EVALUATION OF THE ANALGESIC AND ANTIPYRETIC ACTIONS OF THE *SARACA ASOCA* LEAVES IN EXPERIMENTAL ANIMAL MODELS.

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Summary

The bark of *Saraca asoca* (Caesalpiniaceae) has been commonly used in Indian medicine. It is used for menorrhagia, leucorrhoea, visceromegaly, dipsia, ulcers and fevers. In this study we evaluated the acute toxicity study, analgesic and antipyretic effects of ethyl acetate extract of *Saraca asoca* leaves. The results showed that the extract significantly inhibited the tail flick response of rats and increased the reaction time after dose administration. *Saraca asoca* cause a significant antipyretic effect in rats which was found to be independent of the route of administration of the drug.

Keywords: *Saraca asoca*, Analgesic activity, Antipyretic activity.

Introduction

Saraca asoca is an indigenous herbal drug belonging to the family Caesalpiniaceae. *Saraca asoca* is distributed in evergreen forests of India up to an elevation of about 750 meters. It is cultivated in many gardens because of its decorative orange red flowers and evergreen beautiful foliage. The leaves are parpinnate with 6 to 12 leaflets, oblong and rigidly sum-coriaceous. The fruit is a pod flat, leathery with 4 to 8 ellipsoid- oblong and compressed seeds. The whole plant is claimed to possess medicinal properties. For example, the bark of *Saraca asoca* is used by traditional healers for the treatment of menorrhagia, leucorrhoea, visceromegaly, dipsia, ulcers and fevers. [1, 2] The flowers also regarded medicinal property and used in diabetes, cancer and haemorrhagic dysentery. Seeds of *S. asoca* used for treating bone fracture, strangury and vesical calculi. Despite these, the antimicrobial property of1984; different parts of plant including bark, leaves, flower and flower buds are scientifically reported. [3, 4, 5, 6, 7, 8, 9, 10, 11] The Phytochemical study show in the bark of plant presence of (-) epicatechin, procyanidin p2,11'-deoxyprocyanidin B, (+) catechin, (24, £)- 24- methyl-cholesta-5-en-3p-ol (22 E, 21£)-24-ethycholesta-5, 22 dien-33-ol, (24£)-24-ethylcholesta-5-en-3-p-ol, leucopelargonidin-3-O-p-D glucoside, leucopelargonidin and leucocyanidin.The flower part of plant contain Oleic, linoleic, palmitic and stearic acids,P-sitosterol, quercetin, kaempferol- 3-0-P-D- glucoside, quercetin- 3-0-P-D-glucoside, apigenin- 7-0-p-D-glucoside, pelargonidin- 3, 5- diglucoside, cyanidin-3, 5- diglucoside, palmitic, stearic, linolenic, linoleic, p and y sitosterols, leucocyanidin and gallic acid.

Seed and Pod contains oleic, linoleic, palmitic and stearic acids, catechol, (-) epicatechol and leucocyanidin. Five lignan glycosides, lyoniside, nudiposide, 5-methoxy-9-β-xylopyranosyl-(−)-isolariciresinol, icariside E_3, and schizandriside, and three flavonoids, (−)-epicatechin, epiafzelechin-(4β→8)-epicatechin and procyanidin B_2, together with β-sitosterol glucoside, were isolated from dried bark. [12, 13, 14]
The work is aimed at investigating the analgesic and antipyretic action of the leaves extract of *S. asoca* as claimed by Indian traditional medicinal literature including Charaka Samhita and Susruta Samhita. [15, 16] This is with a view to giving adequate scientific backing and explanations to the use of *S. asoca* in the treatment of pain and fever.

Material and Methods

Collection and Authentication of Plant Material: The leaves of *S. asoca* were collected in February, 2009, from Mahaveer Nagar, Jaipur, Rajasthan, India. The species was authenticated by Mr. Vinod Sharma, Herbarium incharge, Department of botany, Rajasthan University, Jaipur. A voucher specimen (Voucher No. RUBL- 20532) was deposited in the herbarium of the Department of Botany, Rajasthan University, Rajasthan, India.

Preparation of Extracts: The leaves of the plant were air dried at room temperature for 15 days. The dried material was powered by means of an electric blender. Sixty grams of powdered leaves was extracted with ethyl acetate using a Soxhlet apparatus for 18 hours. A percentage yield of 7.8% was obtained after extraction and evaporation to dryness at room temperature. The ethyl acetate extract of *S. asoca* was administered as a suspension in 1% Tween 80 to the animals. [17]

Animals Used: Male Albino rats weighing 150-200 gm were obtained from School of Pharmaceutical Sciences, Jaipur National University. All the animals were housed in clean metabolic cages placed in well ventilated house conditions (temperature: 28 ± 1°C; photoperiod: 12 h natural and dark; humidity: 45 - 50%). The experimental protocols were approved by Institutional Animal Ethics Committee (IAEC No.-008/2009/IAEC/JNU) after scrutinization.

Acute Toxicity Study: Twenty Albino rats were divided into four groups of five rats each and were given graded doses (400, 800, 2000 and 5000 mg/kg body weight) of the extract by gavage using a incubation canula. The rats were observed individually for signs of toxicity and death once in first 30 min., after 24 hour and thereafter for a total of 14 days. The All four group received single oral dose of extract which prepared in Tween 80 solution. In observation including changes are; body weights of the rats before and after drug administration, onset of toxicity and sign of toxicity like change in skin and fur, eyes and mucous membrane and also respiratory, circulatory, autonomic and central nervous system and somatomotor activity, behavior pattern, sign of tremors, convulsions, salvation, lethargy, sleep and coma was noted. [18]

Analgesic Activity

Tail Flick Method: The Analgesic activity was tested by tail flick method in Albino rats. After overnight fasting healthy albino rats were divided four groups with six animals in each group. The tail flick latencies of the animals were assessed by Analgesiometer, basal reaction time was taken by placing the tip (last 2.5 cm) of the tail on the radiant heat source. Tail withdrawl from the heat (flicking response) was taken as the end point. The cut-off reaction time was fixed at 10 sec to avoid tissue damage. The mean reaction time was recorded at pre drug 15,30,60,90,120,150 and 180 min. after administration of vehicle or drugs by i.p. route. Pethidine (5 mg/kg) used as standard drug for comparing the analgesic action of plant extract. [19]

Antipyretic Activity: Male Albino rats weighing 150–200gm were injected subcutaneously (20 ml/kg) aqueous suspension of dried Brewer's Yeast (20%) Rats developing $1^{o}C$ or more rises in rectal temperature at 18^{th} hr after injection were treated with 0.9% normal saline and served as control. Group II & III received the extract (200mg/Kg) and (400mg/Kg) respectively. Group IV was treated with aspirin (300mg/Kg) and served as reference standard. Temperature was recorded at time intervals of 1, 2, 3 & 4 hours. [20]

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:

Ethyl Acetate extract did not show any toxicity and mortality up to maximum dose of 5000 mg/kg of body weight and weight of rat had a normal variation after 14 days of observations. Common side effects such as mild diarrhoea, loss of weight and depression were not recorded.

Analgesic Activity:

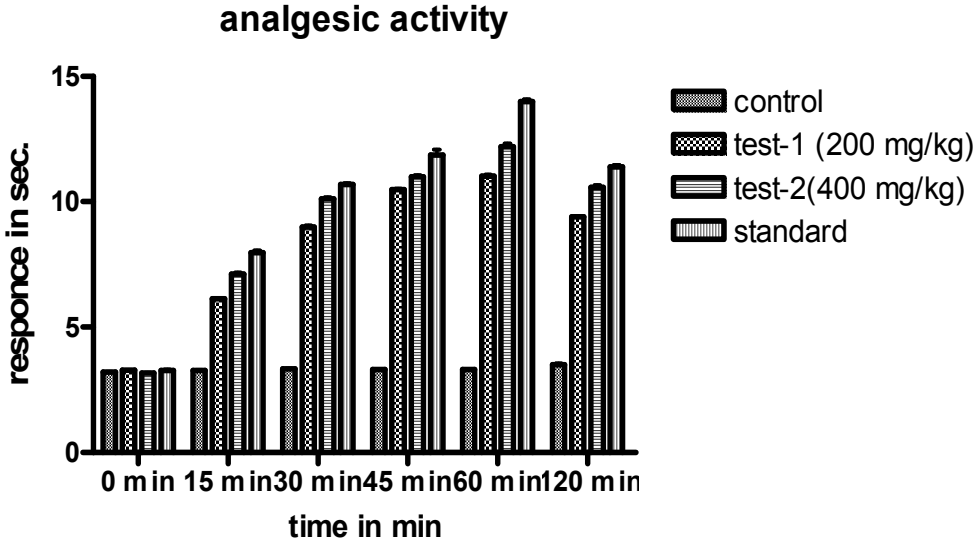
In radiant heat tail-flick the ethyl acetate extract against acute inflammatory pain was moderate as compared to potent inhibitory activity of Pethidine. In the present study, ethyl acetate extract (200 & 400 mg/kg) significantly increased the reaction time in tail flick method (Table 1, Graph 1). Ethyl acetate extract showed significant reduction in pain at 86.42% ($p<0.05$) 200mg/kg & 116.5% ($p<0.05$) 400mg/kg dose while standard drug showed reduction in pain 142% ($p<0.05$) at dose 5 mg/kg at 15 minute dosing time in radiant heat tail-flick method.

Table 1. Effect of leaf extract on tail flick nociception response in rats

Drug → Time ↓	Control	Extract 200 mg/kg	Extract 400 mg/kg	Pethidine 5 mg/kg
0 min.	3.210 ± 0.005	3.290 ± 0.015	3.167 ± 0.008	3.280 ± 0.037
15 min.	3.286 ± 0.017	6.126 ± 0.020	7.117 ± 0.063	7.970 ± 0.090
30 min.	3.339 ± 0.020	8.990 ± 0.066	10.113 ± 0.063	10.698 ± 0.050
45 min.	3.313 ± 0.026	10.480 ± 0.040	10.993 ± 0.040	11.850 ± 0.231
60 min.	3.323 ± 0.026	11.023 ± 0.059	12.200 ± 0.115	13.990 ± 0.091
120 min.	3.503 ± 0.049	9.397 ± 0.026	10.567 ± 0.084	11.400 ± 0.071

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

Table 1. Showing Effect of leaf extract on tail flick nociception response in rats.



Graph 1: Analgesic activity

Antipyretic Activity:

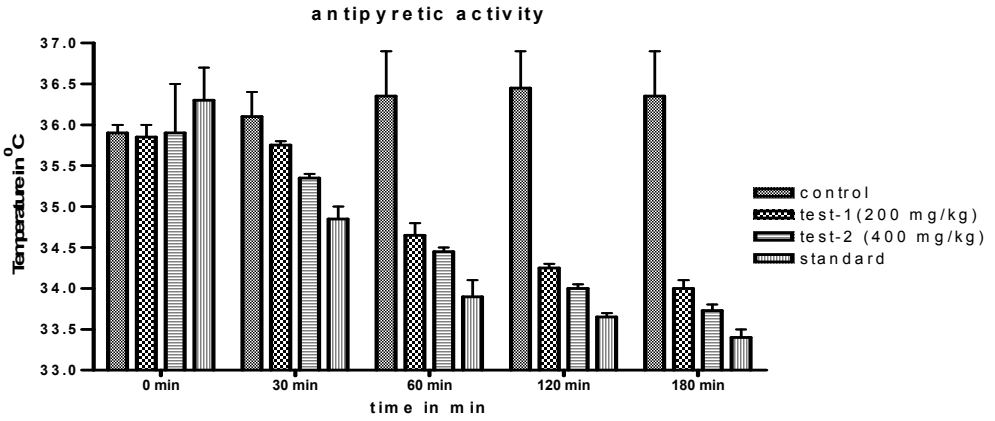
In Brewer's yeast-induced pyrexia method the crude ethyl acetate extract at doses 200 mg/kg & 400 mg/kg produced marked antipyretic activity in Brewer's yeast induced febrile rats (Table 2, Graph 2,). The ethyl acetate extract showed significant percentage reduction in pyrexia 6.46% ($p<0.05$) at 200mg/kg dose and 7.48% ($p<0.05$) at 400mg/kg dose while standard drug (300mg/kg) showed reduction in pyrexia 8.15% ($p<0.05$) at 180 minute dosing time in Brewer's yeast induced pyrexia method.

Table 2. Effect of leaf extract on brewer's yeast induced pyrexia in rats

Time→ Treatment ↓	**0 min.**	**30 min.**	**60 min.**	**120 min.**	**180 min.**
Control	35.90 ± 10	36.10 ± 30	36.35 ± 55	36.45 ± .45	36.35 ± 0.5
Extract 200 mg/kg	35.85 ± 15	35.75 ± .05	34.65 ± .15	34.25 ± 0.5	34.00 ± .10
Extract 400 mg/kg	35.90 ± 0.6	35.35 ± .05	34.45 ± .05	34.00 ± .05	33.73 ± .07
Aspirin 300 mg/kg	36.30 ± 0.4	34.85 ± .15	33.90 ± .20	33.65 ± .05	33.40 ± .10

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

Table 2. Showing Effect of leaf extract on brewer's yeast induced pyrexia in rats.



Graph 2: Antipyretic Activity

Discussion

The present study shows that *Saraca asoca* possesses analgesic activity on tail flick method. The tail flick model is an index that is used to evaluate acute pain in animals. Tail flick response is predominantly considered to be selective for centrally acting analgesic activity. The analgesic activity of *Saraca asoca* leaf extracts against acute inflammatory pain was compared to potent inhibitory activity of aspirin. Prostaglandin and bradykinin are important mediator in the pain process. Aspirin suppress the formation of pain substances in peripheral tissues and produced reduction in pain. Similar to *Saraca asoca* leaf extract suppress these substances and produces analgesic activity. [20, 21] From qualitative test presence of flavonoids in *Saraca asoca* leaves confirmed. Flavonoids inhibit the prostaglandin synthetase and prostaglandin show analgesic activity. In the present study, ethyl acetate extract (200 & 400 mg/kg) significantly increased the reaction time in tail flick method and shows significant analgesic activity. Similar to in antipyretic activity, fever may be as a result of infection or one of the sequelance of tissue damage, inflammation, graft rejection or other diseased states. Regulation of body temperature requires a delicate balance between the production and loss of heat. [22] Present study shows in Brewer's yeast-induced pyrexia method the crude ethyl acetate extract at doses 200 mg/kg & 400 mg/kg produced marked antipyretic activity in Brewer's yeast induced febrile rats. The reduction in the brewer's yeast induced fever by the extract in this study suggests some influence on the prostaglandin in biosynthesis since it is responsible to be regulating of body temperature. [23]

Conclusion

Above studies shows that *Saraca asoca* leaves extract shows significant analgesic and antipyretic activities**.**

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