

## Evaluating Pesticide Resistance in Red Flour Beetle Populations: Significance for Food Security

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Received on: 26 -07-2024

Accepted on: 28- 08 -2024

### Abstract

The Red Flour Beetle (*Tribolium castaneum* H.) is a cosmopolitan pest of stored grains and cereals. This pest develops rapidly under favorable conditions in storage facilities such as warehouses, mills, and flour depots, affecting the quality of flour, causing significant economic loss, and threatening food security. The purpose of this study is to assess the resistance and efficacy of red flour beetle. The present study will play a very crucial role in the principle of integrated pest management (IPM), which enhances food security. Botanical pesticides Neem Tree (*Azadirachta indica*), AK or Milkweed (*Calotropis gigantea*) powder, Lemon Tree (*Citrus limetta*), and Babur/Kikar (*Acacia nilotica*) were evaluated. Neem recorded the highest mortality rate (20%), followed by (12%), lemon (11%), and the lowest Babur (9). The differences between the group means are statistically significant. P-value (0.00177) showed a low p-value (<0.05). A higher F-value suggests greater disparity between group means. F-value. Since  $9.421769 > 3.490295$ , the differences between the groups are significant. The mortality rate is directly proportional to the increased dose. The neem appears to be the most effective, with the highest mortality rate of 20% at 20 g. Babur, on the other hand, has the lowest mortality rate, starting at just 2% at 5 g.

**Keywords:** Red Flour Beetle, Botanical Pesticides, Mortality, Efficacy

## 1. INTRODUCTION

The red flour beetle (*Tribolium castaneum*) belongs to the family Tenebrionidae, order Coleoptera. This species is found throughout the world and is a pest of stored grain, especially food grains. [1] The red flour beetles feed on the grain and other products of flour, cereals, beans, and nuts, leading to significant loss. Red flour beetles are pests that can survive in different environmental conditions, such as hot and moist conditions. Usually they infest post-harvest wheat; they directly feed on grain and flour [2]. Grains and processed products are the major source of nutrients for human beings, and they should be continually available to the population of the world [3]. However, it was recorded that about 690 million population is suffering in malnutrition after COVID-19 and by the 2050 the population is expected to increase in large scale to meet the food demand for population, it should be increased food sources 50% more than current volume [4]. There are many aspects of restrictions to safeguarding global food security, including climate change, different environmental factors, and water and land availability [5]. The red flour beetle is a major pest of grain; due to infestation, the weight and nutritional value become less, and infected grain becomes moisturized, which is the main reason for the attack of the microbes that create a bad odor in the grain, which has a direct effect on the quality of grain and flour [6]. Stored grain is mostly protected by the chemical or fumigation, which are the most effective methods to control the red flour beetle; however, the use of the very dangerous chemicals is like [7]. Methyl bromide is used to control the pest, but this chemical is very harmful for humans. So it was decided by January 2015. In the world, another fumigation chemical named phosphine is dominant and is used to control the pest. However, the red flour beetle has developed resistance against this chemical [8]. The European Union has decided to stop phosphine because it is unsafe for humans and may cause serious problems related to health; therefore, some fumigants can be used for control, but still they may cause problems (Navarro, 2006). The control of the red flour beetle is a challenge for the farmers, landlords, and stockholders because this pest damages the grain, resulting in qualitative and quantitative loss. The infected grain is not able to be used due to this loss; it causes an economic loss [9]. The botanical pesticides are eco-friendly; their effects are less on humans as compared to chemical pesticides; they are used for the control of red flour beetles. Therefore, botanical pesticides are recommended to use for control [10]. Many researchers recommended that botanical pesticides are very effective. The oil of neem, in which azadirachtin is very useful, affects the molting of red flour beetles and disturbs the reproduction results, reducing the population of the pest [11]. Oil of the eucalyptus and mentha is mostly used as a fumigant to control the pest. The use of powder from many leaf plants and seeds is very helpful; it reduces the oviposition and direct effects on the population of the pest. The botanical pesticides also affect the metabolism of the pest. Terpenoids and alkaloids of some plants have direct effects on the nervous system, leading to paralysis of the pest and ultimately death [12]. The use of botanical pesticides to control the red flour beetle has some challenges, like limited shelf life and higher production costs compared to synthetic pesticides. Advances in formulation technology, such as Nano encapsulation are addressing these limitations and enhancing the stability and bioavailability of plant-derived pesticides [13].

## 2. RESEARCH METHODOLOGY

### 2.1 Study area

The research work was done in the laboratory of the Department of Zoology, GC University Hyderabad. The leaves of the neem tree (*Azadirachta indica*), milkweed (*Calotropis gigantean*), lemon tree (*Citrus limetta*), and babur/kikar (*Acacia nilotica*) were collected around the field of the GC University Hyderabad. Red flour beetles (*Tribolium castaneum* H.) were collected from the different markets of Hyderabad.

### 2.2 Plants selected for botanical pesticides

For the assessment, four different types of botanical pesticides were selected to evaluate the pesticide resistance in the red flour beetle (*Tribolium castaneum*).

I. Neem Tree (*Azadirachta indica*)

II. Milkweed (*Calotropis gigantean*)

III. Lemon Tree (*Citrus limetta*)

IV. Babur/Kikar (*Acacia nilotica*)

### 2.3 Preparation of Insecticides

Leaves of 4 different plants, namely the neem tree (*Azadirachta indica*), milkweed (*Calotropis gigantean*), lemon tree (*Citrus limetta*), and Babur/Kikar (*Acacia nilotica*), were washed with tap water to clean the dust off of them. The clean leaves were placed in sunlight for a week. After that, leaves were blended into a blender and the powder was sifted through a cloth. These fine powders of botanical pesticides were tested in four doses of 5 g, 10 g, 15 g, and 20 g [14].

### 2.4 Experiment Design

Wheat grain sterilized at 60°C for 60 to 90 minutes. Weigh 5.0 grams of Neem Tree (*Azadirachta indica*) powder mixed with 1 kilogram of wheat and leave for 3 days, ensuring the wheat grain is saturated with the neem powder. 30 adult individuals (male and female) of Red Flour Beetle (*Tribolium castaneum*) were released in a jar with 50 grams of treated wheat grain and covered with muslin cloth under laboratory environmental conditions at  $25 \pm 1^\circ\text{C}$  temperature with a relative humidity of  $60\% \pm 5\%$ . This process was repeated for other botanical pesticides, Milkweed (*Calotropis gigantean*) powder, Lemon Tree (*Citrus limetta*) powder, and Babur/Kikar (*Acacia nilotica*), respectively. In addition, for the control treatment, 30 adult individuals (male and female) of red flour beetle (*Tribolium castaneum*) were released with 50 grams of untreated wheat grain. Each experiment was replicated four times at different doses such as 5 g, 10 g, 15 g, and 20 g. Insect mortality and feeding behavior were recorded on a daily basis.

### 2.5 Statistical analysis

Data was analyzed statistically using ANOVA (one way) using statistical software (Ver. 8.1) to find the highest significant difference between Neem Tree (*Azadirachta indica*), Milkweed (*Calotropis gigantean*) powder, Lemon Tree (*Citrus limetta*), and Babur/Kikar (*Acacia nilotica*), respectively. The result was expressed at F value, P value, and F crit.

### 3. RESULTS AND DISCUSSION

#### 3.1 Morphological description of Red Flour Beetle (*Tribolium castaneum*)

Red flour beetle reddish brown to rust in colour, size about 3-4mm long. Head; small and slightly elongated, clubbed antennae with gradual tapering toward the tip, Eyes; compound eyes, well developed, mouthparts; chewing mouthparts, adapted for grains and flour. Thorax; convex, slightly smooth, pronotum; slightly rounded sides, three pair well developed legs, elytra; hard forewings, covered the membrane, hind wings; longitudinal grooves or ridges. Abdomen; segmented, elongated, ventral side flat, dorsal side covered with elytra, ends rounded, distinguished it from other similar species (Figure 1)

#### 3.2 Efficacy of pesticides

After 60 days of treatment among 4 different botanical pesticides, Neem Tree (*Azadirachta indica*), Milkweed (*Calotropis gigantea*) powder, Lemon Tree (*Citrus limetta*), and Babur/Kikar (*Acacia nilotica*) were evaluated. Neem recorded the highest mortality rate (20%), followed by (12%), Lemon (11%), and the lowest Babur (9) (Table 1). P-value (0.00177) showed a low p-value ( $<0.05$ ), indicating that the differences between the group means are statistically significant. F-value (9.421769): Indicates the ratio of the variance between the groups to the variance within the groups. A higher F-value suggests greater disparity between group means. F crit (3.490295): This is the critical value to compare against the F-value. Since  $9.421769 > 3.490295$ , the differences between the groups are significant (Table 2). Table 1 shows the mortality rate of the red flour beetle (*Tribolium castaneum*) after exposure to four different botanical pesticides: neem tree (*Azadirachta indica*), milkweed (*Calotropis gigantea*) powder, lemon tree (*Citrus limetta*), and Babur/Kikar (*Acacia nilotica*). The mortality rate is directly proportional to the increased dose. The neem appears to be the most effective, with the highest mortality rate of 20% at 20 g. Babur, on the other hand, has the lowest mortality rate, starting at just 2% at 5 g.

#### 3.2 Resistance in the Red Flour Beetle against pesticides

The highest mortality on Neem indicated that it had the least resistance among the four botanical pesticides. The mortality rate increases significantly with higher dosages, from 12% at 5 grams to 20% at 20 grams. This suggests that Neem was highly effective even at lower doses, and its effectiveness continues to improve as the dosage increases. It had moderate resistance, with mortality rates increasing from 7% at 5 grams to 12% at 20 grams. Although it was not as effective as Neem, it showed a notable increase in mortality with higher dosages, indicating its potential as a botanical pesticide, with higher resistance compared to Neem. Lemon showed slightly higher resistance than milkweed, with mortality rates ranging from 4% at 5 grams to 11% at 20 grams. The increase in mortality was consistent but less pronounced than Neem and Milkweed, suggesting that while it had pesticidal properties, it required higher dosages to achieve similar effectiveness. Babur showed the highest resistance among the tested pesticides, with the lowest mortality rates overall. Starting at just 2% at 5 grams, it only reaches 9% at 20 grams. This indicated that Babur is the least effective botanical pesticide in this group, showing high resistance and requiring much higher dosages to have any significant impact on the pests (Figure 3).

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### 4. CONCLUSION

Among 4 different botanical pesticides are Neem Tree (*Azadirachta indica*), Milkweed (*Calotropis gigantea*) powder, Lemon Tree (*Citrus limetta*), and Babur/Kikar (*Acacia nilotica*). Neem has the highest average value (16.25) and demonstrates the greatest efficacy among the pesticides tested (Figure 2). Babur has the lowest average value (5.75), indicating the least efficacy (Figure 3). The significant differences (low P-value and high F-value) suggest that Neem is statistically more effective than the other pesticides, and Lemon has moderate average values, with a lower variance, indicating more consistent results.

### 5. ACKNOWLEDGMENT

I am very grateful to GC University Hyderabad for financial support for research under the Research Support Program (RSP-GCUH).

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**Table No.1. Efficacy of four different botanical pesticides against *Tribolium castaneum***

Dozes in grams	Neem Mortality %	Milkweed Mortality %	Lemon Mortality %	Babur Mortality %
5	12	7	4	2
10	15	8	6	4
15	18	9	8	8
20	20	12	11	9

**Table No.2. Showing the SS, df, MS, F, P-value and F crit of four different botanical pesticides against *Tribolium castaneum***

<i>Botanical pesticide</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>		
Neem	4	65	16.25	12.25		
Milk weed	4	36	9	4.666667		
Lemon	4	29	7.25	8.916667		
Babur	4	23	5.75	10.91667		
ANOVA						
Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	259.6875	3	86.5625	9.421769	0.001772	3.490295
Within Groups	110.25	12	9.1875			
Total	369.9375	15				

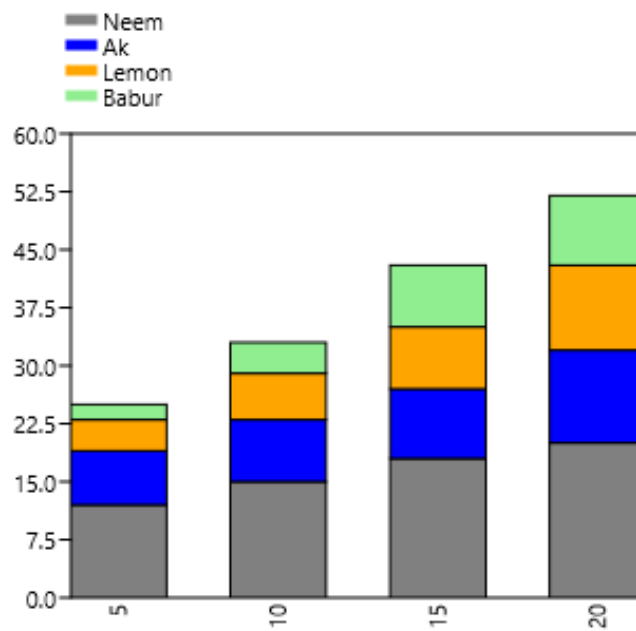
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**Figure 1. Red Flour  
(*Tribolium castaneum*)**



**Beetle**

**Fig 2: Efficacy of  
botanicals  
against *Tribolium***

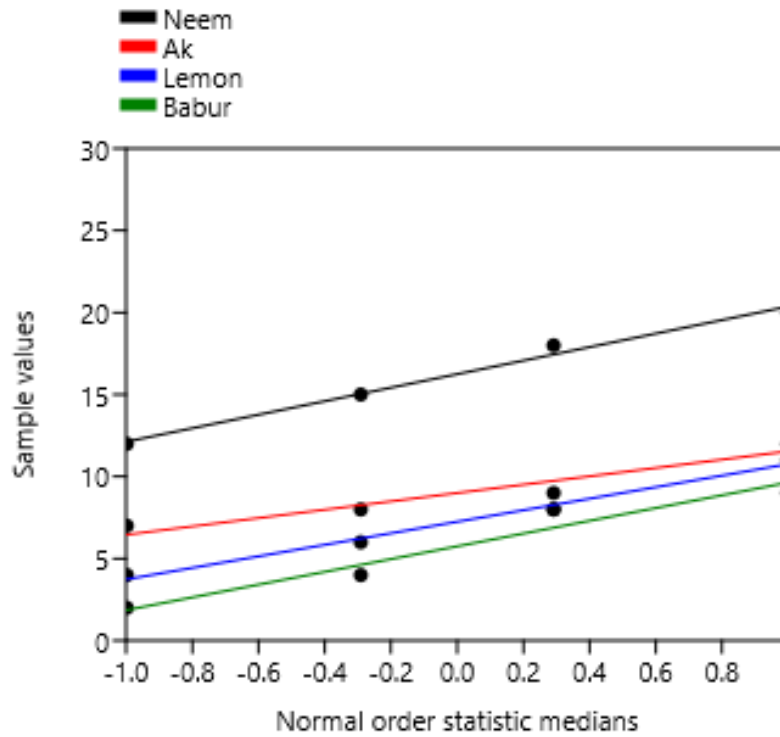


different  
pesticides  
*castaneum*



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Fig 3:  
*Tribolium*  
 on  
 botanical



Resistant in  
*castaneum*  
 different  
 pesticides