Effect of clove oil(Syzygium aromaticum) and neem oil (Azedirecta indica) on *Tribolium castaneum* (Herbst) (coleopteran: tenebrionidae)

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Abstract

Tribolium castaneum (Herbst) (Coleoptera: Tenebrionidae) is one of the most damaging strored – product insecxt pest in India. Though synthetic insecticides have promising resulta against *T. castaneum* but use of synthetic insecticides can be hazardous for the environment. Further, while resistance development continues to be an issue for many synthetic pesticides, it is likely that resistance will develop more slowly to essential-oil-based pesticides owing to the complex mixtures of constituents that characterize many of these oils. Replacing these synthetic insecticides with plant materials to control this pest, however, can be a safe method with low environmental risk especially in stored products. So three important essential oils i.e. neem oil, clove oil were evaluated against *T. castaneum* Four different doses of each of the oils were prepared. Mortality percentages on different doses with respect to time were not only compared with each other. Results showed that mortality was directly proportional to dose and time in case of treated oils .

Key words: Neem oil, Clove oil, *Tribolium castaneum*.

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Introduction

Red flour beetle, Tribolium castaneum (Herbst) is one of the worldwide insect pests of mills, food warehouses, retail stores, and urban homes (Rees 2004). Scientifically it has been reported that the germ part (embryo portion) of the grain is destroyed by red flour beetle, T. castaneum. Their presence in stored grain directly affects both the quantity and quality of the commodity (1). *T. castaneum* is a serious secondary pest of stored cereals in tropical and subtropical regions of the world requiring prior infestation by an internal feeder, or some form of mechanical damage (Haines, 1991). Both adults and larvae are highly

mobile in patches of flour and broken gains (4). Ability of the females to lay more or less continuously throughout their life span result in high population in the substrate it colonizes (2)m .

In order to keep these stored grain products free from pest attack, various synthetic chemicals have been used.

Synthetic pesticides are currently the method of choice to protect stored grain from insect damage. But, continuous or heavy uses of synthetic pesticides has

Created serious problems arising from factors such as direct toxicity to parasites, predators, pollinators, fish and man. It also develops pesticides resistance susceptibility of crop plant to insect pests and increased environmental and social cost. Therefore, environment needs some other alternatives of chemical pesticides. One alternative to synthetic insecticides is the botanical pesticides i.e. insecticidal

plants or plant compound and the use of natural compounds, such as essential oils that result from secondary metabolism in plants. Essential oil and their constituents have been shown to be a potent source of

botanical pesticide. Plant essential oils have several advantages; low mammalian toxicity, low resides on grain, and novel chemical structures. Given that essential oils are very different in chemical structure than the currently used stored-product insecticides, we do not expect the insects that have resistance to the commonly used insecticides to also be resistant to the essential oils. Therefore, essential oils fulfill the requirements of pesticides in the 21st century and they may be widely used to control stored product pest. The essential oil from *Syzygium aromaticum* (clove oil) and neem oil (Azedirecta indica) possesses many compounds with biological activity, and it is used to control insects, fungus, mildews in stored grains.

Essential oils may act as fumigants , contact insecticides, antifeedants and may affect some biological parameters such as ovicidal activity, growth rate, life span and reproduction inhibition.

Therefore, this experiment have been designed to see the effect of clove oil and Neem oil on *Tribolium castaneum* .

Material and method

Plant material

The plants used were neem (*Azadirachta indica L.*) and clove (Syzygium aromaticum).

NEEM

The neem tree (*Azadirachta indica L.*) is a tropical evergreen plant with wide adaptability and known resistance to insect infestation. The tree is known to possess some compounds such as *limonoids*, which gives it a bitter taste while the principal bioactive content, *Azadirachtin*, is a repellent and anti-feedant to many insects.

CLOVE OIL

Syzygium aromaticum (L.) Merr and Perry commonly called clove, which belongs to the family *Myrtaceae*, is an important aromatic spice. The main components of essential oil were eugenol, eucalyptol ,caryophyllene which show insecticidal properties.

Material used

Insect culture

The insects were collected from the infected drugs and were cultured in flour at room temperature in glass beakers.

Essential oil

Essential oil was collected from locally available medical store.

Treatment of insects

Serial dilution essential oils were prepared using acetone as a solvent. considering azadirectine as active ingredient four doses i.e. 10%, 15%, 20%, 25% of neem oil and clove oil were prepared along with control having 0% of oil. Four sets of petri–dishes as replications of each concentration were made for both oils. Labeling of petri–dishes were done at 1 to 4 (1 to 4; starting from 1 for highest concentration and ending on 4 for lowest concentration of neem seed oil and number 5 for control). Aliquots of 2 ml of the dilutions were thoroughly mixed with 2 grams of flour with the help of glass rod in the petri-dish. The solvent was allowed to evaporate for 1 hour and five adult insects were released to each petri–dish. Whereas, controls were treated only with acetone alone. Three replicate of each concentration were made. 15 adults of *T. castaneum* were used for each concentration and same quantity was used for control. The petri–dishes were kept at room temperature and mortality was observed after 12, 24, 36 and 48 hours of exposure. Mortality percentage was recorded after 12, 24, 36 and 48 hours of exposure. The comparison of essential oils was done.

RESULTS

The essential oil showed variable toxicity to adult T. castaneum. The colve oil revealed 100% mortality at the higher concentration (25) after 48h of treatment . the neem oil revealed only 86.9% mortality at higher dose(25) after a period of 48 hour treatment . according to figure 1 and 2 as the concentration and treatment time increases mortality also increases. Therefore, mortality is directly proportional to concentration and time exposed.

Table 1: Toxicity of two essential oil against Tribolium castaneum at different time interval and different concentration

Essential oil	**Different concentration**	**Replicate**	**mortality at different time period**			
			12	24	36	48
Clove oil	10	control	0	0	0	0
		1	0	1	1	2
		2	0	1	1	3
		3	0	1	2	2
	15	control	0	0	0	0
		1	0	2	2	3

		2	0	1	2	2
		3	0	0	1	3
	20	control	0	0	0	0
		1	1	2	3	3
		2	2	2	3	5
		3	1	2	2	4
	25	control	0	0	0	0
		1	1	3	4	5
		2	1	2	3	5
		3	2	3	4	4
Neem oil	10	control	0	0	0	0
		1	0	0	0	0
		2	0	0	1	2
		3	0	0	1	1
	15	control	0	0	0	0
		1	0	1	1	2
		2	0	1	1	1
		3	1	1	2	2
	20	control	0	0	0	0
		1	0	1	2	2
		2	1	2	2	3
		3	1	2	3	3
	25	control	0	0	0	0
		1	1	2	3	4
		2	1	2	3	4
		3	2	3	4	5

Table 2:- Effect of different plant extracts concentrations on mortality of Tribolium castaneum adult at 12, 24 and 48 hours after treatment

	12					24					36					48				
concentraion	con	10	15	20	25	con	10	15	20	25	con	10	15	20	25	con	10	15	20	25
Clove oil	00	0	0	1.33	1.33	00	1	1	2	2.66	00	1	1.66	2.66	3.66	00	2.33	2.66	4	5
Neem oil	00	0	0.33	0.66	1.3	00	0	1	1.66	2.33	00	0.66	2.6	2.3	3.3	00	1	1.66	2.66	4.3

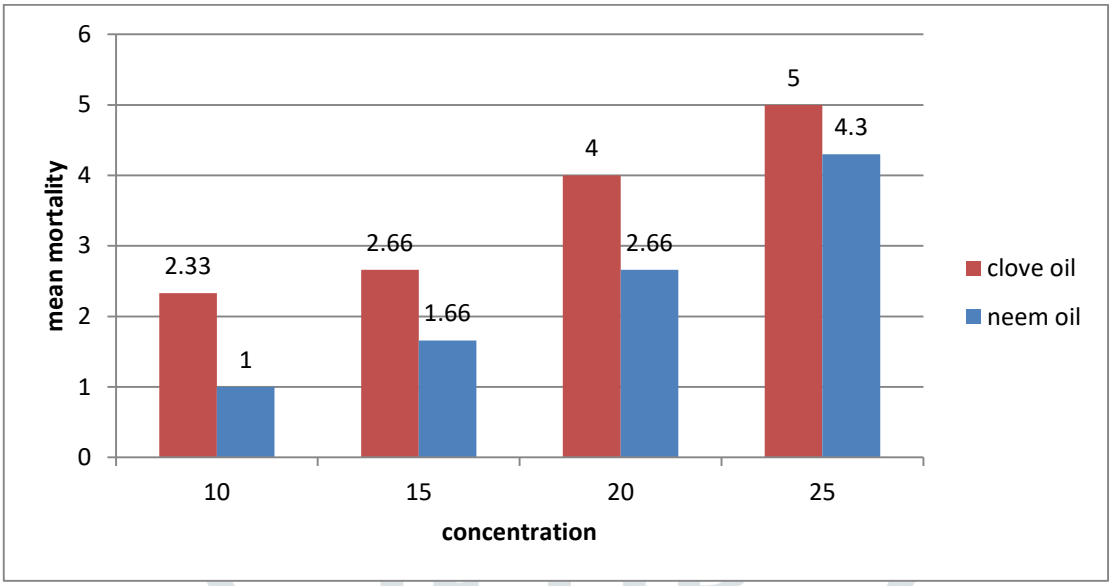

Fig1:- Toxicity of different essential oil concentration on the Tribolium cataneum

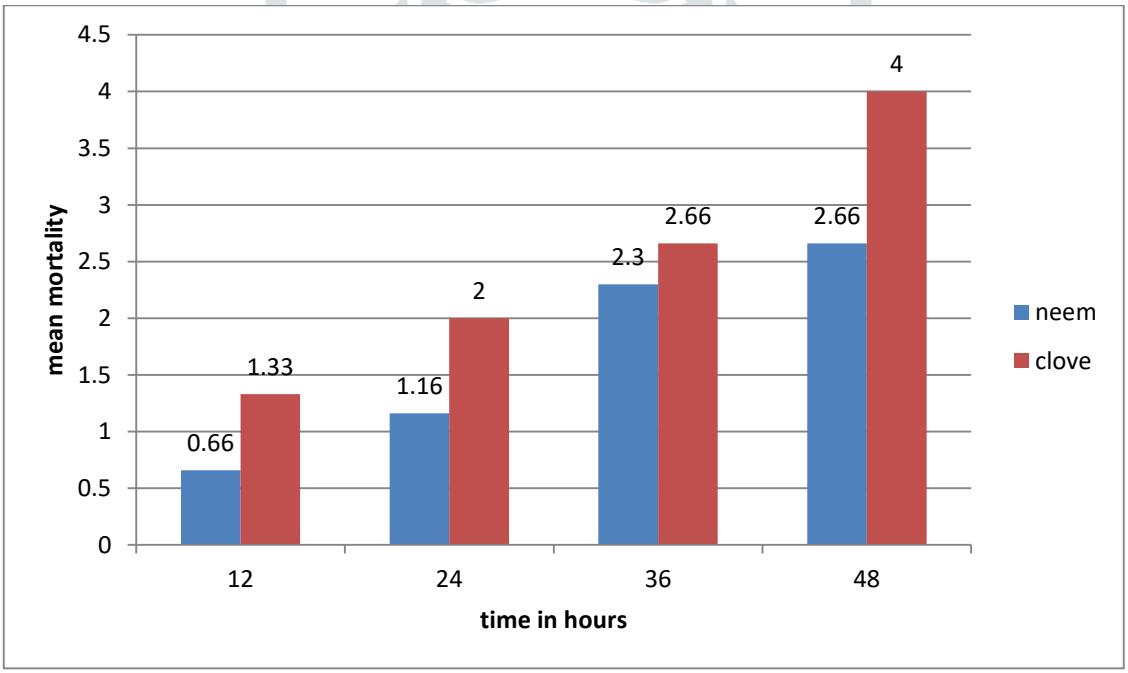

Fig2:-Toxicity of different exposure period on Tribolium castaneum

DISSCUSION

The study done in this experiment indicates high significant difference in susceptibility to clove and neem against adult *T. castaneum* after 48 h. Adults were more sensitive to the clove oil , as well as the percentage mortality increased as the treatment time prolonged and concentration incseaes. After 48 h clove oil showed 100% mortality and neem oil showed 86.6% mortality both at 25% concentration. Therefore the neem oil is less effective as compared to clove oil. The mortality percent depends on time interval and concentration. This result is similar to the result obtained by *Magda M.et.al(2010*) where they have used cove oil ,mustered oil and cumin oil .

The use of spices and medicinal plant which medicinal plants, which are normally used to cure human's illness is a old practice and are known to have effects on stored pests. Neem is probably the oldest botanical, which has been used as botanical pesticides. Its use in stored food protection has also been well known, especially in South Asian countries.

Previously for the management of economic loss caused by *T. castaneum*, several essential oils of botanical origin have been reported for their repellant, toxic and developmental inhibitory activities. Essential oils of *Anethum sowa* (Tripathi *et al*. 2000a), *Artemisia annua* (Tripathi *et al*. 2000b), *Lippia alba* (Verma *et al*. 2000) and *Elletaria cardomum* (Huang *et al*. 2000) have been reported for their repellant and toxic behavior against *T. castaneum*.

Clove buds have been found to repel T. castaneum and kill Ctenocephalides canis and Pediculus humanus humanus (Grainge and Ahmed 1988). Recently, Ho (1995) reported that non-polar clove extracts were very effective against adult S. zeamais and eggs of T. castaneum. Moreover, these extracts could suppress F_1 progeny production in both species of beetles, implying an ovicidal action of these extracts. Besides being insecticidal, clove extracts were repellent to S. oryzae. The repellent effect of cloves against stored grain insects is well documented (Grainge and Ahmed 1988).

The efficienct of the colve oil is due to Two-methoxy-4-(2-propenyl)-phenol (D5) wich is the major compound of clove oil, with a proportion of 83%, followed by trans-caryophyllene (D6), which is 12%. But 2-methoxy-4-(2-propenyl)-phenol (D5) is responsible for the mortality of toxic action of clove oil and the compounds did not act synergistically together. The clove oil and both its two components had repellent and toxicity activity on the 3 important stored grain insect pest species, *R. dominica, S. oryzae* and *T. castaneum*. (Zeng, L. et.al).

Neem seed oil is also a effective contact position and fumigaint against the adult of T.castaneum. Azadirectine in the neem seed oil ia a proven chemical for its insecticidal properties. The ethanol extract of neem from whereas parts also show toxicity against T. castaneum.

The toxic effect of essential oils, apart from the variability of phytochemical patterns, involves several other factors. The point of entry of the toxin is one of them. Commonly, essential oils can be inhaled, ingested or skin absorbed by insects. The fumigant toxicity of essential oils and their main components, the volatile monoterpenes, has been described (Smelyanets and Kuznetsov, 1968;

Netzurubanza, 1991; Weaver *et al*., 1991; Regnault-Roger *et al*., 1993; Regnault-Roger and Hamraoui, 1995).

The essential oils are generally composed of complex mixtures of monoterpenes, biogenetically related phenols, and sesquiterpenes. Examples include 1,8-cineole, the major constituent of oils from rosemary and eucalyptus; eugenol from clove oil; thymol from garden thyme; menthol from various species of mint; asarones from calamus; and carvacrol and linalool from many plant species. This blend of volatile oils can exert toxic, deterrent, antifeedant and repellent effects on insect herbivores . The essential oils seem to possess quadruple effects (ovicidal and emergence–reducing, repellency and toxicity) which should render them effective protectants of stored grains. A number of source plants have been traditionally used for protection of stored commodities, especially in the Mediterranean region and in Southern Asia, but interest in the oils was renewed with emerging demonstration of their fumigant and contact insecticidal activities to a wide range of pests in then 1990s (Isman, 2000).. The rapid action against some pests is indicative of a neurotoxic mode of action, and there is evidence for interference with the neuromodulator octopamine (Kostyukovsky *et al*., 2002) by some oils and with GABA-gated chloride channels by others (Priestley *et al*., 2003). The purified terpenoid constituents of essential oils are moderately toxic to mammals (Table 1), but, with few exceptions, the oils themselves or products based on oils are mostly nontoxic to mammals, birds, and fish (Stroh *et al*., 1998), therefore, justifying their placement under "green pesticides". Owing to their volatility, essential oils have limited persistence under field conditions; therefore, although natural enemies are susceptible via direct contact, predators and parasitoids reinvading a treated crop one or more days after treatment are unlikely to be poisoned by residue contact as often occurs with conventional insecticides. In fact, effects on natural enemies have yet to be evaluated under field conditions. Recent evidence for an octopaminergic mode-of-action for certain monoterpenoids (Bischof and Enan 2004; Kostyukovsky *et al*., 2002), combined with their relative chemical simplicity may yet find these natural products useful as lead structures for the discovery of new neurotoxic insecticides with good mammalian selectivity. The use of plants in this way as insecticides not only ensures safety of the environment and consumption of the treated produces, it is reliable, readily available for production by the farmer and economical, especially for the small scaled indigent farmers. All in all, plants of insecticidal potentials are compelling alternative to synthetic pesticides (Anyanga et al., 2013; Amoabeng et al., 2014; Stevenson, 2014).

CONCLUSION:

This study has revealed that both botanicals clove and neem have toxic effect on adult T. castaneum and clove oil is more effective than neem oil. The clove oil and neem oil are not toxic to mammals and don't have residual effect. As both the oil have shown toxic effect they may be used as an alternative to synthetic insecticide.

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CONFLICT OF INTEREST

The Authors declare no conflict of interest in this work.

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