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Entomotoxicity of plant powders against Pulse beetle (*Callosobruchus chinensis*) on stored mung bean (*Vigna radiata*)

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Abstract
Experiments were conducted in the biology laboratory of the Department of Entomology, Gomal University, Dera Ismail Khan during 2015 to investigate the efficacy of six different botanicals (Neem, Bakain, Dharek, Turmeric, Tumha and AK), each at six different concentrations (0.5, 1.0, 1.5, 2.0, 2.5 and 3%) for the management of *C. chinensis*. Fifty gram of sterilized Mung bean grains were treated with these selected plant powders at six different concentrations viz. 3.0, 2.5, 2.0, 1.5, 1.0, 0.5% and data was recorded after 24, 48, 72 hours and 7 days of exposure period. Neem and Turmeric were found to be more effective in all concentrations against pulse beetle as they showed more toxicity against the tested pest. The Neem and Turmeric powders were found comparatively more effective in controlling progeny production of *C. chinensis* than other tested plant powders. So, it is concluded from the present findings that Neem and Turmeric powders should be incorporated into grain protection practices.

Keywords: mung bean, pulse beetle, plant powders, toxicity

1. Introduction
Mung bean (*Vigna radiata)* is very important leguminous crop. It is a major source of protein and easily digestible especially when used with cereals [1-3]. It plays an important role in nitrogen fixation and restores soil fertility [4]. Mung bean is grown twice in a year in Pakistan. It was cultivated on about 1,20,000 hectares in 2011 with production of 81, 000 metric tons grains [5]. Mung bean was grown majorly in the Punjab province on about 88% area with 85 % of the total production. Cultivation of Mung bean is concentrated in Bhakkar, Rawalpindi, Layyah and Mianwali districts. During July to October Mung bean was grown in Pakistan. It is grown in various rotations of major crops but in about 75% cultivations Mung bean was followed by wheat crop [6].

Mung bean is attacked by different insect pest species from the initial stages to the harvesting period. About 200 insect pests belonging to forty eight families of order Hemiptera, Isoptera, Orthoptera, Coleoptera, Thysanoptera and seven Mites species of Order Acarina are known to attack Mung bean and black gram. On the basis of crop phenology, Mung bean pests were classified as stem, foliage, pod feeders and insect pests of stored products. Major stored grain insect pests include Bruchid species in the Genus *Callosobruchus* Annually 20% yield loss in pigeon pea, 15% in chickpea and 30% in Mung bean and black gram was recorded. Due to stored grain pests about 2.5-3 million tons of grains were lost annually [7]. The Bruchids, *C.chinensis* (L.) cause damage in the field, after that the emerging adults lay their eggs on fresh grains in the storage. In the field the infestation by *C.chinensis* is about 7.8 to 9.9% and whole grains were damaged when infestation was about 9.9 percent[8]. *C. chinensis* (L.) commonly known as pulse beetle infest all kinds of pulses but chickpea and beans are damaged both quantitatively and qualitatively and make them unfit for the human consumption. When stored grains were eaten by the pest the germination capacity of these grains were lost [9, 10].

C. chinensis was first identified in China in 1758, where the species name of beetle was given to it [11]. It is cosmopolitan in nature and distributed in Japan, Srilanka, Philippines, India etc. *C. chinensis* is the cosmopolitan, major destructive insect pest of important pulse crops such as Mung bean, Chickpea, Lentil, Arhar, Black gram, Peas, Cow pea [12, 13]. It is serious insect pest of stored products in the temperate regions with annual loss of about 0.21 million tons of Rs 315 million [4]. Bruchid life cycle was studied on different leguminous crops [15-19] *C. chinensis*

is controlled by fumigation in the storage conditions. Due to the excessive use of these toxic chemicals certain problems develop such as pest resurgence, ill effect on beneficial insects, and environmental pollution, etc. Various efforts are being made to minimize the excessive use of pesticides and emphasize on the IPM techniques including the use of plant powders for safer control of the pest.

2. Materials and Methods

The research work was conducted in the biology laboratory of the Department of Entomology, Faculty of Agriculture, Gomal University, Dera Ismail Khan during 2015.

2.1 Pulse beetle culture

Mung bean grains infested by the Pulse beetle (*C. chinensis*) were collected from Agriculture Research institute (ARI) D. I. Khan and the infested grains were brought to the laboratory. Infested Mung bean seeds were placed in a plastic jar (10x 12cm) and covered with a piece of muslin cloth to avoid escape of pulse beetle adults. Newly emerged adult beetles from this jar were transferred to another plastic jar and were provided sterilized Mung bean seeds for oviposition purpose. The pulse beetle cultures were kept at 27 ± 3ºC and 65 ± 5% R.H with photoperiod of 12:12 hours (light and dark). The freshly emerged subsequent adult generations were used in the experiments.

2.2 Plant materials

The selected plant materials showed in Table 1 were obtained from local market as well as growers and were shade dried and kept under controlled environmental conditions for further experiments.

Table 1: Detail of Plant Materials evaluated for insecticidal activities against *C.chinensis*

S. No	Common Name	Botanical Name	Plant part used
1	Neem seed	*Azadirachta indica*	Seed
2	Bakain seed	*Melia azedarach*	Seed
3	Dharek seed	*Melia azadirach*	Seed
4	Turmeric	*Curcuma longa*	Rhizomes
5	Tumha	*Citrullus colocynthis*	Fruit
6	Akk	*Calotropis gigantean*	Leaves

2.3 Preparation of plant extracts

Seed, fruit, rhizome and leaves of selected plants such as Neem, Bakain, Dharek, Akk, Turmeric and Tumha were collected from different growing areas and were brought to the biological laboratory. All the plant parts were shade dried and were powdered with the help of electric grinder. After that all the powders were sieved out for obtaining a fine powder.

2.4 Toxicity of plant materials against pulse beetle *C. chinensis.*

The experiment was conducted to test the toxicity of selected plant powders mentioned in Table 1 against pulse beetle. The experiment was laid in completely randomized design having 5 replicates at 27 ± 3ºC and 65 ± 5% R.H with 12:12 hours (L: D) The selected plant powders (Neem, Bakain, Dharek, Turmeric, Tumha and Akk) were tested at six different concentrations (0.5, 1, 1.5, 2, 2.5 and 3% w/w). There were 7 treatments including control. In each treatment, twenty grams of sterilized Mung bean grains were treated with the selected plant powders at six different concentrations. The treated seeds were kept in Petri dishes and before pulse beetle release, the Petri dishes were shaken well for complete mixing of selected plant powders. Freshly emerged ten pairs of pulse beetles were released in the test arena. The pulse beetles were starved for one hour before release. The data were recorded up to 100% mortality of the pulse beetle after treatments at 24, 48, 72 hours and 7 days of exposure period. The dead pulse beetles were removed daily from the petri dishes. Corrected percent mortality was calculated as:

$$\text{Corrected \% mortality} = \frac{\text{\% mortality in treatment - \% mortality in control}}{\text{100-\% mortality in control}} \times 100$$

2.5 Statistical analysis

The data of each parameter was analyzed statistically by using statistics 8.1 software and treatment means data was separated by using Fisher protected LSD test significance level of 5 percent [20].

3. Results

3.1 Mortality after 24 hours of exposure period

At each concentration the tested plant materials had significant effect ($P<0.05$) on the mortality of *C. chinensis* after twenty four hour exposure period (Table 2). Maximum mortality of *C.chinensis* was recorded in Mung bean grains treated with Neem while, minimum was recorded with Akk at all concentrations. At each concentration the mortality was significantly different among treatments. Overall trend of increase in mortality was observed with increase in concentration for all the treatments after 24 hours of exposure period. Also the mortality of the test insect increased as the exposure period to the plant powders was increased.

3.2 Mortality after 48 hours of exposure period

Results presented in Table 3 reveal the toxicity of six different concentrations of selected plant powders against the *C.chinensis.* Maximum mortality of *C.chinensis* was recorded in Mung bean grains treated with Neem while, minimum was recorded with Akk at all concentrations after 48 hours of exposure period. Maximum mortality of the test insect was recorded at 3% concentration of Neem whereas; minimum mortality was recorded in case of Akk. Similarly powder of Turmeric was also found effective against the test insect followed by Bakain, Dharek and Akk. The test insect mortality increased with the increase in concentration of the tested powders and also with the exposure time to the plant powders.

3.3 Mortality after 72 hours of exposure period

After an exposure period of 72 hours, the maximum mortality was recorded in Neem powder treatment at 3% concentration which differed significantly ($P<0.05$) from all other treatments; whereas, minimum mortality was recorded in Akk at 3% concentration (Table 4). Similarly Turmeric was also found effective against the *C. chinensis* followed by Bakain, Dharek and Tumha respectively. Overall trend of increase in mortality was observed with increase in concentration for all the treatments after 72 hours of exposure period. Also the mortality of the test insect increased as the exposure period to the plant powders was increased.

3.4 Mortality after 7 days of exposure period

It is evident from the Table 5 that the effect of Neem and Turmeric was found most promising as 100% adult mortality

of the test insect was recorded, at 3% concentration after 7 days exposure period when reared on treated Mung bean grains. At each concentration the mortality was significantly different among treatments. Overall trend of increase in mortality was observed with increase in concentration for all the treatments after 7 days of exposure period. Also the mortality of the test insect increased as the exposure period to the plant powders was increased.

Table 2: Mean percent (± SE) mortality of *C.chinensis* on Mung bean grains treated with different concentrations of Plant powders after 24 hours of exposure period during 2015.

Treatment	Concentrations (%)					
	0.5%	1%	1.5%	2%	2.5%	3%
Neem	21.00 ± 2.23 a	22.00 ± 2.73 a	26.00 ± 2.23 a	31.00 ± 2.23 a	36.00 ± 2.23 a	41.00±2.2 a
Bakain	10.00 ± 2.73 c	10.00 ± 2.23 c	15.00 ± 4.47 c	20.00 ± 2.23 c	25.00 ± 2.73 c	30.00 ± 2.23 c
Dharek	5.00 ± 2.23 d	5.00 ± 2.23d	10.00 ± 2.73 d	15.00 ± 2.73 d	20.00 ± 2.23 d	25.00 ± 2.23 d
Turmeric	15.00 ± 2.23 b	17.00 ± 2.73b	21.00 ± 2.23 b	26.00 ± 2.23 b	31.00 ± 2.23 b	36.00 ± 2.23 b
Tumha	5.00 ± 2.23 d	5.00 ± 2.23 d	10.00 ± 2.23 d	15.00 ± 2.73 d	20.00 ± 0.00 d	25.00 ± 2.23 d
Akk	0.00 ± 2.23 e	5.00 ± 2.23 d	5.000 ± 2.23 e	10.00 ± 2.23 e	15.00 ± 0.00 e	20.00 ± 2.23 e
LSD Value	1.54	1.89	1.54	1.54	1.54	1.54

Each value shows a mean ± S.E of five replications. Means with the same letters in a column are not significantly different at (P>0.05) using LSD Test.

Table 3: Mean Percent (± SE) mortality of *C.chinensis* on Mung bean grains treated with different concentrations of Plant powders after 48 hours of exposure period during 2015.

Treatment	Concentration (%)					
	0.5%	1%	1.5%	2%	2.5%	3%
Neem	26.00 ± 2.23 a	31.00 ± 2.73 a	36.00 ± 2.23 a	41.00 ± 2.23 a	46.00 ± 2.23 a	51.00 ±2.23a
Bakain	15.00 ± 2.23 c	20.00 ± 2.73 c	25.00± 2.23 c	30.00 ±0.00 c	35.00 ± 0.00 c	40.00 ± 0.00 c
Dharek	10.00 ± 2.73 d	15.00 ± 2.23 d	20.00± 2.73 d	25.00 ± 2.23 d	30.00 ± 2.23 d	35.00 ± 2.73 d
Turmeric	21.00 ± 2.23 b	26.00 ± 2.23 b	31.00 ±2.23 b	36.00 ± 2.73 b	41.00 ± 2.23 b	46.00± 2.23 b
Tumha	10.00 ± 2.23 d	10.00 ± 2.23 e	15.00 ± 2.23 e	20.00 ± 2.23 e	25.00 ± 0.00 e	35.00 ± 0.00 e
Akk	5.00 ± 2.23 e	5.00 ± 0.00 f	10.00 ± 0.00 f	15.00 ± 0.00 f	20.00 ± 2.23 f	20.00 ± 2.23 f
LSD Value	1.54	1.54	1.54	1.54	1.54 1.54	

Each value shows a mean ± S.E of five replications. Means with the same letters in a column are not significantly different at (P>0.05) using LSD Test.

Table 4: Mean Percent (± SE) mortality of *C.chinensis* on Mung bean grains treated with different concentrations of plant powders after 72 hours of exposure period during 2015.

Treatment	Concentration (%)					
	0.5%	1%	1.5%	2%	2.5%	3%
Neem	41.00 ± 2.23 a	46.00 ± 2.23 a	51.00 ± 2.23 a	61.00 ± 2.23 a	80.00 ± 2.23a	81.00 ± 2.23 a
Bakain	20.00 ± 2.23 c	25.00 ± 2.23 c	30.00 ± 0.00 c	35.00 ± 4.47 c	40.00 ±2.23 c	45.00 ± 0.00 c
Dharek	15.00 ± 2.73 d	20.00 ± 2.73 d	25.00 ± 2.23 d	30.00 ± 2.23 d	35.00 ± 0.00 d	40.00 ± 2.73 d
Turmeric	26.00 ± 2.23 b	41.00 ± 2.23 b	46.00 ± 0.00 b	51.00 ± 0.00 b	56.00 ± 2.23b	61.00 ± 2.23 b
Tumha	15.00 ± 2.73 d	20.00 ± 0.00 d	25.00 ± 2.23 d	25.00 ± 2.23 e	30.00 ± 0.00 e	35.00 ± 2.73 e
Akk	10.00 ± 2.23 e	15.00 ± 2.23 e	20.00 ± 2.73 e	25.00 ± 2.73 e	30.00 ± 2.23 e	31.00 ± 2.23 f
LSD Value	1.54	1.54	1.56	1.54	1.00	1.89

Each value shows a mean ± S.E of five replications. Means with the same letters in a column are not significantly different at (P>0.05) using LSD Test.

Table 5: Mean Percent (± SE) mortality of *C.chinensis* on Mung bean grains treated with different concentrations of plant powders after 7 days of exposure period during 2015.

Treatment	Concentration (%)					
	0.5%	1%	1.5%	2%	2.5%	3%
Neem	51.00 ± 1.17 a	55.57 ± 1.29 a	75.78 ± 1.76 a	100.00 ± 0.00 a	100.00 ± 2.23 a	100.00 ±2.23a
Bakain	29.26 ± 1.65c	30.00± 1.65c	34.31 ± 1.53 c	39.36 ±1.41 c	44.42 ± 1.29 c	49.47 ± 1.18 b
Dharek	24.21 ± 1.76 d	25.00± 1.76 d	29.26 ± 1.65 d	34.31 ± 1.53 d	39.36 ± 2.92 d	44.42 ± 1.29 c
Turmeric	41.00 ± 0.93 b	45.47 ± 1.05 b	65.68 ± 1.52 b	90.94 ± 2.11 b	96.00 ± 2.23 b	100.00 ± 2.23 a
Tumha	20.00 ±1.76 d	24.21 ± 1.76d	29.26 ± 1.65 d	34.31 ± 1.53 d	39.36 ± 1.41 d	44.42 ± 1.29 c
Akk	19.15± 1.88 e	24.21± 1.88 e	25.00± 1.76 e	29.26 ± 1.65 e	34.31 ± 2.51 e	39.37 ± 1.41 d
LSD Value	1.54	1.91	1.98	1.82	1.76	1.67

Each value shows a mean ± S.E of five replications. Means with the same letters in a column are not significantly different at (P>0.05) using LSD Test

4. Discussion

Among the tested plant powders, Neem and Turmeric had repellent and deterrent effects on oviposition and emergence of *Callosobruchus chinensis.* Also the behavior, survival, reproduction and longevity of the test insects were significantly affected by some of the tested plant powders. Among all the treatments, Neem seed powder was recorded statistically most effective followed by Turmeric. After an exposure period of 7 days, Neem powder caused 100 % mortality of *C. chinensis* which was significantly much better from all the other treatments. These findings are in accordance with Kavillieratos *et al* [21] They concluded that

mortality of *Sitophilus oryzae*a nd *Tribolium confusum* reared on the wheat and maize can occur when azadiracht in based insecticides were ingested. Bio-efficacy of Neem seed powder has been identified for its effectiveness against many stored grain pests [22]. *C. chinensis* developmental duration was also prolonged when Neem treated Mung bean grains were offered to the tested insect and also very few adults emerged from these grains. The results revealed that Neem and Turmeric powders affect the post embryonic development of the *C.chinensis* which resulted in less adult emergence and prolonged growth. Plant products have the ability to penetrate the insect egg chorion with the help of micropyle, resulting in death of the embryos [23-25]. Based on the amount and pest type Neem insecticides have growth retardation and toxic properties [26]. These results are in accordance with previous studies [27] where clove seed powder and cashew nut powder reduced the sorghum grains weight loss by *C. chinensis.* Powder of red pepper and clove have been observed to reduce the percent infestation by Bruchids @ 25 gram per kg [28].

5. Conclusion and recommendation

Amongst the plant powders tested, Neem and Turmeric were found to be more effective against *C.chinensis* as they showed more toxicity *A. indica* and *C. longa* powders were found comparatively more effective in controlling progeny production of *C.chinensis* than other tested plant powders. Based on the conclusions, it is recommended that *A. indica* and *C. longa* powders should be incorporated into grain protection practices.

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