

Original article (Orijinal araştırma)

Insecticidal activity of weed plants, *Euphorbia prostrata* and *Chenopodium murale* against stored grain insect pest *Trogoderma granarium* Everts, 1898 (Coleoptera: Dermestidae)

Yabani bitkiler *Euphorbia prostrata* ve *Chenopodium murale*'nin depolanmış tahıl zararlısı *Trogoderma granarium* Everts, 1898 (Coleoptera: Dermestidae)'a karşı insektisidal aktivitesi

**Kishwar SULTANA¹ Muhammad Kashif ZAHOOR^{1*} Muhammad SAGHEER²
Shabab NASIR¹ Muhammad Asif ZAHOOR³ Farhat JABEEN⁴
Bushra RIAZ¹**

Summary

The efficiency of petroleum ether extracts of weed plants, *Euphorbia prostrata* and *Chenopodium murale*, for the control of *Trogoderma granarium* was investigated. The extracts were prepared in petroleum ether using the whole plants. Diet incorporation was used for mortality bioassay and area preference was used for repellency against third instar larvae of *T. granarium*. The results showed relatively high rate of larval mortality after 6 days with extract concentrations of 10, 20 and 30%. At 30% the corresponding mortality rates induced by *E. prostrata* and *C. murale* were 20 and 25%, respectively. Low larval mortalities were obtained for both plant extracts at 10%. Similarly, repellency assay at 30% extracts found the maximum proportion of larvae that moved away from the treated region of the filter paper to be 88 and 87% for *E. prostrata* and *C. murale* extracts, respectively. The repellency of both plant extracts had a positive relationship applied dosage but was negatively correlated with exposure time. The lowest mean number of larvae in F₁ generation was found with 30% *E. prostrata* and *C. murale* extracts (60 and 53, respectively) as compared to the control (149). Overall the results indicated that the *C. murale* extract had a higher insecticidal activity against *T. granarium* than the *E. prostrata* extract.

Keywords: Mortality, progeny reduction, repellency, *Trogoderma granarium*, weed plants

Özet

Trogoderma granarium'un mücadelesinde, *Euphorbia prostrata* ve *Chenopodium murale* yabani bitkilerinin petrol eteri ekstraktlarının verimliliği araştırılmıştır. Ekstraktlar tüm bitki kullanılarak, petrol eteri içinde hazırlanmıştır. *Trogoderma granarium*'un üçüncü dönem larvalarında, ölüm denemeleri için diyet karışımı ve kaçırıcı etki için alan tercihi kullanılmıştır. Sonuçlar, %10, 20 ve 30 ekstrakt konsantrasyonlarında 6 gün sonra nispeten yüksek larva ölüm oranı göstermiştir. %30 konsantrasyona karşılık gelen ölüm oranları *E. prostrata* ve *C. murale* için sırasıyla, %20 ve 25 olmuştur. Düşük larva ölümleri her iki bitkinin %10'luk ekstraktında elde edilmiştir. Benzer şekilde, %30 ekstraktıyla yapılan kaçırıcı etki denemelerinde filtre kağıdının uygulama yapılmış bölgesinden daha uzağa hareket eden larvaların maksimum oranı, *E. prostrata* ve *C. murale* için sırasıyla %88 ve 87 olarak bulunmuştur. Her iki bitki ekstraktının kaçırıcı etkisinde uygulanma dozlarında pozitif bir ilişki varken maruz kalma süresi için olumsuz korelasyon bulunmuştur. F₁ neslinde en düşük ortalama larva sayısı kontrol (149) ile karşılaştırıldığında *E. prostrata* ve *C. murale* için (sırasıyla 60 ve 53) %30 konsantrasyonda bulunmuştur. Tüm sonuçlar, *C. murale* ekstraktının *T. granarium*'a karşı *E. prostrata* ekstraktından daha yüksek bir insektisidal aktiviteye sahip olduğunu göstermiştir.

Anahtar sözcükler: Ölüm, döl azalma, kaçırıcı etki, *Trogoderma granarium*, yabani bitkiler

¹ Entomology Laboratory, Department of Zoology, Government College University, Faisalabad, Pakistan

² Grain Research, Training and Storage Management Cell, Department of Entomology, University of Agriculture Faisalabad, Pakistan

³ Department of Microbiology, Government College University, Faisalabad, Pakistan

⁴ Department of Zoology, Government College University, Faisalabad, Pakistan

* Corresponding author (Sorumlu yazar): kashif.zahoor@gcuf.edu.pk

Received (Alınış): 21.12.2015 Accepted (Kabul ediliş): 05.08.2016 Published Online (Çevrimiçi Yayın Tarihi): 27.09.2016

Introduction

The khapra beetle, *Trogoderma granarium* Everts, 1898 (Coleoptera: Dermestidae) is the most serious storage pest of dried plant and animal matter, especially wheat (Hinton, 1945; Szito, 2006). It reduces the quantity, as well as the quality, of stored grain. Severe infestation may cause 5 to 30% weight loss which may increase to 70%, if the beetle is left undisturbed (USDA-APHIS-PPQ, 1983). *Trogoderma granarium* seriously deteriorates the nutritional quality of stored wheat. Infested grain suffers from reduction in proteins, gluten, crude fat, ash, starch, reducing and non-reducing sugars, and mineral components (Girish et al., 1975; Jood et al., 1992; Mason, 2002). Heavily infested grain contains the barbed hairs of larvae, which cause dermal and gastric problems in people handling such grain (Morison, 1925; Pruthi & Singh, 1950; Mason, 2002; Stibick, 2007). Consumption of infested grain can cause vomiting, diarrhea and food refusal especially in the young children (Anonymous, 2001).

The management of stored insect pests mostly relies on the use of synthetic insecticides such as deltamethrin, permethrin, pirimiphos-methyl, chlorpyrifos-methyl and fumigation with methyl bromide or phosphine (White & Leesch, 1995). Such treatments are simple and inexpensive, and even a single application of fumigation can control insect infestation for a substantial period (Daglish & Bengston, 1991; Alsarar et al., 2014). However, widespread use of these chemicals can lead to development of insect resistance (Waiss et al., 1981; Zettler & Cuperus, 1990; White, 1995; Tapondjou et al., 2002), health and environment hazards (Taylor, 1994; Prakash & Rao, 2006; Rahman et al., 2009), toxicity to non-target organisms, and pesticide residues inducing mutagenic and carcinogenic effects on human health (Lee et al., 2004; Isman, 2006). Also, methyl bromide is now banned due to its ozone depleting ability (Taylor, 1994).

Recently, there has been growing interest in the use of botanicals which contain chemicals produced by plants that are repellent, feeding deterrents and disrupting to insect behavior and physiology, and toxic to a number of stored grain insect pests (Hiremath et al., 1997; Verma & Dubey, 1999; Isman et al., 2001; Wheeler & Isman, 2001; Isikber et al., 2006; Isman, 2006; Moreira et al., 2007; Srinivasan, 2008). Plants are a rich source of bioactive chemicals. Both primary as well as secondary plant metabolites can be evaluated against the target pests (Salunke et al., 2009), have insecticidal activity (Dev & Koul, 1997) and are used throughout the world due to their environment friendly nature (Belmain et al., 2001).

During the last few years, weeds are being increasingly investigated for their phytochemical, pharmacological and biological properties (Naqvi & Parveen, 1991; Ahmad et al., 2003 a, b). Weeds are generally considered as unwanted plants and crop pests; however, they have shown insecticidal properties for many insects (Sagheer et al., 2013; Alkan et al., 2015; Vázquez-Covarrubias et al., 2015). Some weeds are poisonous (Shamsuddin, 2001); for example, *Euphorbia prostrata* Aiton (Euphorbiaceae) is a prostrate annual herb found all over India (Singla & Pathak, 1989; Chen et al., 1992), which is used for the treatment of bleeding hemorrhoids, chronic fevers and abdominal diseases as a nerve tonic and blood purifier (Qureshi et al., 2009). It is also used as an antidote for bites of venomous insects (such as wasps and scorpions) and to fight against infertility and painful menstruation to avoid the miscarriage (Schmelzer & Gurib-Fakim, 2008). Likewise, *Chenopodium murale* (L.) S. Fuentes, Uotila & Borsch (Amaranthaceae) is an annual, widespread herbaceous noxious weed about 20-70 cm long, found in more than 43 countries (Zohary, 1966). Nanoparticles synthesized from *C. murale* have been reported to have antioxidant and antibacterial activities (Abdel-Aziz et al., 2014). Moreover, chemical constituents extracted from *C. murale* including essential oils, flavonoids, sterols, alkaloids and coumarins shown antibacterial, antifungal, phytotoxic and insecticidal activities (Naqvi & Parveen, 1991; Ahmad et al., 2003a).

According to the literature, very little work has been carried out to investigate the insecticidal potential of these weed plants on the stored grain insect pests (Moreira et al., 2007). Thus, the present study was designed to evaluate the insecticidal and repellent potential of two weed plants; *E. prostrata* and *C. murale* at different concentrations and exposure intervals for control of stored grain insect pest, *T. granarium*.

Materials and Methods

Bioassays were performed in the Entomology Laboratory, Government College University, Faisalabad to investigate the insecticidal effects of the selected weed plants against larvae of *T. granarium*.

Mass rearing of *Trogoderma granarium*

Trogoderma granarium were reared on healthy wheat grain apparently free from insect infestation in sterilized plastic jars (1 kg capacity) under optimum conditions of temperature and relative humidity, $30\pm 2^{\circ}\text{C}$ and $65\pm 5\%$, respectively. Whole common wheat (*Triticum aestivum* L., cv. Nela, 14% moisture), was used as the culture media. The larvae were sieved through a 2.0 mm aperture sieve. The larvae were counted with the aid of magnifying lens. One hundred beetles were released into labeled 500 ml glass jars having 200 g of sterilized whole wheat and covered with muslin to insect prevent entry or escape. Adults were allowed to mate and lay eggs with the incubator. Homogeneous population was achieved after a time period of 28-35 days. Third instar larvae were then used for the assays (Sagheer et al., 2013).

Preparation of plant extracts

Euphorbia prostrata and *C. murale* were collected from the vicinity of Faisalabad and identified by the Department of Botany, Government College University Faisalabad. Whole plants were cleaned by washing in water and then dried in the shade (Alkan et al., 2015). A grinder was used to crush the plant material into fine powder. The extraction was made by mixing 100 g of ground sieved sample and 300 ml of petroleum ether (40-60%) in the ratio of 1:3 (w/v) and shaking for 24 h using a rotary shaker at 220 rpm. After 24 h, the extract was filtered through Whatman No. 1 filter paper. After filtration, the extracts were stored in clean and airtight bottles at 4°C until used. Concentrations of 10, 20 and 30% (v/v) were prepared using petroleum ether from the stock solutions of each plant (Sagheer et al., 2013; Alkan et al., 2015).

Mortality bioassay (diet incorporation method)

A bioassay was performed to observe the toxic effect of the plant extracts on the larvae of *T. granarium*. The three extract concentrations in petroleum ether were applied on 50 g wheat (5 ml crude plant extract on 50 g wheat @ 0.1 ml/g). For the control, the wheat was only treated with petroleum ether, which was air dried to evaporate the petroleum ether and then poured into 250 ml sterilized plastic jars. Thirty larvae were released in each jar and the jar covered with muslin secured with a rubber band. These jars were placed in incubator at $30\pm 2^{\circ}\text{C}$ and $65\pm 5\%$ RH (Moreira et al., 2007). Each treatment was replicated three times in a completely randomized design. The insects were confirmed dead when there was no response to probing the abdomen with sharp pin. Percentage mortality of the larvae was recorded 2, 4 and 6 days after treatment. Mortality in controls was used to correct the mortality according to Abbot's formula (Abbot, 1925).

Repellency bioassay

In another bioassay, the repellent effect of the plant extracts was checked against *T. granarium* larvae by using a modification of the area preference method described by McDonald et al. (1970). For repellency test, 80 mm diameter Whatman No. 1 filter paper was cut into two equal halves. The three extract concentrations in petroleum ether were applied separately to one half of the filter paper placed in a Petri dish (100 x 15 mm) using 10 μl micropipette and petroleum ether alone was applied to the other half (10 $\mu\text{l}/\text{cm}^2$ on treated area). After air-drying for 10 min, each treated half of the filter paper was attached lengthwise to untreated half using adhesive tape and placed in a Petri dish. Twenty third-instar larvae of *T. granarium* were released separately at the center of both halves in each petri dish. Petri dishes were covered with a lid to prevent the escape of test insects and kept under controlled conditions ($30\pm 2^{\circ}\text{C}$ and $65\pm 5\%$ RH). Each treatment was replicated three times and counts of the larvae on each filter paper disk were made after 24, 48 and 72 h. Wheat grain (0.5 g) were also provided on both sides in order to avoid the mortality due to starvation.

Percent repellency (RP) was calculated by using the following formula:

$$\text{PR} = [(\text{NC}-\text{NT}) / (\text{NC}+\text{NT})] \times 100$$

Where, NC= number of larvae present on control half and NT= number of larvae present on treated half.

Growth regulatory effect of plant extracts on the larvae of *Trogoderma granarium*

The larval emergence and inhibition of *T. granarium* in F_1 generation were recorded in order to investigate the growth regulatory effect of the plant extracts on *T. granarium*. The wheat grains were sterilized and the three extract concentrations in petroleum ether were applied by spraying and mixing

(0.1 ml/g) on wheat grain including a control treatment. The solvent was allowed to evaporate and 50 g treated grain was placed separate plastic jars (250 ml). Thirty third-instars were released into each jar and then incubated under optimum conditions ($30\pm 2^{\circ}\text{C}$ and $65\pm 5\%$ RH). The mean emergence and percent inhibition of F_1 larvae was recorded after 35 days (Sagheer et al., 2013; Alkan et al., 2015).

Statistical analysis

The data of corrected mortality, repellency and growth regulation was subjected to ANOVA using Statistica 13.0 for Windows. The means were separated using Tukey's HSD test with $P < 0.05$ considered statistically significant (Tapondjou et al., 2002; Sagheer et al., 2013; Pandir & Bas, 2016).

Results and Discussion

Mortality of *Trogoderma granarium* larvae

Mortalities of *T. granarium* larvae were observed at various exposure time and concentrations of both *E. prostrata* and *C. murale* extracts in petroleum ether. The comparison of mean mortality rates of the third instar larvae of *T. granarium* induced by various concentrations of *E. prostrata* extract during 6 days of exposure period is shown in Table 1. There were significant differences in percent mortality of *T. granarium* larvae at the three concentrations of *E. prostrata* extract after 6 days ($F=3.65$; $P<0.05$). The highest mean mortality (20) was found at 30% *E. prostrata* extract, followed by mortality of 16 and 9% at 20 and 10% concentrations, respectively. The 30% concentration of *E. prostrata* extract resulted in significantly higher mortality of *T. granarium* larvae than at 10%, while the mortality at 20 and 30% *E. prostrata* extract were not statistically different. These results indicated that the larval mortality increased with increasing of extract concentration.

Table 1. Mean percent mortalities of *Trogoderma granarium* larvae exposed to different concentrations of *Euphorbia prostrata* extract for 6 days exposure time

Concentration (%)	Mortality (%) \pm SE
10	9 \pm 4.9 b*
20	16 \pm 6.2 ab
30	20 \pm 7.1 a

* According to Tukey's HSD test, means with same lowercase letter are not significantly different at $P=0.05$.

Table 2 shows that exposure time had a significant effect on the mortality of *T. granarium* larvae exposed to 30% *E. prostrata* extract ($F=39.3$; $P<0.05$). The highest mortality (35%) was found after 6 days while the lowest mortality (0.7%) was observed after 2 days. The larval mortality after 6 days was significantly higher than after 2 and 4 days, but there was no significant difference between mortalities after 2 and 4 days. The results indicated that the larval mortality increased with increasing of exposure time.

Table 2. Mean percent larval mortalities of *Trogoderma granarium* exposed to 30% concentration of *Euphorbia prostrata* extract for different exposure intervals

Exposure interval (days)	Mortality (%) \pm SE
2	0.7 \pm 0.49 b*
4	9 \pm 2.8 b
6	35 \pm 4.9 a

* According to Tukey's HSD test, means with same lowercase letter are not significantly different at $P=0.05$.

The mean mortality rates of *T. granarium* larvae exposed to *C. murale* extract at the three concentrations are shown in Table 3. The highest mean mortality (25%) was observed at 30% concentration and the lowest at 10% (14%). Extract concentration had significant effect on mortality of *T. granarius* larvae ($F=6.85$; $P<0.05$). The mean mortality at 30% concentration was significantly higher than at 10%. However, the mortalities at 20 and 30% *C. murale* extract were not statistically different.

Table 3. Mean percent larval mortalities of *Trogoderma granarium* exposed to different concentrations of *Chenopodium murale* extract for 6 days exposure time

Concentration (%)	Mortality (%) \pm SE
10	14 \pm 4.6 b*
20	20 \pm 6.0 ab
30	25 \pm 7.4 a

* According to Tukey's HSD test, means with same lowercase letter are not significantly different at P=0.05.

Mean percent mortality of *T. granarium* larvae exposed to 30% *C. murale* extract for different times are given in Table 4. The larval mortalities were 0, 19 and 40% at 2, 4 and 6 days after treatment, respectively. There were significant differences in larval mortalities between the exposure periods (F=85.39; P<0.05). The larval mortality after 6 days was significantly higher than those after 2 and 4 days. The results showed that the insecticidal potential of *C. murale* extract increased with the increased exposure time. It is concluded that increase in both concentration of *C. murale* extract and exposure period resulted in the increase of mortality of *T. granarium* larvae.

Table 4. Mean percent mortalities of *Trogoderma granarium* larvae exposed to 30% concentration of *Chenopodium murale* extract for different exposure times

Exposure interval (days)	Mortality (%) \pm SE
2	12 \pm 0.87 c*
4	19 \pm 2.5 b
6	40 \pm 4.1 a

* According to Tukey's HSD test, means with same lower-case letter(s) are not significantly different at P=0.05.

Repellence of *Trogoderma granarium* larvae

The repellency of different concentrations of *E. prostrata* extract against *T. granarium* larvae is shown in Table 5. *Euphorbia prostrata* extracts exhibited significant repellent effect at all treatment concentrations. At 10% extract, only 39% repellency was observed, whereas at 20 and 30% extracts gave 68% and 88% repellency, respectively. The statistical analysis indicated 30% extract resulted in higher percentage of larval repellency than the two lower extract concentrations (F=23.68; P<0.05; Table 5).

Table 5. Mean percent larval repellency in *Trogoderma granarium* exposed to different concentrations of *Euphorbia prostrata* extract for 72 h

Concentration (%)	Repellency (%) \pm SE
10	39 \pm 8.2 c*
20	68 \pm 6.2 b
30	88 \pm 4.0 a

* According to Tukey's HSD test, means with same lowercase letter(s) are not significantly different at P=0.05.

The repellence of *T. granarium* exposed to *E. prostrata* extract at various exposure intervals at 30% concentration is shown in Table 6. The highest mean repellency was observed (80%) after 24 h exposure, followed by 66 and 49% after 48 and 72 h, respectively. The repellency at all exposure times were significantly different from each other (F=9.5; P<0.05). The repellency after 24 h was significantly higher than after 72 h. Overall the results indicated that larval repellency decreased with increasing exposure period.

Table 6. Mean percent larval repellency in *Trogoderma granarium* exposed to 30% concentration of *Euphorbia prostrata* extract for different exposure intervals

Exposure interval (hours)	Repellency (%) ± SE
24	80 ± 7.6 a*
48	66 ± 8.2 ab
72	49 ± 9.3 b

* According to Tukey's HSD test, means with same lower-case letter(s) are not significantly different at P=0.05.

The repellency of *C. murale* extract applied at different concentrations on *T. granarium* larvae is summarized in Table 7. At 10% extract, 40% exposed larvae were repelled, which it increased to 61% with 20% extract. There was no difference in repellency for 10% and 20% extracts. However, 30% extract gave a significantly higher repellency (87%) for 10 and 20% (F=13.14; P<0.05).

Table 7. Mean percent larval repellency in *Trogoderma granarium* exposed to different concentration of *Chenopodium murale* extract for 72 h

Concentration (%)	Repellency (%) ± SE
10	40 ± 7.1 b*
20	61 ± 8.4 b
30	87 ± 5.3 a

* According to Tukey's HSD test, means with same lowercase letter are not significantly different at P=0.05.

Repellence of *T. granarium* larvae exposed to *C. murale* extract at 30% concentration for different exposure intervals is shown in Table 8. After 24 h, the highest repellency was observed (76%), followed by 66 and 47% repellency after 48 and 72 h, respectively. Statistical analysis indicated that the exposure time had significant effect on repellency of *T. granarium* larvae (F=5.18; P<0.05). Repellency after 24 h was significantly higher than after 72 h. The results indicated the time dependent efficacy of *C. murale* extract, showing decreased repellency with the increase of exposure duration.

Table 8. Mean percent larval repellency in *Trogoderma granarium* exposed to 30% concentration of *Chenopodium murale* extract for different exposure intervals

Exposure interval (hours)	Repellency (%) ± SE
24	76 ± 7.7 a*
48	66 ± 9.4 ab
72	47 ± 9.1 b

* According to Tukey's HSD test, means with same lowercase letter are not significantly different at P=0.05.

Effect of extracts of *Euphorbia prostrata* and *Chenopodium murale* on progeny of *Trogoderma granarium*

The mean larval emergence and inhibition of *T. granarium* in F₁ generation at various concentrations of *E. prostrata* and *C. murale* were recorded after 35 days of infestation (Table 9). The results indicated that lower number of larvae emerged in F₁ generation at three concentrations of both plant extracts as compared to the control. Moreover, there was considerable difference observed in the number of larvae in F₁ generation of both extracts. The lowest mean number of larvae (53) and highest larval inhibition (64%) in F₁ generation was observed in 30% *C. murale* extract treatment followed by *E. prostrata* (60), also at 30%, as compared to 101 and 112 larvae with 10% *C. murale* and *E. prostrata* extracts, which induced 32 and 24% larval inhibition, respectively. The highest number of larvae (149) in F₁ generation was obtained when the wheat grains were left untreated for 35 days (Table 9).

Table 9. Mean number of emerged larvae and their percent inhibition in F1 progeny of *Trogoderma granarium* following treatment with *Euphorbia prostrata* and *Chenopodium murale* whole plant extracts at different concentrations

Concentration (%)	F1 progeny ± SE			
	<i>Euphorbia prostrata</i>		<i>Chenopodium murale</i>	
	Mean no. of larvae	Percent larval inhibition	Mean no. of larvae	Percent larval inhibition
10%	112 ± 5.9 b*	24 ± 4.0 b	101 ± 5.2 b	32 ± 3.5 b
20%	94 ± 4.7 b	37 ± 3.2 b	83 ± 7.5 b	44 ± 5.1 b
30%	60 ± 3.1 c	60 ± 2.1 c	53 ± 3.3 c	65 ± 2.2 c
Control	149 ± 7.8 a		149 ± 7.8 a	

* According to Tukey's HSD test, means with same lowercase letter are not significantly different at P=0.05.

Table 9 also shows inhibition of *T. granarium* at the three concentrations of both extracts. The numbers of emerged larvae at the higher concentrations were significantly lower than at the lower concentrations and, also the inhibition increased with increased concentration of both *C. murale* and *E. prostrata* extracts. The statistical analysis indicated that both *C. murale* (F=19.19; P<0.05) and *E. prostrata* (F=31.84; P<0.05) extracts had significant effects on emergence and inhibition in F₁ generation of *T. granarium*.

Trogoderma granarium has been considered the worst insect pest of stored grain because it causes huge quantitative and qualitative losses to stored commodities. Synthetic pesticides have been the main control strategy. However, due to their adverse effects on humans and the environment, there is a need to find a safe and environmentally friendly method of control. Thus, many investigations are being carried out around the world to evaluate the pesticidal and repellent properties of plant extracts. The present research work is a continuation to this research to discover the insecticidal potential of weeds, which are commonly considered as unwanted, and pests of land. The results obtained indicated that *C. murale* extract was most effective resulting in the highest mortality (25%) of *T. granarium* larvae after 6 days of exposure, followed by 20% larval mortality (*E. prostrata*) at 30% concentration. The lowest percent mortality (9%) was obtained after 2 days with 10% *E. prostrate* extract. It was clear that larval mortality is directly related to applied concentration and exposure time. Similarly the highest larval repellency was obtained at the 30% *E. prostrata* (88%) and *C. murale* (87%) extracts compared to 39 and 40% at the lowest concentrations, respectively, showing no difference in the efficacy of both plants. In relation to treatment time, *E. prostrata* and *C. murale* repelled higher numbers of larvae after 24 h, 80 and 76%, respectively. The repellency of *E. prostrata* and *C. murale* extracts was decreased with the increase of exposure time. These repellency results revealed no difference in the efficacy of the two plants, giving almost same results. However, a negative relationship between the larval repellency and exposure interval was found.

This pattern is in line with the previous findings (Odeyemi & Ashamo, 2005) that the plant extracts become more toxic increased dose and exposure time. The efficacy of various plant extracts in reducing reproduction and increasing mortality of adult stored grain insects was evaluated by Tapondjou et al. (2002). They used powders and essential oils from leaves of *Dysphania ambrosioides* (L.) Mosyakin & Clemants against six stored grain insect pests; *Callosobruchus chinensis* L., 1758, *Callosobruchus maculatus* Fab., 1775, *Acanthoscelides obtectus* (Say, 1831), *Sitophilus granaries* L., 1758, *Sitophilus zeamais* Motschulsky, 1855 and *Prostephanus truncates* (Horn, 1878). The powdered dry leaves were mixed with grains at different rates ranging from 0.05 to 6.4% (w/w). A dosage of 0.4% killed more than 60% of *C. chinensis* and *C. maculatus* two days after treatment, while a dosage of 6.4% caused total mortality of *S. granarius* and *S. zeamais* within the same exposure time. Moreover, all the concentrations inhibited F1 progeny production and adult emergence of the tested insects. A concentration of 0.2 µl/cm² of the essential oil almost killed all the tested insects (80-100%). Consistently, 10µl/cm² of extracts was used instead of using essential oils and gave up to 60% progeny inhibition. The insecticidal efficacy of essential oil from *Foeniculum vulgare* Mill.,

Teucrium polium L. and *Satureja hortensis* L. was studied by Heydarzade & Moravvej (2012) against adult *C. maculatus* using a contact toxicity assay. They found essential oil from *S. hortensis* as highly persistent, while *T. polium* showed lower persistency. An increase in mortality with an increase in concentration was observed, which is consistent to the present study. Subsequently, Hasan et al. (2014) used essential oils of four plants, viz., *Azadirachta indica* A. Juss., *Curcuma longa* L., *Nigella sativa* L. and *Piper nigrum* L., for repellency and toxicity studies on *T. granarium*. Concentrations of 5, 10, 15 and 20% were used to test the insecticidal potential to protect stored wheat. The highest mean repellency (90%) was obtained with *A. indica* at 20% and the lowest repellency (26%) with *P. nigrum* at 5%. The highest mortality (31%) was recorded with *A. indica*, whereas lowest mortality (15%) was obtained after 30 days at 20%. The study indicated significant *T. granarium* mortality and feeding deterrence in larvae feeding on wheat grain indicating that these essential oils could be used to develop new botanical insecticides.

Derbalah (2012) tested seven plant extracts [*Argyranthemum frutescens* (L.) Sch.Bip., *Bauhinia purpurea* L., *Caesalpinia gilliesii* (Wallich ex Hook.) Wallich ex D. Dietr., *Cassia fistula* L., *Euonymus japonicus* L., *Senna alexandrina* Mill. and *Thespesia populnea* var. *acutiloba* Bak.] against *T. granarium* and found that all the extracts induced significant mortality and reduced the F₁ progeny emergence. However, *S. alexandrina* was found to be the most effective of these botanical extracts. In other work, Pacual-Villalobos (1998) reported 14% larval mortality in *T. granarium* treated with Neem extract (*A. indica*) compared to 7% with *P. nigrum* extract. Recently, Pandir & Bas (2016) tested the essential oils from basil (*Ocimum basilicum* L.), paprika (*Capsicum annum* L.), peppermint (*Mentha x piperita* L.) and rosemary (*Rosmarinus officinalis* L.) at the concentration of 0.1, 1, 5, 10, 20, 50 and 100 µl/L for the control of different stages of *Ephesia kuehniella* Zeller, 1879. Among different life stages of *E. kuehniella*, the larval stage was found to be the most tolerant to essential oils. Overall it was concluded that insecticidal potential of the essential oils of these plants increased with increased application concentration.

Similarly, Al-Moajel (2004) reported the dose dependent mortality and progeny reduction in *T. granarium* while testing the botanical powders from eleven different plants [*Albizia lebbek* (L.) Benth., *Allium cepa* L., *Allium cepa* var. *ascalonicum* Don., *Capsicum frutescens* L., *Carthamus tinctorius* L., *Delonix regia* (Boj. ex Hook.) Raf., *Eruca sativa* Mill., *Lawsonia inermis* L., *Mesua ferrea* L., *Raphanus sativus* L. and *Vachellia farnesiana* (L.) Wight & Arn.]. *Capsicum frutescens* induced the highest mortality (77-85%) of *T. granarium*, while F₁ progeny was significantly reduced by *R. sativus* (71%), *C. tinctorius* (67%), *C. frutescens* (59%), *A. cepa* var. *ascalonicum* (53%) and *L. inermis* (47%) powders. It was concluded that *C. frutescens* and *L. inermis* showed significant effect on mortality of both adults and larvae of *T. granarium* and also significantly reduced the F₁ progeny. Sagheer et al. (2013) also evaluated the repellent potential of four medicinal plants extracted in acetone against *T. granarium* larvae, resulting in the 55, 52, 51 and 47% larval repellency from *Nicotiana tobaccum* L., *Peganum harmala* L., *Salsola baryosma* (Schult.) Dandy and *Saussurea costus* (Falc.) Lipsch. extracts at 20% concentration, respectively. They also concluded that the repellent effect of plant extracts was increased with the increase in application concentration.

Alkan et al. (2015) also investigated the antifeedant activity and growth inhibition effects of *Achillea millefolium* L., *Heracleum platytaenium* Boiss. and *Humulus lupulus* L. extracts against third instar larvae of Colorado potato beetle (*Leptinotarsa decemlineata* Say, 1824). They found that a concentration of 50 g/L of all plant extracts induced antifeedant activity. It was suggested that *H. platytaenium* and *H. lupulus* extracts were excellent antifeedants and larval growth inhibitors. Although, there are many studies reporting the use of plant extracts against stored grain insect pests, the plant extracts used in the present study has not previously been evaluated for the control of *T. granarium* larvae.

Conclusion

The results demonstrate that extracts of *E. prostrata* and *C. murale* have insecticidal, repellent and progeny reduction potential against *T. granarium*. Overall, the results indicated that petroleum ether extract of *C. murale* had higher insecticidal activity than the extract of *E. prostrata*. More research work on weeds, so called the pest of crops, is needed for their use in stored grain insect pest management programs.

Acknowledgments

The authors thank Stored Grain Research, Training and Storage Management Cell of Department of Entomology, University of Agriculture Faisalabad, Pakistan for the technical assistance provided during the development of this study.

REFERENCES

- Abbott, W. S., 1925. A method of computing the effectiveness, of an insecticide. *Journal of Economic Entomology*, 18: 265-267.
- Abdel-Aziz, M. S., M. S. Shaheen, A. A. El-Nekeety & M. A. Abdel-Wahhab, 2014. Antioxidant and antibacterial activity of silver nanoparticles biosynthesized using *Chenopodium murale* leaf extract. *Journal of Saudi Chemical Society*, 18 (4): 356-363.
- Ahmad, B., Q. Jan, S. Bashir, M. Nisar, F. Shaheen & M. Ahmad, 2003a. Pharmacological and biological investigations of *Chenopodium murale* Linn. *Asian Journal of Plant Sciences*, 2: 1107-1111.
- Ahmad, B., Q. Jan, S. Bashir, M. I. Choudhary & M. Nisar, 2003b. Phytochemical evaluation of *Chenopodium murale* Linn. *Asian Journal of Plant Sciences*, 2 (15-16): 1072-1078.
- Alkan, M., A. Gökçe & K. Kara, 2015. Antifeedant activity and growth inhibition effects of some plant extracts against larvae of Colorado potato beetle [*Leptinotarsa decemlineata* Say (Col: Chrysomelidae)] under laboratory conditions. *Turkish Journal of Entomology*, 39 (4): 345-353. DOI: 10.16970/ted.35600.
- Al-Moajel, N. H., 2004. Testing some various botanical powders for protection of wheat grains against *Trogoderma granarium* Everts. *Journal of Biological Sciences*, 4 (5): 592-597.
- Alsarar, A. S., H. I. Hussein, Y. Abobakr, A. E. Bayoumi & M. T. Alotaibi, 2014. Fumigant toxicity and antiacetylcholinesterase activity of Saudi *Mentha longifolia* and *Lavandula dentata* Species against *Callosobruchus maculatus* (F.) (Coleoptera: Bruchidae). *Turkish Journal of Entomology*, 38 (1): 11-18.
- Anonymous, 2001. Bad bugs. Insects that can cause health problems. *Pest Notes*, 3 (8): 20-21.
- Belmain, S. R., G. E. Neal, D. E. Ray & P. Golop, 2001. Insecticidal and vertebrate toxicity associated with ethnobotanicals used as postharvest protectants in Ghana. *Food and Chemical Toxicology*, 39: 287-291.
- Chen, L., R. Chen & K. Wai, 1992. Constituents of tannins from *Euphorbia prostrata*. *Ait China Journal of Chinese Material Medica*, 17 (4): 225-226.
- Daglish, G. & M. Bengston, 1991. Grain moisture and the activity of protectants. *GASGA Newsletter* 15: 18, In: Groupior Assistance on Systems relating to Grain After-harvest (GASGA), Montpellier, Chatham and Hamburg.
- Derbalah, A. S., 2012. Efficacy of some botanical extracts against *Trogoderma granarium* in wheat grains with toxicity evaluation. *The Scientific World Journal*, April 19, 2012: 639854, 1-9. DOI:10.1100/2012/639854.
- Dev, S. & O. Koul. 1997. *Insecticides of natural origin*. New York: Harwood Academic. 352pp.
- Girish, G. K., A. Kumar & S. K. Jain, 1975. Part - VI: assessment of the quality loss in wheat damaged by *Trogoderma granarium* Everts during storage. *Bulletin of Grain Technology*, 13(1): 26-32.
- Hasan, M., M. Sagheer, M. R. Q. Ali, C. M. Hanif & H. Anwar, 2014. Evaluation of some plant essential oils as repellent and toxicant against *Trogoderma granarium* (Everts) (Coleoptera: Dermestidae). *Journal of Global Innovation in Agricultural and Social Sciences*, 2: 65-69.
- Heydarzade, A. & G. Moravvej, 2012. Contact toxicity and persistence of essential oils from *Foeniculum vulgare*, *Teucrium polium* and *Satureja hortensis* against *Callosobruchus maculatus* (Fabricius) adults (Coleoptera: Bruchidae). *Turkish Journal of Entomology*, 36 (4): 507-518.
- Hinton, H. E., 1945. A monograph of the beetles associated with stored products. *British Museum of Natural History*, 1: 387-395.
- Hiremath, I. G., A. Youngjoon, K. Soonl, Y. J. Ahn & S. I. Kim, 1997. Insecticidal activity of Indian plant extracts against *Nilaparvata lugens* (Homoptera: Delphacidae). *Applied Entomology and Zoology*, 32 (1): 159-166.
- Isikber, A. A., M. H. Alma, M. Kanat & A. Karci, 2006. Fumigant toxicity of essential oils from *Laurus nobilis* and *Rosmarinus officinalis* against all life stages of *Tribolium confusum*. *Phytoparasitica*, 34: 167-177.

- Isman, M. B., 2006. Botanical insecticides, deterrents, and repellents in modern agriculture and an increasingly regulated world. *Annual Review of Entomology*, 51: 45–66.
- Isman, M. B., A. J. Wan & C. M. Passreiter, 2001. Insecticidal activity of essential oils to the tobacco cutworm, *Spodoptera littura*. *Fitoterapia*, 72: 65-68.
- Jood, S., A. C. Kapoor & R. Singh, 1992. Mineral contents of cereal grains as affected by storage and insect infestation. *Journal of Stored Products Research*, 28(3): 147-151.
- Lee, B. H., P. C. Annis, F. Tumaalii & W. S. Choi, 2004. Fumigant toxicity of essential oils from the Myrtaceae family and 1,8-cineol against 3 major stored-grain insects. *Journal of Stored Products Research*, 40: 553-564.
- Mason, L. J., 2002. Khapra Beetle. *Stored Grain Management*. Country Journal Publishing Co., Decatur, Illinois. Grainnet. 2002. (Web page: http://www.grainnet.com/articles/Khapra_Beetle-16160.html)
- McDonald, L., R. Guy & R. Speirs, 1970. Preliminary evaluation of new candidate materials as toxicants, repellents and attractants against stored product insects. *Marketing Research Report*, p. 882.
- Moreira, M. D., M. C. Picanço, L. C. D. A. Barbosa, R. N. C. Guedes, M. R. D. Campos, G. A. Silva & J. C. Martins, 2007. Plant compounds insecticide activity against Coleoptera pests of stored products. *Pesquisa Agropecuária Brasileira*, 42 (7): 909-915.
- Morison, G. D., 1925. The Khapra beetle (*Trogoderma granarium* Everts). *Proceedings of the Royal Physical Society, Edinburgh*, 21: 10-13.
- Naqvi, S. N. H. & F. Parveen, 1991. Toxicity and Residual Effect of *Nerium indicum* crude extract as compared with coopex against adults of *Tribolium castaneum*. *Pakistan Journal of Entomology*, 6: 35-44.
- Odeyemi, O. O. & M. O. Ashamo, 2005. Efficacy of neem plant (*Azadirachta indica*) extracts in the control of *Trogoderma granarium*, a pest of stored groundnuts. *Journal of Plant Disease Protection*, 112 (6): 586- 593.
- Pacual-Villalobos, M. J., 1998. Repelencia inhibición del crecimiento y toxicidad de extractos vegetales en larvas de *Tribolium castaneum* Herbst. (Coleoptera:Tenebrionidae). *Boletín de sanidad vegetal, Plages*, 24: 143-154.
- Pandir, D. & H. Baş, 2016. Compositional analysis and toxicity of four plant essential oils to different stages of Mediterranean flour moth, *Ephesia kuehniella* Zeller (Lepidoptera: Pyralidae). *Turkish Journal of Entomology*, 40 (2): 185-195.
- Prakash, A. & J. Rao, 2006. Exploitation of newer botanicals as rice grain protectants against Angonmois grain moth *Sitotroga cerealella* Oliv. *Entomon*, 31: 1–8.
- Pruthi, H. S. & M. Singh, 1950. Pests of stored grain and their control. *Indian Journal of Agricultural Sciences*, 18: 1-88.
- Qureshi, R., A. Waheed, M. Arshad & T. Umbreen, 2009. Medico-ethnobotanical inventory of Tehsil Chakwal, Pakistan. *Pakistan Journal of Botany*, 41 (2): 529-538.
- Rahman, M. M., W. Islam & K. N. Ahmed, 2009. Functional response of the predator *Xylocoris flavipes* to three stored product insect pests. *International Journal of Agriculture and Biology*, 11: 316–320.
- Sagheer, M., M. Hasan, A. Majid, Q. Ali, M. I. Shahid & K. Ali, 2013. Repellent effect of four medicinal plant extracts to *Trogoderma granarium* Everts (Coleoptera: Dermestidae). *Journal of Global Innovation in Agricultural and Social Sciences*, 1: 9-11.
- Salunki, B. K., K. Parkash, K. S. Vishwakarma & V. L. Maheshwari, 2009. Plant metabolites: an alternative and sustainable approach towards post harvest pest management in pulses. *Physiology and Molecular Biology of Plants*, 15 (3): 185–197.
- Schmelzer, G. H. & A. Gurib-Fakim, 2008. *Plant resources of Tropical Africa (Program). Plant resources of tropical Africa: medicinal plants 1, Vol. 11, PROTA Foundation, Wageningen.*
- Shamsuddin, T., 2001. Weed control technology of wheat. *Industry and Economy*, Feb. 19-25. (Web page: <http://www.pakistaneconomist.com/issue2001/issue8/i&e5.htm>).
- Singla, A. K. & K. Pathak, 1989. Anti-inflammatory studies on *Euphorbia prostrata*. *Journal of Ethnopharmacology*, 27: 55–61.
- Srinivasan, G., 2008. Efficacy of certain plant oils as seed Protectant against pulse beetle, *Callosobruchus chinensis* L. on pigeon pea. *Pesticide Research Journal*, 20: 13-15.

- Stibick, J., 2007. New Pest Response Guidelines: Khapra Beetle. USDA–APHIS–PPQ–Emergency and Domestic Programs, Riverdale, Maryland. (Web site: http://www.aphis.usda.gov/import_export/plants/manuals/online_manuals.shtml).
- Szito, A., 2006. *Trogoderma granarium* (insect). Global Invasive Species Database. Invasive Species Specialist Group (ISSG), IUCN Species Survival Commission, 2006. (Web page:: <http://www.issg.org/>).
- Tapondjou, L. A., C. Adler, H. Bouda & D. A. Fontem, 2002. Efficacy of powder and essential oil from *Chenopodium ambrosioides* leaves as post-harvest grain protectants against six-stored product beetles. *Journal of Stored Products Research*, 38 (4): 395- 402.
- Taylor, R. W. D., 1994. Methyl bromide: Is there any future for this fumigant? *Journal of Stored Product Research*, 30: 253- 260.
- USDA–APHIS–PPQ, 1983. Khapra Beetle. Pests not known to occur in the United States or of limited distribution. APHIS, 81-43 (30): 1-11.
- Vázquez-Covarrubias, D. A., A. Jiménez-Pérez, F. Castrejón-Ayala, R. Figueroa-Brito & R. M. Belmont, 2015. Effects of five species of *Chenopodiaceae* on the development and reproductive potential of *Copitarsia decolora* (Lepidoptera: Noctuidae). *Florida Entomologist*, 98 (1): 80-85.
- Verma, J. & N. K. Dubey, 1999. Prospectives of botanical and microbial products as pesticides of tomorrow. *Current Science*, 76: 172-179.
- Waiss, A. C. J., B. G. Chem, C. A. Elligen, D. L. Dryer, R. G. Binder & R. C. Guedner, 1981. Insect growth inhibitor in crop plants. *Bulletin of the Entomological Society of America*, 27: 217-221.
- Wheeler, D. A. & M. Isman, 2001. Antifeedant and toxic activity off *Trichilia americana* extract against the larvae of *Spodoptera litura*. *Entomologia Experimentaliset Applicata*, 98: 9-16. ISSN 0013-8703.
- White, N. D. G. & I. G. Leesch, 1995. "Chemical Control 287-330". In: *Integrated Management of Insects in Stored Products*; (Eds.: B. Sub-ramanyam & D. W. Hagstrum), Marcel Dekker, New York, NY, USA.
- White, N. D. G., 1995. "Insects, Mites and Insecticides in Stored Grain Ecosystems, 123-168". In: *Stored Grain Ecosystem*, (Eds.: D. S. Jayus, N. D. G. White & W. E. Vir), Marcel Dekkar, New York.
- Zettler, J. L. & G. W. Cuperus, 1990. Pesticide resistance in *Tribolium castaneum* (Coleoptera: Tenebrionidae) and *Rhyzopertha dominica* (Coleoptera: Bostrichidae) in wheat. *Journal of Economic Entomology*, 83: 1677-1681.
- Zohary, M., 1966. *Flora Palaestina*. Part One. Jerusalem, Israel: The Israel Academy of Sciences and Humanities, 186pp.