



## Screening of antisnake venom activity of *Dichrostachys cinerea* W. & A.

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### Abstract

**Objective :** To study the antisnake venom effect of roots of *Dichrostachys cinerea* W. & A. **Materials and methods :** Antisanke venom property of *D. cinerea* was evaluated by using Russell's viper venom (*Daboia russelli*) and albino mice. Potency of methanolic and etherial extract was compared. **Results :** Methanolic extract of the roots of *Dichrostachys cinerea* has shown significant decrease in the mortality percentage in venom induced mice. **Conclusion:** *D. cinerea* has shown a positive effect of nullifying the viper venom toxicity in mice and can be recommended for further studies as an antidote to snakebite poisoning.

**Key words :** *Dichrostachys cinerea* W. & A., Antisnake venom activity, Russell's Viper venom.

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### 1. Introduction

Snake bite [1,2] poisoning has always remained a tremendous cause of morbidity in human beings, especially in the tropical and subtropical countries where adequate measures of treatment [3] are not easily available to the remote areas. Venomous snake bite also poses a great problem for the farmers venturing in the fields. Out of the species of venomous snakes all over the world, snake bite poisoning occurs in India with four prominent species viz. *naja naja* (cobra), *Bungarus caeruleus* (common and banded kraits), *Echis carinatus* and Russell's viper (true vipers).

Treating the snake bite victims with the help of herbal remedies [4,5] has been an age old concept. Though administration of intravenous antisnake venom serum is the only remedy available till today. Many such medicinal plants [6,7] are also reported previously for their antisnake venom properties in the classical literature.

Our ethnomedical survey has also revealed such plants which are consistently utilised by the tribals of Satpura range forests of the Jalgaon and Dhulia district for treating the victims of snake bite envenomation. Bhamre [8] has

reported such eleven plants used by the aboriginies of Dhulia district. It is always observed that most of the medicinal plants or plant parts are administered orally in the form of a decoction or its crushed pulp is applied on the bitten part.

The root of *Dichrostachys cinerea* [8,9,10] W. &A. locally called 'Hiver' is found throughout this region and is used for the treatment of many diseases as well as urinary and ophthalmic problems.

It is also used by the tribals of Satpura range forests viz. Kokni, Pawara, Vasave, Bhil for treating snake bite victims. This present investigation was conducted to investigate in vivo effectiveness of *D. cinerea* as an antidote to snake poison and its lethal effects.

## 2. Materials and methods

### 2.1 Plant material and extracts

Fresh roots of the plant *D. cinerea* W. & A. were obtained from the wild sources in the month of August and identified with the authentic sources. A voucher specimen has been deposited at the Department of Pharmacognosy, college of pharmacy, Chopda. The collected root was dried in the shade and crushed to a coarse powder.

Table1.  
Effect of the methanol extract on the lethality of *Daboia russelli* venom when administered to mice 1 min after LD<sub>50</sub> dose (47.5 µg / 20g) of the venom

Treatment	Dose mg/Kg	% Mortality Test
Vehicle	-	-
Methanolic extract of <i>D.cinerea</i>	50	37.55% ± 1
	100	31.25% ± 0.5*
	200	18.75% ± 1.5*

n=4; Each value is mean of two determination Mean ± SEM; \*P<0.05

The material was first extracted with Petroleum ether at 50-60°C in soxhlet apparatus for 72 h. The material was then extracted with methanol by refluxing at 60-80°C for 72 h in a soxhlet apparatus. Extract was then concentrated to a semisolid mass and was kept in refrigerator until further use.

Before using, the material was dissolved in normal saline, filtered and supernatant was used for further investigation. The methanolic extract was expressed in terms of dry weight (mg/ml) in normal saline.

### 2.2 Snake venom

Lyophilized snake venom of Russell's viper (*vipera russelli*) [11,12,13] was commercially collected from M/s Haffkine Institute, Mumbai (Batch no. 787 J) and preserved at 4°C until further use. The snake venom was dissolved in 0.90% w/v saline and centrifuged for 10 min. and supernatant was used as the venom. The venom concentration was expressed in terms of dry weight (mg/ml stock venom).

### 2.3 Animals

Swiss albino mice (18-20g) of either sex were used in this study. The animals were obtained from M/s National Toxicology Center, Pune and were kept in plastic cages at room temp with a 12 h. light / dark cycle. They had free access to drinking water and the standard laboratory feed (NTC Pune) *ad libitum*.

### 2.4 Phytochemical Screening

The Methanolic extract was subjected to the qualitative phytochemical analysis using standard methods [14].

### 2.5 Biological Methods

The antisnake venom activity of root extract of *D. cinerea* was determined by using the LD<sub>50</sub> of Russell's viper [12] venom in mice by the

subcutaneous route. The LD<sub>50</sub> of the venom was determined in swiss albino mice (18-20 g) 8 groups, four in each.

The venom was dissolved in 0.9 % saline before use and administered subcutaneously. The animals were observed for 2 h and then the number died within 24 h was recorded. The LD<sub>50</sub> was calculated by Karber's [15,16] method. The LD<sub>50</sub> of venom used was 47.5µg / 20g.

The effectiveness of the methanolic extract in modifying the lethal effect of the test dose of venom (47.5 µg / 20g) was investigated by administering the doses of different concentrations by the same route, one minute after the administration of snake venom. For this purpose each experiment was performed twice and each value is mean of such 2 determinations ± S.E. Four mice were used for each dose.

#### 2.6 Statistical analysis

The statistical analysis was carried out using the Students *t* - test and the results were judged significant if *p*<0.05.

### 3. Results and discussion

The LD<sub>50</sub> of venom was established 47.5 µg/ 20g and this was used as a test dose. The methanol extract of *Dichrostachys cinerea* W.&A. significantly protected mice against lethal effect of Russell's viper venom when administered 1 min after the venom (see table).

The best protection was obtained with 200 mg/kg. The etherial and aqueous extracts gave no such protection. In all the experiments controls were performed by administering the viper venom with only normal saline (0.9% w/v) and no significant reduction in the activity of venom is noticed.

Phytochemical analysis revealed the presence of glycosides in the methanolic extract. Further studies to isolate the active and other chemical constituents of *D.cinerea* are in progress.

### 4. Acknowledgement

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