrexia in Rats

Time→ Group↓	Control	200 mg/kg	400 mg/kg	Standard
O min.	35.90 ± 0.10	35.9 ± 0.15	35.80 ± 0.600	36.30 ± 0.40
30 min.	36.10 ± 0.30	35.55 ± 0.05	35.25 ± 0.050	34.85 ± 0.15
60 min.	36.35 ± 0.55	34.75 ± 0.15	34.55 ± 0.050	33.90 ± 0.20
120 min.	36.45 ± 0.45	34.35 ± 0.05	34.05 ± 0.050	33.65 ± 0.05
180 min.	36.35 ± 0.55	34.00 ± 0.10	33.73 ± 0.075	33.40 ± 0.10

Values are means ±SEM (n=6) one way ANOVA, *p< 0.05 Compare to control

Graph 2 - Antipyretic Activity



Discussion

The purpose of the present study was to establish scientific evidences for the usage of this plant in analgesic, antipyretic and antimicrobial conditions. The alcoholic maceration extract was studied for its modulatory effects on pain and fever induced by chemical and thermal stimuli. The analgesic effect of alcoholic extract was tested by tail flick model in rats whereas antipyretic effect was observed by Brewer's Yeast-Induced Pyrexia in Rats. Drugs that act centrally inhibit pain produced by thermal stimuli. [27] The alcoholic maceration extract produced analgesic effect in radiant heat tail-flick method. The crude ethanolic maceration extract was increased 77.29% ($p<0.05$) and 95.80% ($p<0.05$) %elongation of tail flicking time as compaire to control after 15 minutes oral doses of 200 and 400 mg/kg respectively. The crude extract is produced Maximum Analgesic effect 60 min. after oral doses 200 mg/kg and 400 mg/kg. against thermal induced pain stimuli in rats in tail flick method at various points indicates that it might be centrally acting. In the present study, diclofenac also inhibited the pain produced by tail flick method. Although, this model is specific for centrally inhibited pain, there are certain evidences that support; NSAID's also inhibit the pain induced by thermal stimuli. [28, 29] The observations from tail flick model suggest that alcoholic extract inhibited the pain thermal stimuli.

The alcoholic maceration extract produced antipyretic effect against pyrexia induced by Brewer's yeast in rats. the crude ethanolic extract reduced 6.46% ($p<0.05$) and 7.20% ($p<0.05$) of Temperature after 180 minutes oral doses of 200 and 400 mg/kg respectively in Brewer's

yeast-induced pyrexia method. In the present study, Asprin also produced antipyretic effect in Brewer's yeast induced pyrexia in rats produced by tail flick method. The observations from Brewer's yeast induced pyrexia in rats suggest that alcoholic extract inhibited the fever induced by chemical.

Conclusion

The results of present study revealed antipyretic and analgesic activity of alcoholic extract of *Albizzia lebbeck* leaves. Thus it substantiates the traditionally proven effectiveness of this plant in painful and fever conditions.

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References

1. Agarwal S.S. Paridhavi M. Herbal Drug Technology, Universities Press (India) Private Limited, Hyderabad, 2007.
2. Kirtikar KR and Basu BD. In Indian Medicinal Plants, India, 1980: 937.
3. Gupta RS, Kachhawa JB, Chaudhary R. Antifertility effects of methanolic pod extract of Albizia lebbeck benth. in male rats. Asian J. Androl 2004; 6(2):155-159.
4. Gupta RS, Chaudhary R, Yadav RK, Verma SK, Dobhal MP. Effect of Saponins of Albizia lebbeck benth. bark on the reproductive system of male albino rats. J. Ethnopharmacol 2005; 96(1-2):31-36.
5. Pratibha N, Saxena VS, Amit A, D'Souza P, Bagchi M, Bagchi D. Anti-inflammatory activities of Aller-7, a novel polyherbal formulation for allergic rhinitis. Int. J. Tissue. React. 2004; 26(1-2):43-51.
6. Chintawar SD, Somani RS, Kasture VS, Kasture SB. Nootropic activity of Albizia lebbeck in mice. J. Ethnopharmacol 2002; 81(3):299-305.
7. Rashid RB, Chowdhury R, Jabbar A, Hasan CM, Rashid MA. Constituents of Albizia lebbeck and antibacterial activity of an isolated flavone derivatives. Saudi Pharm. J. 2003; 11(1-2):52-6.
8. Barua AK, Raman SP. The constitution of albigenic acid-A new triterpenoid sapogenin from Albizia lebbeck Benth. Tetrahedron 1959; 7:19-23.
9. Ueda M, Tokunaga T, Okazaki M, Sata NU, Ueda K, Yamamura S. Albiziahexoside: a potential source of bioactive saponin from the leaves of Albizzia lebbeck. Nat. Prod. Res 2003; 17(5):329-335.
10. Pal BC, Achari B, Yoshikawa K, Arihara S. Saponins from Albizia lebbeck. Phytochemistry 1995; 38(5):1287-1291.

11. Misra LN, Dixit AK, Wagner H. N-demethyl budmunchiamines from Albizia lebbeck seeds. Phytochemistry 1995; 39(1):247-249.
12. Maa YT, Hsiaob SC, Chenb HF, Hsu FL. Tannins from Albizia lebbeck. Phytochemistry 1997; 46(8):1451-1452.
13. El-Mousallamy AMD. Leaf flavonoids of Albizzia lebbeck. Phytochemistry 1998; 48(4): 759-761.
14. Swamy GK, Bhattathiri PPN, Rao PV, Acharya NV, Bikshapathi T. Clinical evaluation of Sirisa Twak Kvatha in the management of Tamaka Shwasa[bronchial asthma]. Journal of Research in Ayurveda and Siddha 1997; 18:21-7.
15. Ganguli NB, Bhatt RM. Mode of action of active principles from stem bark of *Albizzia lebbeck.* Indian J Exp Biol 1993; 31:125-29.
16. El Mousallamy AM. Leaf flavanoids of *Albizzia lebbeck.* Phytochemistry 1998; 48:759-61.
17. Ueda M, Tokunaga T, Okazaki M, Sata NU, K, Yamamura S. Albizziahexoside; a hexaglycosylated saponin isolated from leaves of *Albizzia lebbeck.* Natural Product Research 2003; 17:29-35.
18. Negi AS, Karnani LK. Shakil NA. Phenolic acids asguaiacyl and coumaryl lignins in cell walls of forages and tree leaves. Indian Journal of Animal Nutrition 2000; 17:259-64.
19. Kasture VS, Chopde CT, Deshmukh VK. Anticonvulsive activity of Albizzia lebbeck, Hibiscus rosa sinesis and Butea monosperma in experimental animals. J. Ethanopharmacol 2000; 71:65-75.
20. Kapoor BBS, Bhumika, Khatri JS. Antimicrobial activity of some medicinal tree species of Hanumangarh district of Rajasthan. Journal of Phytological Research 2007; 20:325-326.
21. Shashidhara S, Bhandarkar AV, Deepak M. Comparative evaluation of successive extracts of leaf and stem bark of Albizzia lebbeck for mast cell stabilization activity. Fitoterapia 2008; 79:301-2.
22. Chintawar SD, Somani RS, Kasture VS, Kasture SB. Nootropic activity of Albizzia *lebbeck* in mice. Journal of Ethnopharmacology 2002; 81:299-305.
23. OECD, Guidance Document on Acute Oral Toxicity. Environmental Health and Safety Monograph Series on Testing and Assessment 2000;24:01-24
24. Barman S. Sahu N. Deka. S. Dutta S. Pharmacologyonline 2009; 1027-1034.
25. Gurav S. Gulkari V. Duragkara N. Sakharwade S. Analgesic and anti-inflammatory activity of flacourtia ramontchi L. Herit. Pharmacologyonline 2007; 2:20-31.
26. Hajare S. W. Chandra S. Tandan S. K. Sarma J. Analgesic and Antipyretic activities of Dalbergia sissoo leaves. Ind J Pharmacology 2000 ; 32:357-360
27. Janseen PAJ, Niemegeers CJE, Dony JGH. The inhibitory effects of fentanyl and other morphine like analgesics on the warm water induced tail withdrawal reflex in rats. Arzneimittel Forschung Drug Research 1963; 6:502-507.
28. Almasi R, Petho G, Bolcskei K, Szolcsanyi J. Effect of resiniferatoxin on the noxious heat threshold temperature in the rat: a novel heat allodynia model sensitive to analgesics. British Journal of Pharmacology 2003; 139:49-58.
29. Grace RF, Lin Y, Edwards SR, Power I, Mather LE. Effect of diclofenac in the rat tail ischemia – reperfusion injury model of acute hyperalgesia. Pain 2001; 89:117-25.