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PHARMACOLOGICAL STUDIES OF *ARAUCARIA BIDWILLI HOOK*

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ABSTRACT: *Leaf and oleoresin fractions of both alcoholic and petroleum ether extracts of Araucaria bidwillii exhibited significant prolongation of pentobarbitones sleeping time at a dose of 300 and 100 mg/kg body weight respectively in mice. This effect is more pronounced for oleoresin than the leaf extracts. Further, the two extracts seem to possess analgesic effect comparable to aspirin by writing method. The analgesic activity is found to be higher in oleoresin than the leaf extracts. Both the extracts are found to show high anti inflammatory activity comparable to phenylbutzaone by cotton pellet granuloma method.*

INTRODUCTION

A. bidwillii Hook (Araucariaceae) popularly termed as Monkey puzzle or Bunya-bunya tree is an important group of living gymnosperms dating back to Jurassic period of 195 million years ago having considerable economic and nutritive significance (Hora, 1981; Guha *et al*., 1971). This genus is commonly distributed in the hill stations of India such as Nilgiris and Kodaikanal. Many scholars are of the view that the origin of this genus-name "Araucaria" may have been derived from South American tribe and the species-name of the plant was named after the British Naturalist J.C. Bidwill (Shani, 1990).

Earlier workers have reported on the phytochemical constituents of this plant (Ilyas *et al.,* 1978; Caputo *et al.,*1976). Anderson (1972) reported use of *A.bidwilli* leaves by the Lahu tribal groups of Northern Thailand as a cure for insomnia in children. Therefore it is thought worth while in the present study of examine some pharmacological properties of this plant.

Materials and Methods

The fresh leaves and oleoresin of *A. bidwillii* have been collected from Government Botanical Garden, Udhagamandalam, Nilgiris (August 1991) and dried, grated and used for extraction. The dried materials are mechanically pounded separately to obtain a coarse powder which is then subjected to successive extraction in Soxhlet apparatus using petroleum ether and methanol solvents. The extracts thus obtained are concentrated at a temperature below 60^0C until semi solid mass is obtained. These are further dried under reduced pressure. The dried extracts are then subjected to pharmacological screening test.

Pentrobarbitone sleeping time has been determined in mice (30 – 40gm) as per the

method (Sheth *et al.*, 1972). Analgesic activity is evaluated in mice (35-40 gm) by writing method (Ghost, 1984). Anti-inflammatory activity is tested in rats (250-275 gm) by cotton pellet granuloma method (Kulkarni, 1987).

Dosage

The leaf extracts of both petroleum ether and methanolic fractions administered at a dose of 3000mg/kg body weight to different groups of mice fail to bring mortality. Hence, a dose of 300mg/kg of petroleum ether and methanolic extracts of *A.bidwillii* were used during the present study. Further, as the LD 50 of petroleum ether extract and methanolic extract of oleoresin was found to be 1g/kg, a dose of 100g/kg was used during the present study.

Experimental Design

Pentabarbitore Sleeping time in mice:

Petroleum ether and methanolic extracts of leaf and oleoresin of *A.bidwillii* are tested on pentobarbitone sleeping time (Sheth *et al.*, 1972). The animals were divided into six groups consisting of six mice each (30 – 40 gm) body weight. Group I received 300mg/kg petroleum ether extracts of leaves, respectively. Group II received 350mg/kg methanolic extracts of oleoresin, respectively. Group IV received 100mg/kg of metanolic extracts of oleoresin, respectively. Group V received pentobarbitone sodium at a dose of 35mg/kg in the same vehicle; refined groundnut oil was used as solvent for all the extracts. The route of administration was interperitoneal. The duration of sleep has been observed as the time between the loss and return of righting reflex. All animals are considered awake when it gives three positive responses within 30 seconds when tested for righting reflex.

Screening for Analgesic activity by writhing method in mice:

Albino mice weighing between 35 – 40gm.were divided into six groups of six animals each. The animals had been tested for writhing response a day before the actual test was performed. Group I & II received 300mg/kg petroleum ether and methanolic extracts of leaves, respectively. Group III & IV received 100gm/kg of petroleum ether and methanolic extracts of oleoresin, respectively. Group V received aspiring 100mg/kg. Group VI served as solvent control (refined groundnut oil). The route of administration was interaperitoneal. Half an hour after the administration of aspirin or plant extracts, the entire animal received 0.6% v/v aqueous acetic acid in a dose of 1 ml / 100g of body weight, interaperitioneally. The writhing response over a period of 20 minutes for each mouse was observed.

The mean number of writhing in different groups are tabulated in Table 2. Percentage protection compared to vehicle treated group was calculated using the following formula.

$$\text{Percentage protection} = 100 - \frac{\text{Observation in test group}}{\text{Observation in control group}} \times 100$$

Screening for Anti-inflammatory activity by cotton-pellet granuloma method in albino rat:

The chronic anti-inflammatory activity of both petroleum ether and methanolic extracts of leaves and oleoresin of *A.bidwillii* were studies in rats (250 – 275 gm). Under ether anesthesia, four sterilized cotton-pellets of 100mg were implanted in each groin by making an incision in the abdomen. The animals were divided into six groups of six animals each. Group I & II received 300mg/kg of petroleum ether and extracts of leaf, respectively. Group III & IV received 100mg/kg of petroleum ether and methanolic extracts of oleoresin, respectively. Group V received phenylbutazone 100mg/kg control group VI received refined groundnut oil. All the drugs were administered by interperitoneal route for seven days. On the eighth day the animals were sacrificed and pellets with granuloma tissues were dissected out, dried at 50^0C and weighed. Percentage protection was calculated using the formula as mentioned above.

Results and Discussion

A.bidwillii was studied in mice fore pentobarbitone sleeping time (Table-1). The results showed a significant increase in pentobarbitone sleeping time after oleoresin administration. The potentiating effect of pentobarbtone sleeping time was more with oleoresin than that of the leaf extracts. It may possibly justify the use of this plant by the Lahu tribals of Northern Thailand to treat insomnia as noticed by Anderson (1986).

The results of analgesic activity are presented in Table-2. It is clear from the data that both leaf and oleoresin extracts exhibit significant analgesic effect comparable to aspirin by writhing method. However, the analgesic activity was found to be more pronounced in oleoresin than the leaf extracts.

It is clear from the results on experimental inflammation that both the extracts show significant anti-inflammatory activity comparable to phenylbutazone (Table-3). The extent of anti-inflammatory activity exhibited by *A.bidwilliii* extracts was found to be more effective than the standard drug phenylbutazone, which however awaits further confirmation in different models through future studies.

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Table – 1
Effects of *A.bidwillii* extracts on Pentobarbitone sleeping time in mice.

S. No.	Drug Extracts	Dose in mg/kg	Mean sleeping time –SEM (min)
1	Petroleum ether extracts of leaf (Group I)	300	144.3 . 1.8*
2	Methanolic extract of leaf (Group II)	300	144.0 . 0.6*
3	Petroleum ether extract of oleoresin (Group III)	100	152.0 . 0.9*
4	Methanolic extract of oleoresin (Group IV)	100	155.8 . 0.5*
5	Pentobarbitone Control group	35	123.5 . 1.8

* $p < 0.001$ – compared with control group

Table – 2
Effects of *A.bidwillii* extracts on acetic acid induced writhing in mice.

S. No.	Drug Extracts	Dose in mg/kg	No. of Wriths mean . SEM	Percentage protection
1	Petroleum ether extracts of leaf (Group I)	300	33.2 . 1.0*	23%
2	Methanolic extract of leaf (Group II)	300	32.2 . 2.2*	26%
3	Petroleum ether extract of oleoresin (Group III)	100	16.0 . 1.4**	63%
4	Methanolic extract of oleoresin (Group IV)	100	19.5 . 1.3**	55%
5	Aspirin (Group V)	100	10.3 . 1.2**	76%
6	Control group	-	43.6 . 3.8	--

* $p < 0.05$ – compared with control group
** $p < 0.001$

$$\text{Percentage protection} = 100 - \frac{\text{Observation in test group}}{\text{Observation in control group}} \times 100$$

Table – 3
Effects of *A.bidwillii* extracts on Experimental inflammation (cotton – pellet granuloma method) in rats.

S. No.	Drug Extracts	Dose in mg/kg	Weight of Pelles (mg) mean . SEM	Percentage protection
1	Petroleum ether extracts of leaf (Group I)	300	28.25 . 1.85*	55%
2	Methanolic extract of leaf (Group II)	300	31.33 . 1.28*	50%
3	Petroleum ether extract of oleoresin (Group III)	100	29.25 . 1.35*	54%
4	Methanolic extract of oleoresin (Group IV)	100	32.25 . 0.82**	49%
5	Phenylbutazone	100	22.37 . 0.78*	65%
6	Refined groundnut oil	-	63.04 . 0.80	--

* $p < 0.05$ – compared with control group
** $p < 0.001$

$$\text{Percentage protection} = 100 - \frac{\text{Observation in test group}}{\text{Observation in control group}} \times 100$$

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