

invitro Antilice activity of *Dichrostachys cinerea* (L.) Wight & Arn

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Abstract: Antilice activity of aqueous and ethanolic extract of *Dichrostachys cinerea* Wight & Arn (family: Mimosaceae) were studied. To provide scientific basis for the traditional claim it was compared with the marketed sample preparation. The ethanolic extract showed 98% mortality in 90 minutes. In order to reduce the mortality time, the ethanolic extract was mixed with carrier oil like Coconut oil, Castor oil and Gingelly oil. The extract with coconut oil as carrier oil showed significant reduction in mortality time from 90 minutes to 60 minutes and showed better activity when compared with castor oil and gingelly oil as carrier oil. From this result we suggest that *D. Cinerea* can be used for the herbal formulation as pediculicidal agent.

Key words: *Dichrostachys cinerea*, Antilice activity, Pediculicidal agent.

Introduction

The head lice *Pediculus humanus capitis* De Gear, is an ectoparasite confined to the scalp of hair of humans. Infestations are prevalent worldwide and especially common among school children in both developed and developing countries. *P. humanus capitis* infection cause skin irritation, prurities and sleep loss as well as occasional secondary bacterial infection. Although the symptoms are relatively mild, infestation by *P. humanus capitis* has resulted in various social, mental and economic problems. To control of human head lice they primarily depends on the synthetic Pediculocides like Organochlorine (DDT and lindane), Organophosphorus (Malathion), Carbamate (carbonyl), Pyrethrin, Pyrethroid (Permethrin) and avermectin (ivermectin- originated from streptomyces avermitilis) insecticides. But repeated usage of these synthetic pediculocides resulted in the development of resistance, increasing levels of resistance, caused multiple and excessive treatments fostering serious human health concerns. Plant essential oils have been suggested as an alternative source of materials for

insect control because they constitute a rich source of bioactive chemicals and are commonly used as fragrances¹.

Every year 6-12 million people in the United States are infested by head lice (Pediculosis). Despite evidence that infestation is not correlated with socioeconomic status, Pediculosis is a socially undesirable parasitic condition accompanied by social stigma, sleeplessness, prurities and last days in school. Head lice are considered a severe nuisance but few researchers have studied head louse biology since the first part of the last century. Research interest in head lice reemerged because report of resistance to tropical pediculocides has been documented around the world².

Dichrostachys cinerea (FAM :Mimosaceae) known as Vurtuli in Hindi, Vidattalai in Tamil, Velantarah in Sanskrit and Sickie bush in English³. The generic name 'Dichrostachys' means '2 coloured spikes. It is a much – branched thorny shrub sometimes a small tree up to 2m in height. It is distributed throughout the dry and warm parts of India. Brushed young shoots used in

the treatment of ophthalmia, astringent, rheumatism and urinary calculi, the leaves are used as fodder^{4,5}.

Materials and methods

Specimen collection

The plant specimens were collected from forest of Perambalur District, Tamil Nadu during August month and identified and authenticated by Botanist. A voucher specimen of leaves (PCG DC 005) has been deposited in the Department of Pharmacognosy herbarium Madurai medical college, Madurai. The leaves portions were washed thoroughly with water and dried in shadow. The shadow dried leaves were powdered, sieved using 60 meshes and stored in well closed container.

Preparation of ethanolic extract^{6,7}

The dried powdered leaves of *D.cinerea* was first defatted with petroleum ether 60°C - 80°C, and then extracted with Ethanol (99%) by using soxhlet apparatus for six hours. The obtained extract was concentrated under reduced pressure and stored in refrigerator until use.

Preparation of aqueous extract

The dried powdered leaves of *D.cinerea* were kept in a round bottomed flask fitted with a reflux condenser and extract with distilled water for six hours. The obtained extract was concentrated under reduced pressure and stored in refrigerator until use.

Phytochemical evaluation^{8,9}

Aqueous and alcoholic extract of *D.cinerea* were studied for its phyto constituents such as glycosides, proteins, terpenoids, tannins, saponins, aminoacids and carbohydrates by phytochemical testing. A portion of ethanolic extract was further fractionated by column chromatography (6x46cm) with the solvent of increasing order of polarity like hexane, ethyl acetate and isopropyl alcohol using silica gel (150) gm as

adsorbent. Twenty two fractions were collected, fractions were evaporated and dried under reduced pressure and identified the constituents by phytochemical evaluations.

Antilice Activity

Nowadays Filter paper assays in petridishes are common bioassay method to determine the level of topical insecticides resistance in head lice which provides informative and comparable results. Insecticide treated lice are likely able to continue blood feeding and blood ingestion alter the availability of the insecticide or the physiology of louse modify subsequent mortality¹⁰. So it was decided to feed head lice with blood meal by placing the lice on the bare lower leg of the one of the author after collection from the host. Microscopic examination was done in the mid gut region and confirmed the blood ingestion¹¹.

Colony of *P. humanus capitis* was collected by combing the hair of 20-25 infected children at the age group of 10-15. Adult lice were placed in small plastic containers (50ml polypropylene containers containing 1.5 cm human hair tufts. The mouth was covered with nylon mesh (1strand/mm) to permit ventilation. For this study the hair tufts was coated with test drugs. The mortality [mortality was defined as lack of limbs and gut and failure to respond when the legs were stroked with forceps.¹²] were observed at every 30 minutes. The activity study was carried out in triplicate at 37±1°C and 65±5% Relative Humidity in dark room, using water as control.

In the majority of situations, a carrier oil has sufficient suffocating capabilities to make almost any mixture of essential and carrier oil effective in killing lice and possible effectiveness of penetration using lipid based compounds.¹³ So the extract was mixed with the different carrier oils like coconut oil, castor oil and gingelly oil in the ratio of 20%, 40% and 60%, performed the antilice activity for the same.

Table No. 1: Anti lice activity of aqueous and ethanolic extract of *D. Cinerea*.

S.No.	Lice released	Test Drug	Concentration g/10ml	Mean ± SEM % mortality 60 min	Mean ± SEM % mortality 90 min
1.	N = 20	Aqueous paste	2	---	3.3333 ± 1.667
	N = 20		4	13.3333 ± 1.667	23.3333 ± 1.667
	N = 20		6	23.3333 ± 1.667	43.3333 ± 1.667
2.	N = 20	EEDC	2	3.3333 ± 1.667	16.666 ± 1.667
	N = 20		4	23.3333 ± 1.667	45.000 ± 2.887
	N = 20		6	61.6666 ± 2.887	98.333 ± 1.667
3.	N = 20	Standard marketed sample	10ml	98.666 ± 0.5774	---

Table No. 2: Anti lice activity of ethanolic extract of *D. Cinerea* with various carrier oil

S.No.	Lice released	Test Drug	Concentration g/10ml	Mean \pm SEM % mortality 60 min	Mean \pm SEM % mortality 90 min
1.	N = 20	EEDC + Coconut Oil	2	23.3333 \pm 1.667	47.61 \pm 1.002
	N = 20		4	41.6666 \pm 3.333	65.32 \pm 2.112
	N = 20		6	98.3333 \pm 1.667	---
2.	N = 20	EEDC + Gingelly oil	2	8.333 \pm 1.667	18.333 \pm 1.667
	N = 20		4	28.333 \pm 1.667	48.333 \pm 1.667
	N = 20		6	68.3333 \pm 1.667	98.333 \pm 1.667
3.	N = 20	EEDC + Castor oil	2	3.3333 \pm 1.667	21.666 \pm 1.667
	N = 20		4	23.3333 \pm 1.667	53.333 \pm 1.667
	N = 20		6	65.24 \pm 1.667	98.333 \pm 1.667
4.	N = 20	Standard marketed sample	10ml	98.666 \pm 0.5774	---

EEDC: Ethanolic extract of *D. Cinerea*

Result and Discussion

Lice infestations are common in worldwide whereas synthetic pediculocides are expensive. Though all of the synthetic pediculicidal agents act efficiently against *P.humanus capities*, some of them are neurotoxic. Hence Non toxic alternative options are needed for treatment of head lice. Natural products have been used in traditional medicine for thousands of years and recently have been of increasing interest. Natural source remain attractive primarily because they are inexpensive, less toxic when compare with synthetic products, readily available, capable of multitude of chemical modifications, potentially degradable and compatible due to their natural origin. We made an attempt to check the extract of leaves of *D.cinerea* for antilice activity.

The Phytochemical evaluation of aqueous and ethanolic extract of leaves of *D.cinerea* revealed that the various phytoconstituents like carbohydrates, proteins, saponins, glycosides, tannins, aminoacids and terpenoids were present in the extracts. Three different concentration of aqueous and alcoholic extracts were prepared and performed antilice activity and observed the mortality for 60 and 90 minutes. The results revealed that the aqueous extract and ethanolic extract

showed the mortality for 90 minutes were 43.33% and 98.33% respectively. The results were comparable with standard marketed sample; the results are shown in Table 1.

Three different concentrations of ethanolic extracts were prepared and mixed with carrier oil and performed Antilice activity, the mortality was observed for 60 and 90 minutes. The results revealed that significant mortality was found within 60 minutes in the case of ethanolic extract with coconut oil as carrier oil. The mortality time was reduced from 90 minutes to 60 minutes, the ethanolic extract of leaves of *D. Cinerea* mixed in coconut oil as carrier possesses significant antilice activity ($p < 0.05$). The results were comparable with standard marketed sample. The results are shown in Table 2.

Conclusion

From the above results we can conclude that the ethanolic extract of leaves of *D. cinerea* with coconut oil as carrier possesses significant antilice activity ($p < 0.05$). Advantages are inexpensive, less toxic, readily available, potentially degradable and compatible. We can suggest that *D. Cinerea* can be used in herbal formulation as pediculicidal agent.

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