

ANTIPYRETIC POTENTIAL OF EXTRACTS FROM SEEDS OF
BIXA ORELLANA L.

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Summary

Bixa orellana L. (Bixaceae), known as the annatto plant, is a small evergreen tree native of the Central and Southern American rain forests. It is commercially grown for the high apocarotenoid pigment content in its seed coat, particularly bixin (BXN) (90Z-6,60-diapocarotene-6,60-dioate). This species enjoys wide folkloric uses in India, China, the Philippines, Brazil and Guiana. This species has wide use in folk-medicine practice in these countries, especially as antioxidant and antipyretic. The present study aimed to evaluate the antipyretic activity of alcoholic and ethylacetate extracts of this species. The result reveals that the seed extracts (Ethylacetate 200 mg/kg) showed comparable antipyretic activity with that of the standard drug (Paracetamol 200 mg/kg). The present results provide a quantitative basis of explaining the traditional folk-medicine has significant antipyretic activity.

Keywords: *Bixa orellana* L., Bixaceae, ethyl acetate extracts, antipyretic activity

Introduction

Annatto, *Bixa orellana* L. (also known as 'achiote') is a perennial tree, native to the Neotropics ¹ and the only species in the Bixaceae family. Annatto is widely used for coloring in cosmetic and pharmaceutical products ². The plant is alexipharmic; useful in headaches, blood disorders, as an anti-emetic and to allay thirst. The seeds are cordial, astringent, febrifuge and a good remedy for gonorrhoea ³. The root bark is also useful in gonorrhoea ⁴ and as an antiperiodic and antipyretic ⁵. An infusion of the leaves and roots is useful in epilepsy, dysentery, fever and jaundice ⁶. Previous phytochemical investigations have revealed the presence of several carotenoid derivatives including bixin and norbixin ⁷, some terpenoids, tocotrienols, arenes and flavonoids (including luteolin and apigenin) in *Bixa orellana* seeds ⁸.

Investigations on *Bixa orellana* leaves have revealed the presence of flavonoid bisulfates⁹ and of an essential oil comprising mainly sesquiterpenes with ishwarane as the major compound¹⁰. The roots have been found to contain the triterpene tomentosic acid¹¹. Previous pharmacological studies have revealed that *Bixa orellana* extracts possess antiprotozoal, anthelmintic and platelet antiaggregant activity¹². Root extracts have been reported to have spasmolytic activity¹³. Extracts of leaves and branches have shown to be effective at neutralising the effects of snake venoms¹⁴. Extracts from different parts (leaves and seeds in particular) have displayed in vitro antimicrobial activity¹⁵. The seed extracts have been reported to exhibit chemopreventive¹⁶ and antioxidant activity¹⁷. Bixin has also been found to have anticlastogenic activity¹⁸. With a view to find the pharmacological rationale for some of the reported and traditional uses of the plant, the ethanolic and ethylacetate extracts of *Bixa orellana* L. seeds was evaluated for antipyretic activity.

Materials and methods

Collection of Plant material

Bixa orellana L. seeds were collected from Kollam, Kerala and identified by Dr. P. Jayaraman, Botanist, plant anatomy research centre (PARC) Chennai. The seeds were washed properly with water to remove the mud or dust; initially it was dried for seven days under shade. The dried seeds were then powdered by means of wood grinder and were sieved through sieve no.60 to get the coarse powder, which was subjected for ethanolic and ethylacetate extraction.

Animals

Healthy adult crossbred male albino rats (weighing 180–250 gm) was taken for the study. Food and water were provided *ad libitum*. Experiments were carried out between 0-3 hrs.

Drugs and chemicals

Paracetamol (Pfizer Ltd., Bangalore). Ethanol, Ethylacetate, Sodium carboxy methyl cellulose, sodium chloride, were purchased from SD Fine chemicals and are analytical grade.

Preparation of extracts

Dried and coarsely powdered seeds of *Bixa orellana* L. (250 g) was subjected to continuous extraction in a Soxhlet extractor for 18-24 hrs using 100% ethanol and ethylacetate as solvents. The solvents were recovered by vacuum distillation in a rotary vacuum evaporator (Buchler Corp.).

Antipyretic potential in rats

Rats were given subcutaneously 10 ml/kg of a 15% aqueous suspension of sterilized brewer's yeast powder. After 19 h, animals showing an increase of rectal temperature $> 0.50^{\circ}\text{C}$ were selected. Control group received normal saline; treated groups received 100 and 200 mg/kg of ethanol and ethylacetate extract of *Bixa orellana* L. seeds 1 to 5, respectively. The 6th group was given standard Paracetamol (200 mg/kg). Rectal temperature was determined by thermal-probe (Ellab thermistor thermometer) 30 min before (pre-treatment) and at 30 min., 1, 2 and 3 hrs after administration.

Statistical analysis

The data were analyzed for statistical significance, using the ANOVA.

Results

The antipyretic effect of *Bixa orellana* L. seed extract on yeast induced pyrexia is shown in Table 1. It was found that the subcutaneous injection of 10 ml kg⁻¹ of 15% w/v of yeast suspension markedly elevated the rectal temperature after 19 h of administration (39.6 °C) and treatment with *Bixa orellana* L. seed extract (100 and 200 mg kg⁻¹ doses) decreased the rectal temperature. The significant finding is the decreases of rectal temperature of the treated rats in a dose-dependent manner by *Bixa orellana* L. seed extract. The results obtained from both, the standard drug paracetamol (200 mg kg⁻¹) and *Bixa orellana* L. seed extract (100 and 200 mg kg⁻¹) treated groups, were compared with the control (5 ml kg⁻¹ sodium carboxy methyl cellulose suspension) group. Previous phytochemical investigations have revealed the presence of several carotenoid derivatives including bixin and norbixin, some terpenoids, tocotrienols, arenes and flavonoids (including luteolin and apigenin) in *Bixa orellana* L. seeds. Fever may be a result of infection or one of the sequelae of tissue damage, inflammation, graft rejection, or other disease states. Regulation of body temperature requires a delicate balance between the production and loss of heat, and the hypothalamus regulates the set point at which body temperature is maintained. In fever, this set point is elevated and drug like paracetamol do not influence body temperature when it is elevated by factors like exercise or increase in ambient temperature. The present results show that the ethylacetate extract of *Bixa orellana* L. seed at 200 mg kg⁻¹ dose possesses a significant anti-pyretic effect, in yeast-provoked elevation of body temperature (39.6 °C) in rats as the temperature was reduced to 38.0 °C, 37.9 °C and 37.4 °C respectively, and this effect is comparable to that of the standard anti-pyretic drug paracetamol which at 200 mg kg⁻¹ reduced the temperature to 37.6 °C. Furthermore, the *Bixa orellana* L. seed extract significantly reduced the normal body temperature of the animals tested, and to know the exact mechanism of action of the extract further study is required.

Table 1. Effect of *Bixa orellana* L. seed extract on elevated body temperature in rats.

Drug Treatment (mg kg ⁻¹)	Rectal temperature in (°C) before and after treatment				
	0 min	30 min	60 min	120 min	180 min
SCMC 5 ml kg ⁻¹	39.7 ± 0.02	39.7 ± 0.03	39.7 ± 0.02	39.7 ± 0.02	38.9 ± 0.01
Paracetamol 200	39.4 ± 0.02	39.1 ± 0.04	38.2 ± 0.01	37.8 ± 0.04	37.1 ± 0.03
Ethylacetate 100	39.9 ± 0.08	39.7 ± 0.06	39.5 ± 0.08	39.5 ± 0.06	38.9 ± 0.08
Ethylacetate 200	39.5 ± 0.07	39.1 ± 0.06	38.0 ± 0.05	37.9 ± 0.05	37.4 ± 0.05
Ethanol 100	39.1 ± 0.05	39.4 ± 0.09	39.4 ± 0.08	39.4 ± 0.06	38.7 ± 0.07
Ethanol 200	39.1 ± 0.06	39.1 ± 0.04	39.2 ± 0.06	39.1 ± 0.07	38.4 ± 0.07

Were” p” value is ≥ 0.05 considered as significant

Conclusion

The present results show that the ethylacetate extract of *Bixa orellana* L. seed at 200 mg kg⁻¹ dose possesses a significant anti-pyretic effect, in yeast-provoked elevation of body temperature (39.6 °C) in rats as the temperature was reduced to 38.0 °C, 37.9 °C and 37.4 °C respectively, and this effect is comparable to that of the standard anti-pyretic drug paracetamol which at 200 mg kg⁻¹ reduced the temperature to 37.6 °C. Furthermore, the *Bixa orellana* L. seed extract significantly reduced the normal body temperature of the animals tested, and to know the exact mechanism of action of the extract further study is required.

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