

Testing the Insecticidal Activity of the Crude Extract S of Vitex Nugendo L Against Two Adult Insects Boophilus Microplus and Coptotermes Heimi



BIOSCIENCES

KEYWORDS: Vitex nugendo, Boophilus microplus (Ticks), Coptotermes heimi (Termites) insecticides

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ABSTRACT

In the study an investigation was made to study the effect of Vitex nugendo L an important medicinal plant against two adult insects Boophilus microplus (Ticks) and Coptotermes heimi (Termites). This initial study has shown that the ethanolic extract of the leaves of this plant does have a good potential for use against these two insect pests. The crude leaf extract has shown mortality percentages of 60% in the case of Ticks and 80% in the case of Termites after exposure to 1000 ppm of the extract for 48 hours, further studies with the purified extract can be carried out as a part of future investigation

INTRODUCTION

Synthetic pesticides have been reported to accumulate in the environment, i.e. seep or leach into ground water systems (Albanis and Hela, 1995; Cerejeira *et al.*, 2003; Hung and Thiemann, 2002; Matin *et al.*, 1998), enter into food webs and accumulate in body fat of fish, reptiles, birds and humans (Campbell and Campbell, 2001; Dong *et al.*, 2004; Hoshi *et al.*, 1998; Minh *et al.*, 2004; Tanabe *et al.*, 1998), where they disrupt physiological body functions and cause diseases like breast cancer (Alavanja *et al.*, 2004; and Mathur *et al.*, 2002). Pesticide accumulation has also been reported in the air where it can cause acid rain (Gong *et al.*, 2010) whereas methyl bromine and organochlorides have been reported to contribute to ozone layer depletion and global warming (Ristaino and Thomas, 1997; Thomas *et al.*, 2004). These disadvantages in mind, environmentalists have tried to lobby for policies that tend towards restricting pesticide use and at the same time look for biologically degradable insecticides, in this search plant based or bio insecticides have proven to a safe and cheap alternative. Neem and other plants have been used as insecticides for the past many years in different parts of the world, they are now accepted as safe alternatives by the scientific world in the present study investigations were carried to study the efficacy of another plant which has been used for the past many decades as an insecticide in certain parts of India. The present study was carried out using the extracts of *Vitex nugendo* against Termites and Ticks.

MATERIALS AND METHODS

A.) PLANT MATERIAL



FIGURE 1

BOTANICAL NAME: *Vitex nugendo* L

COMMON NAME: Nirgundi

FAMILY: Vaerbenaceae.

Vitex nugendo is an erect shrub or small tree growing from 2 to 8 m (6.6 to 26.2 ft) in height. The bark is reddish-brown. Its leaves are digitate, with five lanceolate leaflets, sometimes three.

Each leaflet is around 4 to 10 cm (1.6 to 3.9 in) in length, with the central leaflet being the largest and possessing a stalk. The leaf edges are toothed or serrated and the bottom surface is covered in hair. The numerous flowers are borne in panicles 10 to 20 cm (3.9 to 7.9 in) in length. Each is around 6 to 7 cm (2.4 to 2.8 in) long and is white to blue in color. The petals are of different lengths, with the middle lower lobe being the longest. Both the corolla and calyx are covered in dense hairs. The fruit is a succulent drupe, 4 mm (0.16 in) in diameter, rounded to egg-shaped. It is black or purple when ripe. (Fact Sheet 1999)

Geographical distribution: *Vitex* usually grows from three to nine feet tall, but under cultivation can develop to 20 feet tall. Nirgundi occur in tropical to temperate regions (up to 2200 m from east to west) grows gregariously in wastelands and is also widely used as a hedge-plant. This species is globally distributed in Indo-Malaysia, cultivated in America, Europe, Asia and West Indies. Within India, it is found throughout the greater part of India, ascending to an altitude of 1500 metres in the outer Himalayas. (Ladda and Magdum 20102).



B) TARGET INSECTS

I. Botanical Name: *Rhipicephalus microplus* Canestrini (Syn. *Boophilus microplus*).

Phylum: Arthropoda. **Family:** Ixodidae.

Common/local name: Tick; Pissu.

FIGURE: 2

Description: *Rhipicephalus microplus* is considered to be the most important tick parasite of livestock in the world. It is a hard tick that can be found on many hosts including cattle, buffalo, horses, donkeys, goats, sheep, deer, pigs, dogs and some wild animals. Heavy tick burdens on animals can decrease production of milk and may damage hides. *R. microplus* can also transmit babesiosis (caused by the protozoan parasites *Babesia bigemina* and *Babesia bovis*) and anaplasmosis (caused by *Anaplasma marginale*). Under experimental conditions, this tick can transmit *Babesia equi*, the cause of equine piroplasmosis.

Distribution: *B. microplus* is found worldwide in subtropical and

tropical regions. This tick is endemic in the Indian region, much of tropical and subtropical Asia, North-eastern Australia, Madagascar, South-eastern Africa, the Caribbean, and many countries in South and Central America and Mexico. (Johnson W and Wade F.S.(Retrieved July 25, 2013)).

3. Coptotermes heimi Wasmann.

Phylum: Arthropoda. **Family::** Rhinotermitidae

Common/local name: Termite (Eng);Seonk,Deemak



FIGURE: 3

Description: The insect is found worldwide in all the continents and is a serious pest of wood and wooden products. Termites are generally grouped according to their feeding behaviour. Thus, the commonly used general groupings are subterranean, soil-feeding, dry wood, damp wood, and grass-eating. Of these, subterranean and dry woods are primarily responsible for damage to human-made structures. (Su N-Y. 2003)

METHODS

Preparation of crude extract: The powdered plant part was heated with one polar (ethanol) and one non- polar (hexane) solvent so that the active principle dissolved completely in these solvents . Ethanol and hexane were used as the primary solvents. The extracts were prepared from each plant part – stem, root, leaf, and flower separately. After making the extract, the respective solvents were dried out in vacuum to give a solid crude ex-

RESULTS

TABLE1: MORTALITY PERCENTAGE OF TERMITES &TICKS EXPOSED TO 2000 PPM CONCENTRATION OF ETHANOL EXTRACT OF JUSTICIA ADATHODA

PLANT PART USED	MORTALITY PERCENTAGE AFTER 12 HOURS		MORTALITYPERCENTAGE AFTER 24 HOURS		MORTALITYPERCENTAGE AFTER 36 HOURS		MORTALITYPERCENTAGE AFTER 48 HOURS	
	TERMITES	TICKS	TERMITES	TICKS	TERMITES	TICKS	TERMITES	TICKS
STEM	10	10	10	10	10	10	10	10
LEAVES	10	20	20	20	20	20	20	20
ROOTS	10	10	10	10	10	10	10	10
FLOWER	10	10	10	10	10	10	10	10

The initial tests with the different parts of the plant showed that the Hexane extracts of all the four parts that is the stem, root, leaf and flower were not very effective in killing the target insects and the mortality percentage for the stem extract (Table2) was 10% at 200ppm for termites in all the experiments carried out and 20% for ticks at the same concentration only in the extracts derived from the leaves of the plant. The 2000 ppm ethanol extract of the stem caused a mortality of 10% and 20% in termites and ticks (Table1) after 12 hours, which increased to 70% in ticks and 50% in termites after 48 h . The leaves at 2000 ppm concentration showed a mortality percentage of 30% in case of termites and 40% in the case of ticks (Table 1). This mortality increased to 70% in the case of termites and 90% in the case of ticks after 48h.

TABLE2: MORTALITY PERCENTAGE OF TERMITES &TICKS EXPOSED TO 2000 PPM CONCENTRATION OF HAXENE EXTRACT OF JUSTICIA ADATHODA

PLANT PART USED	MORTALITY PERCENTAGE AFTER 12 HOURS		MORTALITYPERCENTAGE AFTER 24 HOURS		MORTALITYPERCENTAGE AFTER 36 HOURS		MORTALITYPERCENTAGE AFTER 48 HOURS	
	TERMITES	TICKS	TERMITES	TICKS	TERMITES	TICKS	TERMITES	TICKS
STEM	10	20	30	30	60	50	70	50
LEAVES	30	40	70	70	80	60	80	60
ROOTS	10	10	30	30	30	50	30	50
FLOWER	10	20	10	20	20	40	20	40

tract. During this process, temperature was maintained at a level (below 50 c) which does not harm the active principle dissolved in the solvents. A special high dosage of 2000ppm of each plant part was prepared in the two solvents and tested against the two target insects. The part whose extract gave the best results was further tested against the insects by exposing the insects to different doses of the plant extract. Different dosages of solution of the crude extracts were prepared using an emulsifying agent (Tween) and acetone / water as the solvent in concentrations of 100 ppm to 500 ppm. These dosages ranging from 100-1000 ppm were administered to the target insects to check their broad efficacy using standard procedures and precautions. The extracts were administered in triplicates and the average of all three readings was taken as the mortality. The readings were taken after 24 h and 48 h of administering the extracts to the target insects.

Ticks (*Rhipicephalus macropus*) (methodology of Prates, 1998):

Ticks were collected from one broad side of a cow or buffalo and stored in a bottle. One liter of the crude extract of the plant made in Acetone / water: tween mixture was immediately sprayed on the broad side of the animal from which the ticks were counted. Ten stored ticks were then released on to the broad side sprayed with extract. A count of dead ticks was then made after 12 h and 24 h respectively.

3.Termites (*Coptotermes heimi*) (methodology of Spikkett, 2002):

Termites were collected from the field and stored at 20 c. 100 gms of mud containing twigs and dried leaf parts was spread in Petri dishes and 10ml of the crude extract of the plant selected made in : Acetone/water mixture was poured into this and mixed properly. Ten termites were then released into this dish, which was subsequently covered by another Petri dish for 12 h and 24 h. After the stipulated time, the number of dead termites were counted.

The results with the root extract were 10% mortality in both termites and ticks after 12h and this percentage increased to 30% for termites and to 50% for ticks after 48h of exposure. The 2000 ppm flower extract gave an initial mortality of 10% for termites and 20% for ticks after 12h, this mortality increased to 20% in the case of termites and 40% in the case of ticks after 48h. In view of the above mentioned initial results it was decided to pursue the ethanolic extract of the plant and discard the hexane extract for this study. The next stage involved different concentrations of the

ethanolic extract for identifying the exact and effective concentration to be used against the target insects. (Table 3). Concen-

TABLE3: EFFECT OF DIFFERENT CONCENTRATIONS OF CRUDE ETHANOLIC LEAF EXTRACT ON THE TWO TARGET INSECTS (TERMITES AND TICKS)

Name of target insect	Mortality(%age)of insects in different concentration of crude Ethanolic Leaf extract											
	100 ppm		200 ppm		400ppm		600ppm		800ppm		1000ppm	
	After 24 h	After 48 h	After 24 h	After 48 h	After 24 h	After 48 h	After 24 h	After 48 h	After 24 h	After 48 h	After 24 h	After 48 h
Ticks	0	0	10	10	30	50	60	60	60	60	60	60
Termites	0	0	40	40	50	60	60	70	70	80	80	80

It remained at 60% in the case of termites in 600ppm after 24h but increased to 70% after 48 h, for ticks the mortality was 60% after 24h and 48h. In the 800 ppm extract the mortality for ticks was 60% after 24h and 48h whereas in termites it increased to 80% from 60% after 48h. At 1000ppm concentration the mortality was 60% after 24h and after 48h for Ticks and in case of termites the mortality remained at 80% after 24h and 48h.

DISCUSSION

Present investigation shows that the degree of contact toxicity of nishinda extract was different among different solvents as well. In general, the leaf extracts exhibited higher toxicity and mortality. As leaves are the prime attraction for the most of the phytophagous insects and other herbivores, plants accumulate their defence material in the leaves in greater concentration than other parts of their body (Harborne, 1988; Bell et al., 1990;

Houghton, 1996). Insecticidal activities of nishinda, *Vitex negundo* L. on red flour beetle (*Tribolium castaneum* Hbst.) was investigated by Chowdhury et al and they reported that among the solvents, methanolic extracts elucidated more toxicity on both adult and larval instars. (Chowdhury et. al.2009). Leaves of the plant are known to possess mosquito repellent activity for both *Culex* and *aedes* species (Raji 2013) similarly the plant has been shown to control the potato tuber moth with an LD 50 value of 485 ppm (Vishwanathan and Basavaraju 2010) similar results have been reported against Asian army worm, *Spodoptera litura* Fab with a n LD 50 value of 423 by Deepthi et.al. (2010). as also in the present study with the ethanolic extract of the leaves of the plant. This points to a good potential use of the plant leaves as an insecticide and the active principle can be identified by further purification for better trial results against similar insects.

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