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## **Antimalarial Effect of Extracts of *Arisaema jacquemontii* Bl. On *Plasmodium berghei* Vincke and Lips (1948)**

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### **ABSTRACT**

Extracts of tuber, leaves and fruits from *Arisaema jacquemontii* Bl. (Himalayan or Jacquemont's cobra lily) were evaluated for their antimalarial properties against *Plasmodium berghei* (NK-65) maintained in white Swiss mice, *Mus musculus* (BALB/c). Peter's 4 day test was employed and animals were administered to ethanol prepared extracts of plant parts orally. Extracts significantly inhibited multiplication of *Plasmodium berghei*. Tuber extract inhibited about 70% propagation of parasite followed by leaves and fruit while no parasite was seen in the smears of chloroquine treated mice.

**Key words:** *Arisaema jacquemontii* Bl., *Plasmodium berghei*, *Mus musculus* (BALB/c), malaria parasite, antimalarial property

### **INTRODUCTION**

Malaria inflicts poor nations population in warmer area with millions of cases being reported annually (WHO, 2013). In spite of many efforts to control malaria, it still remains a major global health problem. There is currently no licensed vaccine available for malaria and only chemotherapy holds the promising alternative to combat this dreaded disease. Plants have invariably been a rich source for new drugs and some antimalarial drugs that are in use today were either obtained from the plants or developed using their chemical structures as templates (Gessler *et al.*, 1994).

*Arisaema jacquemontii* Bl. (Araceae) commonly known as Himalayan or Jacquemont's cobra lily, mainly found in Afghanistan, China, India, Nepal and Pakistan, has been used in this study to evaluate its antimalarial effect against *Plasmodium berghei* *in vivo*. *Arisaema jacquemontii* Bl. is fairly common in Himalayan forest at 2,300-4,300 m above the sea level. It also occurs in Nilgiri Hills in Southern India, Khasi hills of Northeast India and very common in Northern India. Compounds isolated from this plant exhibits anticancerous activity (Tanveer *et al.*, 2013). The lectins isolated from *Arisaema jacquemontii* Bl. tuber show insecticidal and antiproliferative properties (Kaur *et al.*, 2006).

### **MATERIALS AND METHODS**

Tuber, fruits and leaves of *Arisaema jacquemontii* Bl. were washed separately using distilled water and were air dried, weighed and cut into small pieces. They were homogenized in ethanol, homogenate filtered and centrifuged at 2000 rpm for 10 min (Sigma 3k-30). Supernatant was

boiled and the residual concentrated solid material was used as plant extract. Extract was suspended in Phosphate Buffered Saline (PBS), pH 7.2 before administration to animals. White Swiss mice, *Mus musculus* (BALB/c) was used to maintain *Plasmodium berghei* (NK-65). To maintain the parasite normal, mice were inoculated intraperitoneally (i.p) with  $1 \times 10^5$  *P. berghei* infected erythrocytes in citrate saline (0.85% sodium chloride w/v, 3.8% sodium citrate w/v). Smears from mice were prepared from tail vein, fixed in methanol and stained in Giemsa. Percentage infection (parasitaemia) was calculated by observing infected and normal erythrocytes in different fields on the slide as given:

$$\frac{\text{No. of } P. \text{berghei} \text{ infected erythrocytes}}{\text{Total No. of erythrocytes}} \times 100$$

For screening of plant extracts for their antimalarial activity, Peter's 4 day test was followed (Peters, 1970). On day 0, experimental as well as control groups of mice were inoculated intraperitoneally in each animal with  $1 \times 10^5$  *P. berghei* parasitized red blood cells.

The test extract was given daily from day 0-3 by oral route in concentration of  $500 \text{ mg kg}^{-1} \text{ b.wt. dose}^{-1} \text{ day}^{-1}$  to the experimental animals. Placebo controls received similar volume of PBS. Chloroquine was given to another control group of mice in concentration of  $4 \text{ mg kg}^{-1} \text{ mouse}^{-1} \text{ dose}^{-1}$  for 4 days as standard antimalarial.

On day 4, thin blood smears were prepared from the tail vein of all the animals (extracts treated, chloroquine control and placebo control groups) and the percentage infection was monitored. If any mouse died within 24 h of extract administration, this was considered as toxicity of the herbal product.

The study was carried out with prior permission of Institutional Animal Ethics Committee (IAEC) of H.P. University, Shimla vide number IAEC/Bio/1-2012.

## RESULTS

Extracts of tuber, fruits and leaves of *Arisaema jacquemontii* Bl. were used to evaluate their antimalarial effect. Test showed that none of the animals treated with extracts died during the 4 day test, showing the non-toxic effect of the dose of extract. Mice administered tuber extract showed mean parasitaemia  $6.18 \pm 1.69\%$  and percentage infection ranged between 3.70-7.70% on day 4 (Table 1, Fig. 1). In the fruits extract treated mice, mean parasitaemia was  $14.41 \pm 2.10\%$  and percentage infection ranged between 11.80-16.70% on day 4. While, in mice, treated with the leaves extract, mean parasitaemia being  $12.98 \pm 2.50\%$  and it ranged between 9.90 and 16.70% on day 4. In the placebo control mice, the mean parasitaemia was  $23.30 \pm 2.03\%$  and the animals died within 6-8 days post inoculation. In the mice given chloroquine ( $4 \text{ mg kg}^{-1} \text{ mouse}^{-1} \text{ day}^{-1} \text{ dose}^{-1}$ ),

Table 1: Percentage infection attained by mice on day 4 post inoculation

Experimental groups	Mice No. and parasitaemia (%)					Mean infection (%)
	1	2	3	4	5	
<b><i>Arisaema jacquemontii</i> Bl.</b>						
Tuber	3.70	5.20	7.70	6.90	7.40	6.18±1.69
Leaves	13.70	11.90	12.70	9.90	16.70	12.98±2.50
Fruits	12.70	14.90	16.70	11.80	15.98	14.41±2.10
Placebo control	20.98	26.21	24.23	21.98	23.12	23.30±2.03
Chloroquine control	0.00	0.00	0.00	0.00	0.00	0.000000

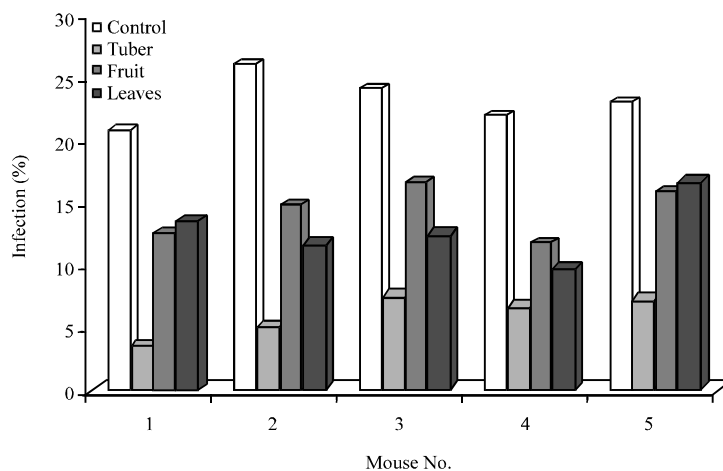


Fig. 1: Histogram showing the percentage infection in placebo control and extracts treated mice

Table 2: Maximum parasitaemia in mice after the administration of plant extract

<i>Arisaema jacquemontii</i> Bl.	Maximum parasitaemia (%)	Maximum parasitaemia on day	Death on day
<b>Tuber</b>			
1	20.04	06	12
2	52.70	10	13
3	43.70	08	11
4	53.85	09	12
5	29.03	07	13
<b>Leaves</b>			
1	44.80	07	9
2	53.70	09	10
3	51.70	08	9
4	52.70	09	10
5	53.09	09	11
<b>Fruit</b>			
1	43.90	08	10
2	51.78	09	10
3	53.72	09	10
4	41.70	08	11
5	50.78	09	11

no parasite was seen in the smears prepared on day 4. Different mice in each group treated with plant extracts showed maximum parasitaemia on a day as given in Table 2. The animals finally succumbed to infection.

## DISCUSSION

In the present study, *P. berghei* was found lethal to white Swiss mice and the animals died due to infection in a week's time. *Arisaema jacquemontii* Bl. is known for its medicinal properties. Rhizome ground with edible oil forms a paste used for massage purposes in order to regain the muscular strength and in skin problems such as blisters, pimples etc. (Khan, 2007). Fruit decoction is used as snake bite antidote. Roots of *Arisaema jacquemontii* Bl. led to the isolation of two new triterpenoids which were characterized by NMR, IR and MS spectra as 30-nor-lanost-5-ene-3 $\beta$ -ol and 30 norlanost-5-ene-3-ones (Jeelani *et al.*, 2010).

This study showed that various parts of this plant have antimalarial properties as the untreated control group showed mean parasitaemia  $23.30 \pm 2.03\%$ , which is quite high as compared to extracts fed group of mice. Of the three plant parts of *A. jacquemontii*, tuber exhibited more antimalarial effect *in vivo* but the molecules responsible for these properties and the exact mechanism is not known. Products derived from the plants like alkaloids and terpenoids have ability to develop drugs which needs to be explored further (Onguene *et al.*, 2013). So, the particular ingredients of this plant responsible for antimalarial activity may be extracted chemically, analyzed and biochemically studied to know its nature and hence its role in checking the propagation of malaria parasite infection.

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