Tarım Bilimleri Dergisi
Tar. Bil. Der.
Dergi web sayfası:
www.agri.ankara.edu.tr/dergi

Journal of Agricultural Sciences
Journal homepage:
www.agri.ankara.edu.tr/journal

Effects of Diatomaceous Earth on the Mortality and Progeny Production of *Rhyzopertha dominica* (Coleoptera: Bostrychidae)

Gülay BERİŞ[a], Ahmet Güray FERİZLİ[a], Mevlüt EMEKÇİ[a]

[a] *Ankara University, Faculty of Agriculture, Department of Plant Protection, 06110, Ankara, TURKEY*

ARTICLE INFO

Research Article — Crop Production DOI: 10.1501/Tarimbil_0000001160
Corresponding author: Ahmet Güray FERİZLİ, e-mail: ferizli@agri.ankara.edu.tr, Tel: +90(312) 596 11 36
Received: 13 May 2011, Received in revised form: 10 July 2011, Accepted: 19 July 2011

ABSTRACT

Laboratory experiments were conducted to assess the insecticidal effect of the diatomaceous earth (DE) formulation Insecto® against adult stage of *Rhyzopertha dominica* (F.) at two levels of relative humidity i.e. 40 and 55%, and three exposure periods of 7, 14, and 21 days. Test insects were placed in vials containing 40 g of soft winter wheat mixed with 0.25, 0.50, 1.00, 1.50 and 2.00 g kg^{-1} DE. After respective exposure periods, mortality in all dose rates except 0.25 and 0.50 g kg^{-1} was found to be significantly different from mortality in control. After each exposure interval, dead and live insects were counted and removed, and then the vial containing wheat was returned to corresponding humidity chamber for 8 weeks until F_1 adults emerged. For F_1 production, applications at all dose rates were significantly different from control group. Mortality regardless of dosage for 7, 14 and 21 days exposure intervals were between 27.67– 33.40 %. Despite the fact that mortality increased with the increasing dose rate, total mortality was not obtained even at the highest rate of DE. F_1 production decreased with the increasing dose rates for both r.h. conditions. Containment of population was achieved at 1.50 g kg^{-1} of DE for 7 days of exposure period and at 2.00 g kg^{-1} of DE for 14 and 21 days of exposure periods. For each exposure intervals, F_1 production of *R. dominica* decreased with the increasing dose rates of DE. Mortality regardless of exposure interval was 59% at 1.50 g kg^{-1} of DE and 75% at 2.00 g kg^{-1} of DE.

Keywords: Insecto®; *Rhyzopertha dominica*; F_1 adult production; Mortality

Diyatom Toprağının *Rhyzopertha dominica* (Coleoptera: Bostrychidae)'nın Ölüm ve Yavru Verimi Üzerine Etkileri

ESER BİLGİSİ

Araştırma Makalesi — Bitkisel Üretim
Sorumlu Yazar: Ahmet Güray FERİZLİ, e-mail: ferizli@agri.ankara.edu.tr, Tel: +90(312) 596 11 36
Geliş tarihi: 13 Mayıs 2011, Düzeltmelerin gelişi: 10 Temmuz 2011, Kabul: 19 Temmuz 2011

ÖZET

Bu araştırmada diyatom toprağı olarak Insecto®'nun *Rhyzopertha dominica* (F.)'nın ergin evresine öldürücü etkisi %40 ve %55 orantılı nem koşullarında 7, 14 ve 21 günlük üç farklı uygulama süresinde etkinliği belirlenmiştir.

Ergin böcekler içerisinde 0.25, 0.50, 1.00, 1.50 ve 2.00 g kg^{-1} dozda Insecto® karıştırılmış 40 g yumuşak kışlık buğday bulunan kaplara bırakılmıştır. Üç farklı uygulama süresinde belirlenen ölümler (0.25 ve 0.50 g kg^{-1}'daki hariç) kontrol grubunda belirlenen ölüm düzeyinden önemli düzeyde farklı bulunmuştur. Her bir uygulama süresi sonunda, ölü ve canlı böcekler sayılmış ve kaptan uzaklaştırılmış; ardından buğday tekrar kaba alınarak deneme süresince tutulduğu nem kabinine alınmış ve burada F_1 erginleri çıkıncaya kadar 8 hafta tutulmuştur. F_1 verimi uygulanan tüm dozlarda kontroldekinden önemli düzeyde farklı bulunmuştur. Doz dikkate alınmaksızın ölümler 7, 14 ve 21 günlük uygulamada %27.67– 33.40 arasında olmuştur. Ölümler artan doz oranı ile birlikte artmış olmasına rağmen, mutlak ölüm en yüksek dozda da gerçekleşmemiştir. F_1 verimi her iki nem koşulunda da artan doz ile birlikte azalmıştır. Özet olarak, Insecto® uygulaması F_1 veriminde düşüşe neden olmuş ve popülasyon 7 günlük uygulamada 1.50 g kg^{-1} ve 14 ve 21 günlük uygulamada ise 2.00 g kg^{-1} diyatom toprağı dozunda sabit düzeyde kalmıştır. Süre dikkate alınmaksızın ölümler 1.50 g kg^{-1} dozunda %59 ve 2.00 g kg^{-1} dozunda ise %75 düzeyinde gerçekleşmiştir.

Anahtar sözcükler: Insecto®; *Rhyzopertha dominica*; F_1 ergin üretimi; Ölüm

1. Introduction

Long-term storage of grains inevitably necessitates taking action against pests. Although large scale storage efforts are mainly concentrated on the use of fumigants due to economic and practicability reasons, environmental considerations have limited the use of common fumigants. Methyl bromide, for example, has been banned by Montreal Protocol worldwide due to its ozone depleting properties, while many pests have already developed phosphine-resistant strains due to improper fumigations (Zettler & Arthur 2000). Additionally, sulphuryl fluoride is now being considered as a gas to the greenhouse effect and global warming (Mühle et al 2009). Moreover, few residual insecticides are currently registered for use in grain storage; however, they are also subject to health, environmental, and pest resistance considerations (Subramanyam & Hagstrum 1995).

Non-chemical alternatives, which are effective, environmentally sound, economically feasible, and user friendly, have gained an important position in the protection of stored grains. Diatomaceous earth (DE) that causes insects to die from dehydration meets all those criteria and can be incorporated in pest control programs (Subramanyam & Roesli 2000). It is organic in origin, and does not pose any environmental and/or health risk, having a low mammalian toxicity (e.g., DE rat oral LD_{50}, > 5000 mg per kg of body weight (Subramanyam & Roesli 2000). DEs provide stable and long-term grain protection without any toxic residue, and their potential in the control of stored grain pests has received much research attention (e.g., Banks & Fields 1995; Arthur 1996; 2000; 2002; 2004; Arthur & Throne 2003; Fields & Korunic 2000; Subramanyam & Roesli 2000; Athanassiou et al 2003). Studies on DE are mainly focused on the effectiveness against pest species, application rate and exposure periods at different temperature and relative humidity (r.h.) levels.

The lesser grain borer, *Rhyzopertha dominica* (F.), is a serious pest of stored grain worldwide (Potter 1935) and has been present in Turkey since 1957 (Özer 1957). Several reports showed that the lesser grain borer is one of the most difficult stored product insects to control using DE (Quarles 1992; Fields & Korunic 2000; Subramanyam & Roesli 2000; Stathers et al 2002a,b). There has been comparatively little work done on mortality of insects and suppression of progeny production based on wide range of Insecto® concentrations. This study aimed to test the effectiveness of Insecto®, which is a commercially available DE formulation, on adults and F_1 adult production of *R. dominica* (F.) in a wide range of dosages at different exposure intervals and r.h. conditions.

2. Materials and Methods

2.1. Insects

R. dominica adults were reared on a mixture of broken wheat and 5% yeast (by weight) at 25 ± 1°C and 60 ± 5% r.h. Bioassays were conducted

using 1- to 2-week-old, unsexed adults. The insects were from a culture maintained in the laboratory for at least five years, with no history of exposure to insecticides.

2.2. Experimental protocol

Exposure studies were conducted at 40 and 55% r.h. and 0.00 (control), 0.25, 0.50, 1.00, 1.50 and 2.00 g kg^{-1} of Insecto® formulation of DE (Natural Insecto Products, Inc., CA, USA). Humidity chambers were created by pouring 750 ml of saturated K_2CO_3 or NaBr solutions into two sets of plastic boxes (26 cm × 36.5 cm × 15 cm) having waffle-type grids cut to fit the bottom (Arthur 2000). These solutions maintained humidity at approximately 40 and 55%, respectively (Greenspan 1977). Humidity and temperature during the experiment were monitored using HOBO ProTemp/RH data logger (Onset Computer Corporation, MA, USA) during the experiment. Each humidity chamber was kept at 25 ± 1°C.

Clean wheat of known origin was used in the tests. The moisture content as determined by a Dickey–John moisture meter (Dickey-John Multigrain CAC II, Dickey-John Co, KS, USA) of the wheat was approximately 11.8% (11.6-12.0%). Before use in experiments, the wheat was stored for 3 days at -18°C to kill any residual insect infestation and then transferred to each humidity chamber and equilibrated to test conditions for 2 weeks.

The experimental unit consisted of 40 g of soft white winter wheat weighed into a cylindrical plastic vial (6 by 5 cm) with perforated lids covered with US standard sieves mesh #120. The diameter of the lid perforation was 1.5 cm. The appropriate amount of DE was added to 600 g wheat in 1 l glass jars for each humidity and dose rate. Jars were tightly closed with the lids and thoroughly shaken for 5 min to ensure even distribution. Jars were left for 10 min to allow dust settled before dividing into test vials. Bioassays were conducted at each exposure period (7, 14, and 21 days) and humidity combinations. Ten (1-2 wk old) adults were placed in each vial containing wheat treated with each dose rate. After adult introduction, vials were closed with lids and then, test vials were placed in temperature controlled humidity chambers. There were 5 replicates (min.) at each exposure period and control (75 vials for treatment and 15 vials for control at each humidity level). After each exposure interval, dead and live individuals were counted and removed, and then the test vial containing wheat was returned to the corresponding humidity chamber for eight weeks till the emergence of F_1 adults. Live and dead F_1 individuals were counted at the end of each experiment.

2.3. Data analysis

Mean ± SE mortality of adults in the control treatment was found between 0.0 ± 0.0 and 2.0 ± 2.0%. Therefore, dose - mortality data were not corrected for control mortality. Prior to the statistical analysis, an arcsin transformation of the mortality data as well as a square root transformation of the F_1 adult production data was performed to standardize the data. The ANOVA procedure of the SAS (SAS Institute 1987) was used to determine the significance of exposure interval, r.h., and dose rate of DE on the adult mortality and the number of F_1 adults. The Bonferroni Simultaneous test was used to determine significant differences between control and treatments. Duncan's multiple range tests at $P<0.05$ was used to determine statistical differences among treatments (Sokal & Rohlf 1995). The lethal dose for 50% of the population (LD_{50}) was calculated using probit analysis (LeOra Software 2003).

3. Results

GLM statistical analysis showed that mortality and F_1 adult production at five dose rates of DE differed from control group; thus, the main effect of dosages on mortality ($F = 165.97$; $df = 5, 195$; $P<0.001$) and F_1 production ($F = 257.12$; $df = 5, 195$; $P<0.001$) were significant. Bonferroni Simultaneous test indicated that mortality at all dose rates except 0.25 and 0.50 g kg^{-1} were significantly different than that of control Table 1. F_1 production at all dose rates was significantly different than that of control (Table 1).

Table 1-Bonferroni Simultaneous Tests results for the comparison of mortality and F_1 adult production of *Rhyzopertha dominica* (control vs dose rate of DE)(*P*<0.05)

Çizelge 1-Rhyzopertha dominica'da belirlenen ölümler ve F_1 ergin veriminin Benforoni Simultaneous Testi ile kıyaslaması (diyatom toprağının dozlarına karşı kontrol) (P<0.05)

	Mortality	F_1
Dose rate, g kg^{-1}	Adjusted *P*-Value	Adjusted *P*-Value
0.25	1.0000	0.0000
0.50	0.2229	0.0000
1.00	0.0000	0.0000
1.50	0.0000	0.0000
2.00	0.0000	0.0000

The ANOVA analysis for mortality revealed significant differences at each exposure interval (F = 10.66; df = 2, 165; P < 0.001) and dose rate (F = 171.35; df = 5, 165; P < 0.001), but not for humidity (F = 0.22; df = 1, 165) and interactions (Table 2). Mortality obtained after 7, 14 and 21 days of exposures were 27.67±4.03-33.40±4.36% with the highest mortality obtained after 21 days of exposure (Figure 1). According to dose rate analysis, total mortality was low at 0.00, 0.25 and 0.50 g kg^{-1} without any significant differences among groups (Figure 2). Despite the fact that mortality increased with dose rate, only 75% mortality was obtained even at the highest dose of DE (Figure 2). Calculated LD_{50} values for 7, 14 and 21 days exposure intervals at both humidity levels were 0.93 - 1.59 g kg^{-1} (Table 3).

For F_1 adult production of *R. dominica*, the ANOVA analysis showed significant differences for main effect of humidity (F = 32.65; df = 1, 165; P<0.001), exposure interval (F = 68.64; df = 2, 165; P<0.001) and dose rate (F = 465.47; df = 5, 165; P<0.001) (Table 2). All associated interactions except exposure interval × dose rate (F = 5.75; df = 10, 165; P<0.001) (Table 2) were not significant. Relative humidity level caused significant differences in the number of F_1 adults on wheat treated with DE (Figure 3).

According to exposure interval × dose rate interaction, F_1 adult production of *R. dominica* was highest at 0.00 g kg^{-1} and decreased with increasing dose rates at all exposure interval (Table 4). However, no significant differences were found in F_1 adult production between 1.50 and 2.00 g kg^{-1} dose rate for each exposure interval. Although F_1 production at 0.25 g kg^{-1} was significantly lower than that of control for each exposure interval, containment of population were obtained at 1.50 g kg^{-1} of DE for 7 days of exposure period and at 2.00 g kg^{-1} of DE for 14 and 21 days exposure.

4. Discussion

Current study showed that adult mortality of *R. dominica* is dose-dependent, a common phenomenon for DE applications (Fields & Korunic 2000; Athanassiou et al 2003; Stathers et al 2002b, 2004; Vayias & Athanassiou 2004; Ferizli & Beris 2005). Adult mortality at 0.25 and 0.50 g kg^{-1} were 2 and 5%, respectively, while it was 42.67% at 1.00 g kg^{-1}. The highest mortality (75.00±3.09) was obtained at 2.00 g kg^{-1}.

Table 2-ANOVA parameters for main effects and interactions for mortality and F_1 adults production of *Rhyzopertha dominica* (total *df* = 194)

Çizelge 2-Rhyzopertha dominica'da belirlenen ölümler ve F_1 ergin adedine ilişkin varyans analiz sonuçları (total df=194)

		Mortality		F_1	
Source	*df*	*F*	*P*	*F*	*P*
Humidity (r.h.)	1	0.22	0.641	32.65	0.000
Exposure interval	2	10.66	0.000	68.64	0.000
Dose rate	5	171.35	0.000	465.47	0.000
r.h.× exposure interval	2	0.32	0.726	1.56	0.213
r.h. × dose rate	5	0.86	0.512	1.37	0.238
Exposure interval× dose rate	10	1.18	0.306	5.75	0.000
r.h. ×exposure interval× dose rate	10	0.45	0.920	0.45	0.917
Error	165				

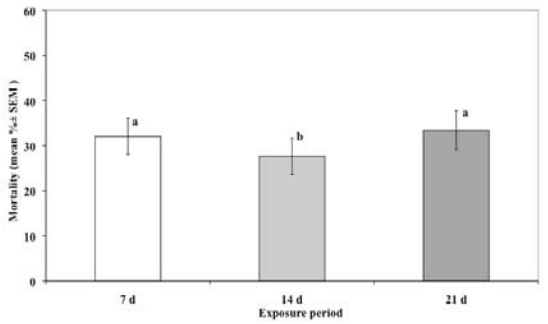

Figure 1-Mortality (mean % ± SEM) of *Rhyzopertha dominica* adults after 7, 14 and 21 d of exposure (means followed by the same letter are not significantly different; *P*>0.05; Duncan's multiple range test)

Şekil 1-Rhyzopertha dominica erginlerinde 7, 14 ve 21 günlük uygulama sonunda belirlenen ölümler (farklı uygulama sürelerinde aynı harfi taşıyan ortalamalar birbirinden önemli düzeyde farklı bulunmamıştır; P>0.05; Duncan'ın multiple range testi).

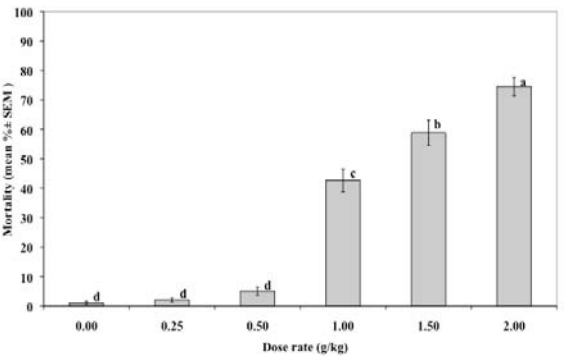

Figure 2-Mortality (mean % ± SEM) of *Rhyzopertha dominica* adults on treated wheat with six different dose rates of DE (means followed by the same letter are not significantly different; *P*>0.05; Duncan's multiple range test)

Şekil 2-Altı farklı dozda diyatom toprağı karıştırılmış buğdayda Rhyzopertha dominica erginlerinde belirlenen ölümler (farklı dozlarda aynı harfi taşıyan ortalamalar birbirinden önemli düzeyde farklı bulunmamıştır; P>0.05; Duncan'ın multiple range testi)

Table 3-Calculated LD_{50} (g kg^{-1}) values of *Rhyzopertha dominica* on treated wheat with six different dose rates of Insecto® after 7, 14 and 21 d of exposure at 25°C

Çizelge 3-Diyatom toprağının altı farklı dozu ile muamele edilmiş buğdayda 25°C sıcaklıkta 7, 14 ve 21 günlük uygulama sonunda Rhyzopertha dominica'da hesaplanan LD_{50} (g kg^{-1})

Humidity, % r.h.	*Exposure, day*	*LD_{50} FL**	*Slope*	*Slope SE*	*χ^2*
40	7	1.43 (1.19-1.74)a**	3.60	0.44	94.9
	14	1.31 (1.16-1.48)ab	4.52	0.61	25.4
	21	1.02 (0.90-1.16)b	3.93	0.43	23.9
55	7	1.59 (1.28-2.20)a	2.67	0.39	37.1
	14	1.46 (1.21-1.84)a	2.98	0.40	32.3
	21	0.93 (0.81-1.06)b	3.49	0.38	23.4

* Fiducial limits were calculated at $P \leq 0.05$ level

** Fiducial limits overlapping comparison

In the control groups, mortality was around 1% as reported by Arthur (2004). Ferizli & Beris (2005) found that Protect-It® produced 3.33 and 20.33% mortality at 0.25 and 0.50 g kg^{-1}, respectively; however, efficacy greatly increased at 1.00 g kg^{-1}, reaching a mortality value of 77.33%. Similarly, a rate of 1.00 g kg^{-1} of Dryacide produced 100% mortality in *R. dominica* (Desmarchelier and Dines, 1987) as well as in *S. oryzae* (McLaughlin, 1994). Increasing the exposure time results in a higher mortality; thus, mortality in *R. dominica* was 72% at 0.3 g kg^{-1} Protect-It® of wheat, after 5 days of exposure and increased to 90% after 14 days of exposure (Fields & Korunic 2000). In the present study, 100% mortality was not obtained even at the highest dose rate (2.00 g kg^{-1}). This is in accordance with the literature, which implies a higher tolerance of *R. dominica* and other

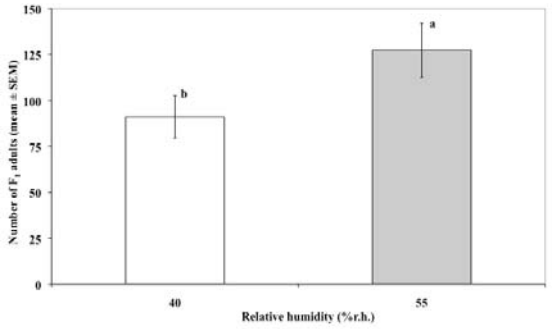

Figure 3-Number of F_1 adults (mean) of *Rhyzopertha dominica* on treated wheat at 25°C; 40% and 55% r.h. (means followed by the same letter are not significantly different; *P*>0.05; Duncan's multiple range test)
Şekil 3. Diyatom toprağı ilave edilmiş buğdayda 25°C ortam sıcaklığında %40 ve %55 orantılı nem koşullarında gelişen Rhyzopertha dominica'nın ergin F_1'leri

bostrichids to DE applications than other stored product pest species (Desmarchelier & Dines 1987; Aldryhim 1993; Fields & Korunic 2000; Stathers et al 2002a,b; Subramanyam & Roesli 2000; Arthur 2002; 2004).

In this study, no significant difference was found between adult mortalities obtained at the r.h. levels of 40 and 55%. Similarly, Ferizli & Beris (2005) did not find any significant mortality difference in *R. dominica* adults treated with diatomaceous earth (Protect-It®) at 40-55% r.h. This could be due to the relatively high moisture content of test wheat (11.6 - 12.0%), which might have eliminated the effects of r.h. values on adult mortality. Probit analysis also showed that LD_{50} values were not significantly different (Table 3). There are also contrary results on DE applications implying that reduced mortality under increased r.h. levels can be obtained (Aldryhim 1993; Arthur 2000; 2002; 2004; Desmarchelier & Dines 1987; Field & Korunic 2000; Vayias & Athanassiou 2004; Subramanyam & Roesli 2000; Stathers et al 2004). Thus, increase in r.h. from 40 to 60% yielded a three-fold increase in LD_{50} for *Tribolium confusum* and *S. oryzae* in Dryacide treated wheat (Aldryhim 1990).

An increase in exposure period to commercial inert DE formulations was shown to increase mortality of stored products beetles (Arthur 2000; Subramanyam & Roesli 2000; Athanassiou et al 2003; Vayias & Athanassiou 2004). However, in this research, we did not observe any increase in mortality at increased exposure periods to DE. Similarly, Ferizli & Beris (2005) found that the mortality of *R. dominica* was not increased with increasing exposure period. Thus, probit analysis estimating the LD_{50} levels for 7, 14 and 21 days exposures at both humidity levels also support this result (Table 3). In contrast to our results, Fields et al (2003) found that LD_{50} values of Protect-It® for *Tribolium castaneum* adults at 7 and 14 d exposure were 638 and 462 ppm, respectively. Arthur (2002) stated that mortality of *S. oryzae* adults exposed to wheat treated with Protect-It® at 57% rh increased when exposure interval and concentration increased. Thus, adult mortality after 7, 14 and 21 days at 71 ppm DE was reported as 50.0, 100.0 and 100.0% (Arthur 2002). The difference between the efficacy in the present work and those in the literature can be attributed to higher tolerance of *R. dominica* to DE in comparison to other stored-grain beetles (Fields & Korunic 2000; Subramanyam & Roesli 2000). Arthur (2002) also reported that higher application rates or longer exposure intervals were necessary for control of *R. dominica*. Because, *R. dominica* adults treated with DE bored into kernels, stayed inside and avoided from continuous contact with the dust. Besides the repellency effect (White et al 1966; Mohan & Fields 2002), DE also reduces the locomotion ability of *R. dominica* adults (Vardeman et al 2007).

In this study, F_1 adult production was lower at 40% r.h. than that produced at 55% r.h. Similarly, Stathers et al (2004) reported reduced progeny production for various stored product pests treated with Protect-It® at two dose rates (0.1 and 0.15 g kg^{-1}). They stated that progeny production of *Prostephanus truncatus* was lower at 50% r.h. than at 60% r.h. Progeny production at 0.1 g kg^{-1} dose rate of Protect-It® at 50% r.h. was close to

Table 4-Number of F_1 adults (mean ± SE) of *Rhyzopertha dominica* on treated wheat with six different dose rates of Insecto® after 7, 14 and 21 days of exposure at 25°C
Çizelge 4-Diyatom toprağının altı farklı dozu ile muamele edilmiş buğdayda 25°C sıcaklıkta 7, 14 ve 21 günlük uygulama sonunda Rhyzopertha dominica'da gelişen F_1 erginleri

Dose rate, g kg^{-1}	*Exposure interval, day*		
	7	14	21
0.00	217.10 ± 21.12c*A**	277.30 ± 26.12bA	387.70 ± 28.32aA
0.25	105.00 ± 9.84cB	156.50 ± 11.32bB	219.90 ± 18.57aB
0.50	60.20 ± 8.22cC	88.40 ± 10.65bC	145.60 ± 13.09aC
1.00	13.30 ± 2.16bD	33.30 ± 8.79aD	34.20 ± 6.53aD
1.50	5.20 ± 1.24bE	12.10 ± 3.49aE	14.70 ± 2.83aE
2.00	2.13 ± 0.71bE	9.70 ± 2.62aE	4.60 ± 1.13bE

* Means of number of F_1 adults (row, lower case letters) at each exposure intervals for each dose rate followed by the same letter are not significantly different (P<0.05; Duncan's multiple range test)

** Means of number of F_1 adults (columns, upper case letters) at each dose rate for each exposure intervals followed by the same letter are not significantly different (P<0.05; Duncan's multiple range test)

that of control group; at the higher dose rate of DE progeny production was reduced.

In our experiments, the effects of DE on suppression of F_1 adults were found to be dose dependant. Thus, at 1.50 g kg^{-1} of DE, population containment was obtained for 7 days of exposure; while it inexplicably took 14 - 21 days to reach the containment at 2.00 g kg^{-1}. However, even at the highest dose rate of DE, complete suppression of F_1 adults failed. By contrast, there are many studies reporting the success of DE suppressing progeny production in *R. dominica*. In similar to our results, Desmarchelier & Dines (1987) reported that at least 1.0 g/kg rate of Dryacide in wheat at 28 days of exposure was necessary for the complete control of adults and their progeny of *R. dominica*; while 0.5 g kg^{-1} of Dryacide was sufficient for containment of the adult population only. In other pest species such as *Tribolium castaneum*, *Oryzaephilus surinamensis* and *Plodia interpunctella*, adult emergence in maize treated with Insecto® (DE) at a rate of 1 g kg^{-1} was decreased by 98-100% (Subramanyam et al 1998). Similarly, a minimum of 0.5 g kg^{-1} Dryacide was found to be sufficient for progeny suppression of *R. dominica* (Aldryhim 1993). According to Ferizli & Beris (2005) progeny production of *R. dominica* decreased with the increased rate of DE (Protect-It®) and *R. dominica* population was suppressed at 1.0 g kg^{-1} DE.

The type of formulation is an important factor on the insecticidal action of DE (Subramanyam & Roesli 2000; Kavallieratos et al 2005; Vayias et al 2006); thus, it affects the progeny production of pests. Insecto® used in our tests contains food additive compounds, but Protect-It® contains silica aerogel. Consequently, all additives used in DE formulation differentiate the insecticidal effect of a given DE (Athanassiou et al 2008).

Subramanyam & Roesli (2000) reported that suppression of population growth of pests for long-term protection of grain is more important than total insect mortality. Adult progeny production of *Cryptolestes ferrugineus*, *C. pusillus,* and *O. mercator* was inhibited in corn treated with 0.5 g kg^{-1} of Insecto®. However, reduction in adult progeny of *P. truncatus*, *S. oryzae*, and *R. dominica* at 1.5 g kg^{-1} was 50-85%. Thus, adults of those insects that are relatively tolerant to Insecto® have enough time to mate and lay eggs in crops treated with DE. Since slow mode of action of DE allows adults oviposit on rice kernels before they die (Subramanyam & Roesli 2000). McLaughlin (1994) reported that Dryacide at 1 g kg^{-1} dose rate caused 100% mortality and 77% reduction in adult progeny of *S. oryzae*, suggesting adult emergence could not be completely inhibited at a rate of 1 g kg^{-1} of Dryacide. As reported by Subramanyam & Roesli (2000) and Athanassiou et al (2003), prevention

of adult emergence is more important than killing the insects. Reduction of F_1 adults was also noted by Arthur (2002), who exposed 10 adult rice weevils for 1 week to wheat treated with 300 ppm Protect-It® at 27°C. In comparison with control, in which 56-212 F_1 adults emerged, he obtained 2, 32 and 107 F_1 adults at the r.h. levels of 40, 55 and 75%, respectively. Increase in r.h. increased progeny production. Arthur & Throne (2003) came to a similar conclusion when Protect-It® applied at a rate of 0.3 g kg^{-1} rice failed to the population suppression of *Sitophilus oryzae* (L.) though it caused a complete adult mortality after 3 weeks of exposure.

DE mixed with grain can adversely affect physical and mechanical properties of a bulk commodity such as reduced flowability, bulk density, visible residues, and dust production during handling (Korunic et al 1998). The negative effects can be avoided by lowering the concentrations of DE to low but still effective levels (Subramanyam & Roesli 2000).

5. Conclusion

In conclusion, Insecto® will cause F_1 and following generations to diminish as the adults of each generation die before reproducing normally. In our tests with Insecto® at 2 g kg^{-1}, irritant effects of DE causing adults to stay inside the kernels resulted with low level of F_1 production. However, Insecto® failed to cause total adult mortality even at a dose of 2 g kg^{-1}. In field application, high DE rates are generally not used due to grain quality and handling problems, however these high rates are needed if DEs are going to effectively control serious storage pests, particularly bostrichids.

Acknowledgments

We thank Ensar Başpınar for statistical analysis. The authors also wish to thank Umut Toprak for their valuable comments. Natural INSECTO Products, Inc. (21 Sherwood Place PO Box 12138Costa Mesa, CA 92627) supplied the DE used in this study.

References

Aldryhim Y N (1990). Efficacy of the amorphous silica dust, Dryacide, against *Tribolium confusum* Duv. and *Sitophilus granarius* (L.) (Coleoptera: Tenebrionidae and Curculionidae). *Journal of Stored Product Research* **26**(4): 207-210

Aldryhim Y N (1993). Combination of classes of wheat and environmental factors affecting the efficacy of amorphous silica dust, Dryacide, against *Rhyzopertha dominica* (F.). *Journal of Stored Product Research* **29**(3): 271-275

Arthur F H (1996). Grain protectants: current status and prospects for the future. *Journal of Stored Product Research* **32**(4): 293–302

Arthur F H (2000). Toxicity of diatomaceous earth to red four beetles and confused four beetles (Coleoptera: Tenebrionidae): effects of temperature and relative humidity. *Journal of Economic Entomology* **93**(2): 526–532

Arthur F H (2002). Survival of *Sitophilus oryzae* (L.) on wheat treated with diatomaceous earth: impact of biological and environmental parameters on product efficacy. *Journal of Stored Product Research* **38**(3): 305–313

Arthur F H (2004). Evaluation of methoprene alone and in combination with diatomaceous earth to control *Rhyzopertha dominica* (Coleoptera: Bostrichidae) on stored wheat. *Journal of Stored Product Research* **40**(5): 485–498

Arthur F H & Throne J E (2003). Efficacy of diatomaceous earth to control internal infestations of rice weevil and maize weevil (Coleoptera: Curculionidae). *Journal of Economic Entomology* **96**(2): 510-518

Athanassiou C G, Kavallieratos N G, Tsaganou F C, Vayias B J, Dimizas C B & Buchelos C Th (2003). Effect of grain type on the insecticidal efficacy of SilicoSec® against *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae). *Crop Protection* **22**(10): 1141-1147

Athanassiou C G, Kavallieratos N G, Vayias B J & Panoussakis E C (2008). Influence of grain type on the susceptibility of different *Sitophilus oryzae* (L.) populations, obtained from different rearing media, to three diatomaceous earth formulations. *Journal of Stored Product Research* **44**(3): 279–284

Banks J H & Fields P (1995). Physical methods for insect control in stored-grain ecosystems. In: Jayas D S, White N D G & Muir W E (Eds.), *Stored-Grain Ecosystems*, Marcel Dekker Inc, New York,

pp. 353-409

Desmarchelier J M & Dines J C (1987). Dryacide treatment of stored wheat: its efficacy against insects, and after processing. *Australian Journal of Experimental Agriculture* **27**(2): 309-312

Ferizli A G & Beris G (2005). Mortality and F_1 progeny of the lesser grain borer (*Rhyzopertha dominica* (F.)) on wheat treated with diatomaceous earth: Effects of dose, exposure interval and relative humidity. *Pest Management Science* **61**(11):1103–1109

Fields P & Korunic Z (2000). The effect of grain moisture content and temperature on the efficacy of diatomaceous earths from different geographical locations against stored-product beetles. *Journal of Stored Product Research* **36**(1): 1–13

Fields P, Allen S, Korunic Z, McLaughlin A & Stathers T (2003). Standardized testing for diatomaceous earth. In: *Proceedings 8th International Working Conference on Stored-Product Protection,* 779-784, Wallingford, United Kingdom

Greenspan L (1977). Humidity fixed points of binary saturated aqueous solutions. *Journal of Research of the National Bureau of Standards - A. Physics and Chemistry* **81**A: 89–96

Kavallieratos N G, Athanassiou C G, Pashalidou F G, Andris N S & Tomanovic Z (2005). Influence of grain type on the insecticidal efficacy of two diatomaceous earth formulations against *Rhyzopertha dominica* (F.) (Coleoptera: Bostrychidae). *Pest Management Science* **61**(7): 660–666

Korunic Z, Cenkowski S & Fields P (1998). Grain bulk density as affected by diatomaceous earth and application method. *Postharvest Biology and Technology* **13**(1): 81–89

LeOra Software (2003). PoloPlus, version 1.0: probit and logit analysis user's guide. LeOra Software, Berkeley, CA, USA

McLaughlin A (1994). Laboratory trials on desiccant dust insecticides. In: *Proceedings 6th International Working Conference on Stored-Product Protection,* 638-645, Canberra, Australia

Mohan S & Fields P G (2002). A simple technique to assess compounds that are repellent or attractive to stored-product insects. *Journal of Stored Product Research* **38**(1): 23–31

Mühle J, Huang J, Weiss R F, Prinn R G, Miller B R, Salameh P K, Harth C M, Fraser P J, Porter L W, Greally B R, O'Doherty S & Simmonds P G (2009). Sulfuryl fluoride in the global atmosphere. *Journal of Geophysical Research* **114**, D05306, doi:10.1029/2008JD011162

Potter C (1935). The biology and distribution of *Rhyzopertha dominica* (FAB.) *Transactions of the Entomological Society of London* **83**: 449-482

Quarles W (1992). Diatomaceous earth for pest control. IPM Practitioner XIV: 1-11

Özer M (1957). Türkiye'de Depo, Ambar ve Silolarda Muhtelif Hububat Taneleri Un ve Mamulleri ile Kuru Meyvelerde ve Tütünlerde Zarar Yapan Böcek Türlerinin Morfolojileri, Kısa Biyolojileri ve Yayılışları Üzerine Araştırmalar. Ankara Üniversitesi Ziraat Fakülltesi Yayınları: 125, A. Ü. Basımevi, 136, Ankara

SAS Institute (1987). SAS/STAT Guide for personal computers, version 6 edition, SAS Institute Inc, Cary, NC, USA

Sokal R R & Rohlf F J (1995). Biometry: The Principles and Practice of Statistics in Biological Research. WH Freeman and Co, New York

Stathers T E, Mvumi B M & Golob P (2002a). Field assessment of the efficacy and persistence of diatomaceous earths in protecting stored grain on small-scale farms in Zimbabwe. *Crop Protection* **21**(10): 1033-1048

Stathers T E, Mvumi B M & Golob P (2002b). Can diatomaceous earths have an integrated role in small-scale tropical grain storage? In: *Proceedings of the Conference of the IOBC/ WPRS Working Group Integrated Protection of Stored Products*, Lisbon, IOBC Bulletin **25**(3): 231-237, 2-5 September, Portugal

Stathers T E, Denniff M & Golob P (2004). The efficacy and persistence of diatomaceous earth admixed with commodity against four tropical stored product beetle pests. *Journal of Stored Product Research* **40**(1): 113-123

Subramanyam Bh & Hagstrum D W (1995). Resistance measurement and management. In: Subramanyam Bh & Hagstrum D W (Eds.), *Integrated Management of Insects in Stored Products*, Marcel Dekker Inc, New York, pp. 331-397

Subramanyam Bh, Madamanchi N & Norwood S (1998). Effectiveness of Insecto® applied to shelled maize against stored-product insect larvae. *Journal of Economic Entomology* **91**(1): 280-286

Subramanyam Bh & Roesli R (2000). Inert dusts. In: Subramanyam Bh & Hagstrum D W (Eds.), *Alternatives to Pesticides in Stored-Product IPM*,

Kluwer Academic Publishers, Boston, pp. 321–379

Vardeman E A, Campbell J F, Arthur F H & Nechols J R (2007). Behavior of female *Rhyzopertha dominica* (Coleoptera: Bostrichidae) in a monolayer of wheat treated with diatomaceous earth. *Journal of Stored Product Research* **43**(3): 297-301

Vayias B J & Athanassiou C G **(**2004). Factors affecting the insecticidal efficacy of the diatomaceous earth formulation SilicoSec against adult and larvae of the confused flour beetle, *Tribolium confusum* DuVal (Coleoptera: Tenebrionidae). *Crop Protection* **23**(7): 565-573

Vayias B J, Athanassiou C G, Kavallieratos N G, Tsesmeli C D & Buchelos C Th (2006). Persistence and efficacy of two diatomaceous earth formulations and a mixture of diatomaceous earth with natural pyrethrum against *Tribolium confusum* Jacquelin du Val (Coleoptera: Tenebrionidae) on wheat and maize. *Pest Management Science* **62**(5): 456–464

White G D, Berndt W L, Schesser J H & Fifield, C C **(**1966). Evaluation of inert dusts for the protection of stored wheat in Kansas from insect attack. Agricultural Research Service, USDA, ARS-51-8, pp. 21

Zettler J L & Arthur F H (2000). Chemical control of stored product insects with fumigants and residual treatments. *Crop Protection* **19**(8-10): 577-582