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Fumigant toxicity of four essential oils and their combination against *Tribolium castaneum* in stored wheat

Ranjeet Kumar, PS Pandey and SN Tiwari

Abstract
The experiment were conduct to find out the fumigant toxicity of by *Murraya koenigii*, *Citrus reticulata, Callistemon citrinus* and *Curcuma longa* essential oils and their combination against *Tribolium castaneum* in stored wheat. 100 percent mortality was achieved by *M. koenigii*, *C. reticulata*, *C. citrinus* oil at 0.2 percent, *M. koenigii* + *C. reticulata*, *M. koenigii* + *C. citrinus* oil at 0.1 percent each and *M. koenigii* + *C. reticulata* + *C. citrinus* oil at 0.07 percent each and *M. koenigii* + *C. reticulate* + *C. longa* + *C. citrinus* oil at 0.05 percent each within 24 hours. The combination containing *C. reticulata* + *C. citrinus* oil at 0.1 percent each and *M. koenigii* + *C. reticulata* + *C. longa* oil at 0.07 percent each caused 93.3 and 86.7 percent mortality, respectively, within 24 hours which increased to 100 percent within 48 hours. The fumigant toxicity in grain treated with *C. reticulata* + *C. longa* + *C. citrinus* oil at 0.07 percent each was 76.7 percent within 24 hours, however, all insects died within 72 hours. The combination containing *C. reticulata* + *C. longa* at 0.1 percent each caused 73.3 percent mortality in 24 hours which increased to 100 percent within 96 hours.

Keywords: Fumigant toxicity, essential oil, *Tribolium castaneum*

Introduction
In India, only aluminium phosphide and methyl bromide are available for fumigation of food grain. The use of aluminium phosphide is restricted by law while methyl bromide needs special infrastructures for its use. Its improper use is also developing resistance in the insect pests of stored grain and in many parts of world. Now a time has reached when we have to think whether it is a boon or bane. The last four decades serious attempts are being made to find its alternative in plant kingdom and hundreds of plants have been screened to search their fumigation potential. And most interesting so many plants have been identified which are equipped with fumigant properties against insect pests of stored grain. Now it has been proved beyond doubt that under laboratory condition the plant components can suppress feeding and breeding of stored grain insect or even kill them completely within a few hours just like chemical fumigants. Essential oils are secondary metabolites that plants produce for their own needs other than nutrition. Generally they are complex mixture of 20-60 organic compounds that provides characteristic odour and flavor to leaves, flowers, fruits, seeds, bark and rhizomes [1]. Several essential oils have the properties of anti parasitical, bactericidal, fungicidal, virucidal and insecticidal properties [2]. Essential oil from more than seventy five plant species belonging to different families, have been studied for fumigant toxicity against several insect pests of storage grain. In view of above mentioned fact the present study was taken up to determine the days of mortality cause by essential oils and their combination against *Tibolium castaneum* in stored wheat by fumigant toxicity test.

Materials and Methods
Culture of the insect
Pure culture of test insects were developed in Biological Oxygen Demand maintained at 27°C±1 temperature and 70±5% relative humidity. Plastic jars of about 1.0 kg capacity were used for rearing purpose. At the center of the lid a hole of 1.8 cm diameter was made and covered with 30 mesh copper wire net to facilitate aeration in the jar. *T. castaneum* was cultured on its flour fortified with 5 per cent yeast powder. Before use, grain was disinfested in the oven at 60 °C for 12 hrs.

After disinfestation the moisture content of the grain was measured and raised to 13.5 per cent by mixing water in the grain. The quantity of water required to raise the moisture content was calculated by using following formula as described by [3]. After mixing the water in grain it was kept in closed polythene bags for a week so that moisture content of grain could equilibrate. The flour of wheat grain was then filled in plastic jar and 100 adults were released in jar after which it was kept in biological demand incubator. First generation adults (0-7 days old) were used for experimental purpose.

Procurement of *Essential oil*

The essential oils selected for the study were extracted from the locally available plants by steam distillation with Clevenger Apparatus in Department of Entomology Laboratory Bihar Agricultural University, Sabour. The extracted essential oils were kept in refrigerator at 4° C till experiment.

Preparation of grain

The experiments on *Tribolium castaneum* were conducted on graded and untreated wheat seed variety DBW-14. Before use, the grain was disinfested and raised moisture content to 13.5 percent as rearing conditions. To ensure the even distribution of water, the grain was spread on a platform and water was sprayed on it by hand sprayer. The grain was then mixed thoroughly and closed in polythene bag for a week for equilibration of moisture content of grain. The Experiments were conducted at same as rearing condition.

Fumigant toxicity test

The experiment was conducted on *T. castaneum* to study the fumigant toxicity of four essential oils and their combinations. The experiment was performed under controlled conditions at 27 ± 1^0C temperature and $70\pm$ 5 percent relative humidity. Fifty gram wheat grain variety DBW-14 (moisture 13.5 percent) was filled in 100 ml capacity of plastic vial. Twenty adult insects (0-7 days) of *T. castaneum* were released in each vial. After 24 hours of releasing the adults, required quantity of oil soaked on blank mat was inserted in each vial after which it was closed and sealed with paraffin wax. Each treatment was replicated three times. Observation was recorded after 24 hrs of treatment up to fifteen days.

Results and Discussion

Fumigant toxicity of four essential oils and their combination against *T. castaneum* in stored wheat

The fumigant toxicity four essential oils and their combination against *T. castaneum* in stored wheat is presented in Table 1 which indicated that 100 percent mortality was achieved by *M. koenigii*, *C. reticulata*, *C. citrinus* oil at 0.2 percent, *M. koenigii* + *C. reticulata*, *M. koenigii* + *C. citrinus* oil at 0.1 percent each and *M. koenigii* + *C. reticulata* + *C. citrinus* oil at 0.07 percent each and *M. koenigii* + *C. reticulate* + *C. longa* + *C. citrinus* oil at 0.05 percent each within 24 hours. The combination containing *C. reticulata* + *C. citrinus* oil at 0.1 percent each and *M. koenigii* + *C. reticulata* + *C. longa* oil at 0.07 percent each caused 93.3 and 86.7 percent mortality, respectively, within 24 hours which increased to 100 percent within 48 hours. The fumigant toxicity in grain treated with *C. reticulata* + *C. longa* + *C. citrinus* oil at 0.07 percent each was 76.7 percent within 24 hours, however, all insects died within 72 hours. The combination containing *C. reticulata* + *C. longa* at 0.1 percent each caused 73.3 percent mortality in 24 hours which increased to 100 percent within 96 hours. The *C. longa* oil at 0.2 percent and *C. longa* + *C. citrinus* oil at 0.1 percent each concentration were found less effective against *T. castaneum* [4]. Tested fumigant toxicity of essential oil from cumin (*Cuminum cyminum*) against eggs of two stored product insects, *T. confusum* and *Ephestia kuehniella,* it caused100 % mortality. Bio- efficacy of leaf oil of Curry leaf plant *Murraya koenigii* (Rutaceae) was evaluated against *C. chinensis* [5]. The essential oils of *Murraya koenigii*, *Citrus reticulata*, *Calistimone citrinus* either alone at 0.2 percent or two component combinations found highly effective against *S. oryzae* and *R. dominica* [6]. Essential oil of Sweet Annie, *Artemisia annua* (Compositae) evaluated by [7] against *T. castaneum* and *C. maculatus* at 1 percent showed adult repellent and with adult emergence of *T. castaneum*. [8] Tested fumigant toxicity of essential oil from cumin (*Cuminum cyminum*) against eggs of two stored product insects, *T. confuxum* and *Ephestia kuhniella,* it caused 100 percent mortality.

The present study concluded that the 100 percent mortality was achieved by *M. koenigii*, *C. reticulata*, *C. citrinus* oil at 0.2 percent, *M. koenigii* + *C. reticulata*, *M. koenigii* + *C. citrinus* oil at 0.1 percent each and *M. koenigii* + *C. reticulata* + *C. citrinus* oil at 0.07 percent each and *M. koenigii* + *C. reticulate* + *C. longa* + *C. citrinus* oil at 0.05 percent each within 24 hours. The combination containing *C. reticulata* + *C. citrinus* oil at 0.1 percent each and *M. koenigii* + *C. reticulata* + *C. longa* oil at 0.07 percent each caused 93.3 and 86.7 percent mortality, respectively, within 24 hours as they completely suppress the feeding and breeding of *Tribolium castaneum* in stored wheat.

Table 1: Fumigant toxicity of four essential oils and their combination against *Tribolium castaneum* in stored wheat

Essential oils	**Conc.%**	**Percent mortality of *Tribolium castaneum* at day after fumigation**														
		1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**13**	**14**	**15**
M. koenigii	0.2	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)
C. reticulata	0.2	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)
C. longa	0.2	33.3 (5.2)	33.3 (5.7)	26.7 (5.2)	36.7 (6.1)	40.0 (6.2)	43.3 (6.5)	43.3 (6.4)	20.0 (4.3)	36.7 (5.9)	70.0 (8.4)	70.0 (8.4)	90.0 (9.5)	93.3 (9.7)	100.0 (10.0)	100.0 (10.0)
C. citrinus	0.2	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)
M. koenigii+C. reticulata	0.2	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)
M. koenigii+C. longa	0.2	60.0 (7.6)	83.3 (9.1)	70.0 (8.4)	76.7 (8.5)	100.0 (10.0)	70.0 (8.4)	80.0 (9.1)	83.3 (9.1)	76.7 (8.7)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)
M. koenigii+C. citrinus	0.2	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)
C. reticulata+C. longa	0.2	73.3 (8.5)	93.3 (9.7)	93.3 (9.7)	100.0 (10.0)	90.0 (9.5)	93.3 (9.7)	70.0 (8.3)	80.0 (8.9)	96.7 (9.8)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)
C. reticulata+C. citrinus	0.2	93.3 (9.7)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)
C. longa+C. citrinus	0.2	26.7 (5.2)	73.3 (8.5)	36.7 (6.1)	50.0 (6.9)	40.0 (6.3)	100.0 (10.0)	73.3 (8.6)	63.3 (7.9)	66.7 (8.2)	96.7 (9.8)	86.7 (9.3)	90.0 (9.5)	83.3 (9.1)	93.3 (9.7)	100.0 (10.0)
M. koenigii+C. reticulata+C. longa	0.2	86.7 (9.3)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)
M. koenigii+C. reticulata+C. citrinus	0.2	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)
C. reticulata+C. longa+C. citrinus	0.2	76.7 (8.7)	96.7 (9.8)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)
M. koenigii+C. reticulata+C. longa+C. citrinus	0.2	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)	100.0 (10.0)
Untreated control		0.0 (1.0)	0.0 (1.0)	0.0 (1.0)	0.0 (1.0)	0.0 (1.0)	0.0 (1.0)	0.0 (1.0)	0.0 (1.0)	0.0 (1.0)	0.0 (1.0)	0.0 (1.0)	0.0 (1.0)	0.0 (1.0)	0.0 (1.0)	0.0 (1.0)
S.Em±		(0.67)	(0.28)	(0.19)	(0.47)	(0.26)	(0.21)	(0.35)	(0.33)	(0.28)	(0.09)	(0.26)	(0.16)	(0.06)	(0.04)	(0.001)
CD at 5%		(1.95)	(0.81)	(0.57)	(1.37)	(0.75)	(0.61)	(1.01)	(0.96)	(0.83)	(0.28)	(0.76)	(0.46)	(0.18)	(0.12)	(0.003)

Data in parenthesis indicate Square root (X+1) transformed value, E= Each

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