Evaluation of Plastic Packaging for Prevention of Damage to Wheat by Trogoderma granarium (Coleoptera: Dermestidae), and Suitability of Phosphine Fumigation

Evaluation of plastic packaging for prevention of damage to wheat by *Trogoderma granarium* (Coleoptera: Dermestidae), and suitability of phosphine fumigation

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Abstract

We evaluated 4 types of plastic packaging containing whole wheat grains for damage to packaging in the form of scratches (damage through which insects cannot enter packages), holes (damage through which insects can enter packages), and penetrations (insects entering the packaging through holes) by *Trogoderma granarium* (Everts) (Coleoptera: Dermestidae) during the third and fourth instars. The plastic packaging used in this study was opaque polyethylene (high density), polypropylene, transparent polyethylene (low density), and polyvinylchloride with thicknesses of 0.021, 0.023, 0.026, and 0.036 mm, respectively. After 90 d, damage (scratches) was common with polypropylene and transparent polyethylene packaging, less so with opaque polyethylene, and no damage was found with polyvinylchloride packaging. Holes were more frequent in polypropylene, followed by opaque polyethylene, but none in the other forms of packaging. Insect penetrations were more frequent in opaque polyethylene, followed by polypropylene, but none in other forms of packaging. We also determined the number of damaged grains, weight of damaged grains, number of undamaged grains, weight of undamaged grains, weight of frass, and percent weight loss of grains caused by insects in the aforementioned types of packaging, and compared this with a fifth treatment consisting of open (unpackaged) grains infested by *T. granarium* larvae. We found that there was more damage to grain, greater weight of damaged grain, greater weight of frass, and higher percentage of weight loss of grains in unpackaged wheat, followed by opaque polyethylene and polypropylene, but not in transparent polyethylene or polyvinylchloride packaging. Microphotography after 90 d to measure the lengths of openings showed the maximum lengths in scratches and holes in opaque polyethylene, followed by polypropylene, although scratches in transparent polyethylene did not have openings for measurement. Following this, the types of packaging that were resistant to damage, i.e., transparent polyethylene and polyvinylchloride, were evaluated further for phosphine fumigation efficacy in terms of mortality of *T. granarium*. Fumigation results showed that in 24 h more larval mortality occurred in transparent polyethylene than in polyvinyl chloride, making transparent polyethylene overall a better foodstuff packaging material for protection from *T. granarium*.

Key Words: foodstuff packaging; insect pests; scratches; holes; weight loss; management

Resumen

Evaluamos 4 clases de envases de plástico que contenían granos de trigo integral para detectar daños en el empaque hechos por *Trogoderma granarium* (Everts) (Coleoptera: Dermestidae) durante el tercer y cuarto estadios en forma de rasguños (daños a través de los cuales los insectos no pueden entrar en los paquetes), orificios (daños a través de los cuales ellos pueden entrar los paquetes), y penetraciones (insectos que ingresan al empaque a través de los orificios). El empaque de plástico utilizado en este estudio fue polietileno opaco (alta densidad), polipropileno, polietileno transparente (baja densidad), y cloruro de polivinilo, de 0.021, 0.023, 0.026, y 0.036 mm de grueso, respectivamente. Después de los 90 días, el daño (rayones) fue común con el empaque de polipropileno y polietileno transparente, menos con el polietileno opaco, y no se encontró daño con el empaque de cloruro de polivinilo. Los orificios fueron más frecuentes en el polipropileno, seguidos del polietileno opaco, pero ninguno en las otras formas de empaque. Las penetraciones de insectos fueron más frecuentes en el polietileno opaco, seguido del polipropileno, pero no habían en las otras formas de empaque. También determinamos el número de granos dañados, el peso de los granos dañados, el número de granos no dañados, el peso de los granos no dañados, el peso de los deshechos (excremento) del insecto y el porcentaje de pérdida de peso de los granos causados por insectos en las clases de empaque mencionados y comparamos esto con un quinto tratamiento consistente en granos abiertos (sin empaquetar) infestados por larvas de *T. granarium*. Encontramos que hubo más grano dañado, una mayor pérdida de peso del grano dañado, del peso de escremento y del porcentaje de pérdida de peso de los granos en el trigo no envasado, seguido por polietileno opaco y polipropileno, pero no en envases de polietileno transparente y cloruro de polivinilo. La microfotografía después de 90 días para medir la longitud de las aberturas mostró el número máximo de rasgaduras y agujeros en el polietileno opaco, seguido del polipropileno, aunque los rasguños en el polietileno transparente no tenían aberturas para la medición. Después de esto, los tipos de empaque que fueron resistentes al daño, como el polietileno transparente y cloruro de polivinilo, se evaluaron adicionalmente para determinar la eficacia de fumigación con fosfina en términos de mortalidad de *T. granarium*. Los resultados de la fumigación mostraron que en 24 horas se produjo una mayor mortalidad de larvas en el polietileno transparente que en el cloruro de polivinilo, lo que hace que el polietileno transparente sea en general un mejor material de envasado de alimentos para la protección de *T. granarium*.

Palabras Clave: envasado de productos alimenticios; plagas de insectos; rasgaduras; agujeros; pérdida de peso; manejo

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Post-harvest losses are an important problem for food manufacturers throughout the world. If even a single insect penetrates packaging, it establishes the potential for significant damage. Not only is there potential for direct damage by insects, but there is greater potential for microorganisms such as fungi to damage the food. Use of infected foods can cause a number of diseases (Jakic-Dimic et al. 2009). Insect infestation may result in the occurrence of insect fragments in wheat flour, which is a major concern for the milling industry because consumers demand high quality grains (Perez-Mendoza et al. 2003). Percentage of loss of wheat and loss of germination potential increases with duration of storage time due to reproduction by insects (Ahmad et al. 2017).

Unfortunately, traditional storage methods have been practiced for many years with little or no change, even though they are not, in some cases, effective any longer for the control of insect pests. The choice of old-style storage structures, plus application of insecticides by farmers, often occurs because they are uninformed about improved techniques (Hall 1970). The negative effects of chemical insecticides include harmful effects on human health and environmental pollution, as well as development of resistance by the insect pests (Masoumzadeh et al. 2014). Therefore, an alternative method of storage is needed. Insect resistant packaging is an alternative method to prevent damage of food from insects. Insect resistant packaging of food material is the last line of defense for the producer against insect attack (Hou et al. 2004).

Numerous pests attack food products during storage. Agricultural products often are vulnerable to insect attack, except when frozen or canned (Mullen 1994). Khapra beetle, *Trogoderma granarium* Everts (Coleoptera: Dermestidae) is the most damaging stored grain insect pest in many regions of the world (Burges 2008). The larvae of *T. granarium* can survive under adverse conditions, such as in hot and dry environments, environments in which most of the stored grain insect pests cannot survive (EPPO 2013). Its larvae are also resistant to many pesticides commonly used for other stored grain insect pests (Bell & Wilson 1995). Presently, khapra beetle is the principal actionable species associated with stored products that are imported into the US (Stibick 2007). There have been numerous instances where *T. granarium* has been able to establish populations in the US and other foreign locations, and eradication of these populations often is difficult and expensive. Its population can increase rapidly in favorable conditions; in an unfavorable environment, it enters diapause, which can allow it to survive for several years. This is the most common reason for the spread of this pest in commercial trade (Hill 2003). Its larvae easily can penetrate packaging and infest stored goods, pet food, and food packets after they gain entry to a home or storage site. Once in a product, they can reproduce and move to cross-infest previously clean materials. Larvae burrow in and out of packaged material, destroying the integrity of the packaging material (CERIS 2004). According to Shukla et al. (1993), *T. granarium* is able to penetrate polyethylene that is less than 0.08 mm thick.

Packaging of products is commonplace, and plastics comprise the major form of packaging of products due to the benefits derived from plastic films. Plastic bags are acceptable to the customer and are lighter in weight than some other forms of packaging (Kindle 2001; Connolly 2011). Plastic often is used as the sole packaging material, but sometimes also is used as an internal liner of woven and gunny bags for wheat and other cereals. Paine and Paine (1993) indicated that plastic packaging materials provide benefits in the form of protection against insects and avoidance of contamination.

Due to the global importance of *T. granarium* as a quarantine pest, and the economic losses caused by this insecticide-resistant and difficult-to-control insect pest, it seemed appropriate to assess different commonly used plastic packaging for the management of this pest in packaged foodstuffs. The specific objectives of this study were to determine the extent of damage caused by *T. granarium* to packaging containing wheat (a primary host for *T. granarium*), and to calculate the damage to wheat grains caused by *T. granarium* after penetration into packaged wheat when contained in 4 commonly used packaging materials: opaque polyethylene (high density) (PEO), transparent polyethylene (low density) (PET), polypropylene (PP), and polyvinylchloride (PVC). Secondly, when packaging comes with insects already infesting the commodity, the ability of fumigant gas to permeate the packaging and kill the infesting larvae is important. Therefore, the packaging resistant to penetration by insects, and displaying the least damage to grains inside the packaging, as determined in the first experiment, were tested further for phosphine gas effectiveness. This was measured by mortality of *T. granarium* larvae packed with wheat inside the packaging.

Materials and Methods

COLLECTION OF INSECTS

Populations of *T. granarium* (adults and larvae) were collected from different locations in Punjab, Pakistan. The insects were cultured together into a single colony in the laboratory of Entomology, University College of Agriculture and Environmental Sciences, Islamia University of Bahawalpur, Pakistan.

CULTURE OF *TROGODERMA GRANARIUM*

The collected insects were reared in clean jars (Blowplast [Private] Ltd., Karachi, Pakistan) (1.0 kg capacity). The jars were covered by perforated lids to prevent the beetles from escaping from the jars, and to allow ventilation. Wheat grains were used as culture medium. We released 50 male and 50 female adults of *T. granarium* into the rearing jars to obtain homogenous age populations of larvae (third to fourth instars). The jars were kept at 30 ± 2 °C and 65 ± 2% RH in the laboratory by an air conditioner in summer, and by an electric heater in winter. Humidity was maintained by a humidifier. Data on daily temperature and relative humidity were recorded by a thermo-hygrometer.

PACKAGING MATERIALS

Four commonly used types of packaging materials, namely opaque polyethylene (high density), transparent polyethylene (low density), polypropylene, and polyvinylchloride were purchased from the wholesale plastics market in Lahore, Pakistan. Small bags of these plastic materials (8 × 12 cm) were prepared in the laboratory using a pair of scissors and an impulse (heat) sealer. Mean thickness of the different packaging materials was determined using a digital micrometer (Mitutoyo Corporation, Kawasaki, Japan) to measure 30 plastic bags per packaging type. Descriptive statistics were calculated using SPSS software Version 16.0 for Windows (SPSS 2007) to generate average thickness of all packaging types, which were 0.021, 0.023, 0.026, 0.036 mm, respectively, for opaque polyethylene, polypropylene, transparent polyethylene, and polyvinylchloride packaging types.

EXPERIMENTAL PROCEDURES

A whole grain, certified wheat variety ('Meraj 08') was obtained from the Regional Agricultural Research Institute in Bahawalpur, Pakistan. The 4 plastic packaging treatments mentioned above were used to create plastic bags (8 × 12 cm) that were filled with wheat grains (30 g each), and were placed vertically in a plastic jar (0.5 L vol. capacity); each jar contained 1 package of each treatment and in all there were 4 packaging types per jar. Then 50 (third and fourth instar) larvae were released in the jar outside of the packaging at the initiation of the experiment, followed by an additional 50 larvae released every 15

d for an additional 75 d (the length of experiment was 90 d). There were 3 replicates for each treatment. Insect damage such as scratches (damage with or without slits or openings of a small size which do not allow insects to enter packaging), holes (damage with slits or openings of sufficient size to allow insect penetration), and insect penetration (insects which entered in packaging through holes) were recorded in these packets. In order to avoid accidental insect invasion (insects gaining entry through sealing defects or imperfections in packaging, but not because of holes by insects), packets displaying sealing defects were replaced immediately with sound similar packets. Also, for the purpose of assessing damage to grain and weight loss of grain inside packaging by the insects that had penetrated, a control treatment of 30 g of unpackaged wheat was poured into a 0.5 L jar, infested with 50 similar larvae, and was included for each replicate to determine the relative difference in damage in grains and weight loss in unpackaged grain compared to packaged grain.

DATA RECORDING

After a period of 30 d from the date of initial insect release, damage data (number of scratches and holes) were recorded on the outside of packets in all replications. Damage was recorded as a single "scratch" when damage consisted of minor damage occurring in 1 location on the packaging, with or without small openings that did not allow insect penetration into the packet. Similarly, a "hole" consisted of damage on 1 location of the packaging with large openings that would allow insect penetration into packaging. In this way, the number of scratches and holes present on each type of packaging per replicate were recorded. The same procedure was followed for all 3 replicates of packaging. Afterwards, packaging in a replicate was opened to count the number of insects that had penetrated into packets through holes; then the number of damaged grains, weight of damaged grains, number of undamaged grains, and weight of undamaged grains were recorded for each plastic packaging, and in unpackaged (control) grains, to determine the difference among different packaging types and unpackaged grains. The infested grains in all treatments, including the control, were sieved to separate grain dust, cast skins, and other waste material from the insects. Weight and number of damaged grains and undamaged grains were calculated, and the following equation was applied for determination of percentage of weight loss according to the formula of Gwinner et al. (1996):

$$\text{Percent weight loss} = \frac{(Wu \times Nd) - (Wd \times Nu)}{Wu \times (Nd + Nu)} \times 100$$

where Wu = weight of undamaged grains; Nu = number of undamaged grains; Wd = weight of damaged grains; and Nd = number of damaged grains.

Following data collection, the grains and insects were repacked into their respective packages and resealed using the impulse sealer. Similarly, after data collection from the unpackaged treatment (control), grains and infesting insects were returned to their respective jars. The same procedure was repeated after 60 and 90 d to obtain subsequent damage data. When calculating weight loss, the weights of frass, including grain dust, cast skins, etc., also were calculated in all treatments, including the control.

FUMIGATION OF PACKAGED WHEAT TO CONTROL *TROGODERMA GRANARIUM* INFESTATION

In order to decide overall about the suitability of the different types of packaging, another experiment was performed for fumigation of packaged wheat infested by the *T. granarium* larvae. Two of the packaging materials that proved most resistant to penetration and damage to grains inside in first experiment, namely transparent polyethylene and polyvinylchloride, were used to check the effect of fumigation efficacy by phosphine on mortality of *T. granarium*. Plastic bags (8 × 12 cm) were filled with wheat grains (30 g each) and were infested with 30 homogenous-aged larvae in each bag, and the bags were sealed. Plastic bags of transparent polyethylene and polyvinylchloride containing wheat and larvae were placed inside a tin container (0.7 m³ volume) with 4 partitions. Two of each type of the plastic bags were placed in each partition; thus, for 4 replicates of experiment there were 8 plastic bags. A 3 g metal phosphide tablet (aluminum phosphide) (Phostoxin®) (Degesch America, Weyers Cave, Virginia, USA) wrapped in muslin cloth was placed in a plate situated at the top of center of the 4 replicates for equivalent generation of phosphine gas in all compartments of the tin container. We used a 0.7 m³ volume tin container to generate 1.5 g phosphine gas per m³ at 28 ± 2 °C and 65 ± 5 RH. The lid of the container was tightly closed with sealing tape to avoid any leakage during the experiment. Mortality was recorded after 24 h. The same experiment was repeated to obtain mortality after 48 h. These experiments were repeated to check natural mortality without the phosphine tablet, and to correct the mortality in treatments using a mortality correction formula (Abbott 1925).

STATISTICAL ANALYSIS

Data on frequency of scratches and holes on packaging, frequency of insect penetrations in the 4 packaging types, and incidence of grain damage in the 5 different packaging types (including unpacked control grains) were tested with 1-way ANOVA using SPSS Version 16.0 for Windows (SPSS 2007). The mean values were separated statistically using Tukey's HSD test at the 5% probability level. Data on fumigation-induced mortality in relation to packaging type were analyzed using corrected mortality percentages with a *t* test.

MICROPHOTOGRAPHY

To assess the susceptibility of packaging, we measured maximum lengths of openings (μm) in scratches and holes by microphotography. For this purpose, damaged areas displaying the maximum size of openings were selected on all packaging types. One cm² damaged areas were cut with a pair of scissors and placed on a glass slide. The glass slides were fitted below a trinocular microscope (Labomed, CXR3, Labo America, Inc., Fremont, California, USA) coupled with a digital camera Model HD 1500 T (Meiji, TECHNO, Saitama, Japan) and T Capture Version 3.9 digital software (T Capture 2017). All photographs were taken at a magnification of 40×.

Results

NUMBERS OF SCRATCHES, HOLES, AND INSECT PENETRATIONS IN PACKAGING CAUSED BY *TROGODERMA GRANARIUM*

Damage in the form of scratches after 90 d were seen more in polypropylene (2.000 ± 1.000) and transparent polyethylene (2.000 ± 1.000) followed by opaque polyethylene (1.333 ± 0.333), but none in polyvinylchloride packaging. However, after 30 or 60 d, there were more scratches in opaque polyethylene than in polypropylene, but none were seen in transparent polyethylene or polyvinylchloride packaging. The number of holes up to 60 d were greater in opaque polyethylene (1.333 ± 0.882) than in polypro-

pylene (0.667 ± 0.667), but none were present in other packaging; after 90 d more holes were found in polypropylene (2.000 ± 1.155) than in opaque polyethylene (1.667 ± 0.882), and no holes were found in other packaging. Insect penetrations through holes up to 90 d were greater in opaque polyethylene (6.333 ± 3.283) followed by polypropylene (6.000 ± 3.786), but no penetrations were seen in other packaging (Table 1; *P* > 0.05).

DAMAGE TO PACKAGED WHEAT BY *TROGODERMA GRANA-RIUM* IN RELATION TO DIFFERENT PLASTIC PACKAGING COMPARED TO UNPACKAGED GRAINS

After all 3 time intervals, the greatest numbers of damaged grains were recorded in unpackaged grains, followed by opaque polyethylene and polypropylene in descending order, but none in other packaging. After 90 d, the maximum numbers of damaged grains were found in unpackaged grains (321.00 ± 9.238), followed by opaque polyethylene (18.000 ± 9.291) and polypropylene (15.333 ± 10.333), but none were found in transparent polyethylene and polyvinylchloride. Similarly, the weight of damaged grains was the greatest in unpackaged grains, followed by opaque polyethylene and polypropylene after all 3 time periods. Correspondingly, the numbers of undamaged grains were least in unpacked grains (481.67 ± 8.413), followed by opaque polyethylene (777.33 ± 12.914), polypropylene (780.67 ± 8.452), transparent polyethylene (782.00 ± 10.440), and polyvinylchloride (784.67 ± 2.186) in ascending order. Similarly, the weight of the undamaged grains was least in unpackaged grains, followed by polypropylene, opaque polyethylene, transparent polyethylene, and polyvinylchloride in ascending order (Table 2; *P* < 0.05).

The weight (g) of frass was greatest in unpackaged grains, followed by opaque polyethylene or in polypropylene, but no frass was detected in transparent polyethylene or polyvinylchloride packaging. Percentage of weight loss was greatest in unpackaged grains (16.100 ± 1.322), followed by opaque polyethylene (0.499 ± 0.266), and polypropylene (0.056 ± 0.085) in descending order, but no weight loss of wheat grains occurred in transparent polyethylene or polyvinylchloride packaging (Table 3; *P* < 0.05).

MORTALITY OF *TROGODERMA GRANARIUM* DUE TO PHOSPHINE FUMIGATION UNDER THE EFFECT OF PACKAGING TYPES

Mortality of *T. granarium* due to phosphine was tested using the packaging materials that proved resistant to damage by *T. granarium*. Results showed that after 24 h maximum mean mortality was generated in polyethylene transparent (89.868 ± 2.045), and least mortality was in polyvinylchloride packaging (52.311 ± 14.869) (Table 4; Sig (2-tailed): *P* = 0.046). After 48 h 100% mean mortality was generated in both packaging materials in all replications (Table 4).

SUSCEPTIBILITY OF PACKAGING DUE TO SIZE OF OPENINGS

Susceptibility of packaging was tested on the basis of maximum length of an opening inside a scratch or hole created by larvae of *T. granarium*. The greatest lengths of scratches and holes were focused under the microscope for measurement. Maximum length of an opening in a hole was 2,495 μm (in opaque polyethylene) followed by 1,943 μm (in polypropylene) (Fig. 1A, B). The maximum length of an opening associated with scratch damage was 688 μm (in opaque polyethylene), whereas the minimum length of an opening associated with scratch damage was 146 μm (in polypropylene) (Fig. 1C, D). Scratches associated with transparent polyethylene did not show slits or openings.

Discussion

Our research on damage to packaging by *T. granarium* revealed that after 90 d scratches made by *T. granarium* were more frequent in polypropylene or transparent polyethylene packaging, followed by opaque polyethylene; no scratches were found in polyvinylchloride packaging. Holes occurred more frequently in polypropylene, followed by opaque polyethylene, but no holes were found in transparent polyethylene or polyvinylchloride packaging. Similarly, penetrations by the insects were more frequent in opaque polyethylene followed by polypropylene, but no insects penetrated the transparent polyethylene or polyvinylchloride packaging. When evaluated at 30, 60, and 90 d, both scratch and hole damage increased with time.

Table 1. Comparison of damage type occurrence in 4 types of plastic packaging at 3 time intervals when exposed to *Trogoderma granarium* larvae.

Damage type	Packaging type	N	30 d (mean ± SE)	60 d (mean ± SE)[a]	90 d (mean ± SE)
Scratches	Polyethylene, opaque	3	0.667 ± 0.667 NS	1.333 ± 0.333 b	1.333 ± 0.333 NS
	Polyethylene, transparent	3	0.000 ± 0.000	0.000 ± 0.000 a	2.000 ± 1.000
	Polypropylene	3	0.333 ± 0.333	0.333 ± 0.333 ab	2.000 ± 1.000
	Polyvinylchloride	3	0.000 ± 0.000	0.000 ± 0.000 a	0.000 ± 0.000
	Total (grand means)	12	0.250 ± 0.179	0.417 ± 0.193	1.333 ± 0.396
	Statistics (df: 3, 8)		*F*: 0.733; *P*:0.561	*F*: 7.167; *P*: 0.012	*F*: 1.684; *P*: 0.247
Holes	Polyethylene, opaque	3	0.333 ± 0.333 NS	1.333 ± 0.882 NS	1.667 ± 0.882 NS
	Polyethylene, transparent	3	0.000 ± 0.000	0.000 ± 0.000	0.000 ± 0.000
	Polypropylene	3	0.333 ± 0.333	0.667 ± 0.667	2.000 ± 1.155
	Polyvinylchloride	3	0.000 ± 0.000	0.000 ± 0.000	0.000 ± 0.000
	Total (grand means)	12	0.167 ± 0.112	0.500 ± 0.289	0.917 ± 0.417
	Statistics (df: 3, 8)		*F*: 0.667; *P*: 0.596	*F*: 1.333; *P*: 0.330	*F*: 2.158; *P*: 0.171
Penetrations	Polyethylene, opaque	3	1.333 ± 1.333NS	4.000 ± 2.082 NS	6.333 ± 3.283 NS
	Polyethylene, transparent	3	0.000 ± 0.000	0.000 ± 0.000	0.000 ± 0.000
	Polypropylene	3	1.000 ± 1.000	2.667 ± 2.667	6.000 ± 3.786
	Polyvinylchloride	3	0.000 ± 0.000	0.000 ± 0.000	0.000 ± 0.000
	Total (grand means)	12	0.583 ± 0.398	1.667 ± 0.890	3.083 ± 1.417
	Statistics (df: 3, 8)		*F*: 0.680; *P*:0.589	*F*: 1.398; *P*: 0.312	*F*: 2.022; *P*: 0.189

[a]Means in a column followed by different lowercase letters are significantly different (*P* ≤ 0.05; ANOVA and Tukey HSD test). N denotes replicate numbers per treatment. NS. Non-significant.

Table 2. Damage (means)[a] in packaged wheat by *Trogoderma granarium* in relation to different types of plastic packaging and unpackaged grains.

Damage type	Packaging type	N	30 d (mean ± SE)	60 d (mean ± SE)	90 d (mean ± SE)
Weight of damaged grains	Polyethylene, opaque	3	0.127 ± 0.127 a	0.383 ± 0.198 a	0.533 ± 0.275 a
	Polyethylene, transparent	3	0.000 ± 0.000 a	0.000 ± 0.000 a	0.000 ± 0.000 a
	Polypropylene	3	0.087 ± 0.087 a	0.283 ± 0.283 a	0.557 ± 0.363 a
	Polyvinylchloride	3	0.000 ± 0.000 a	0.000 ± 0.000 a	0.000 ± 0.000 a
	Control (unpackaged grains)	3	3.953 ± 0.220 b	6.213 ± 0.191 b	8.430 ± 0.370 b
	Total (grand means)	15	0.833 ± 0.420	1.376 ± 0.651	1.904 ± 0.880
	Statistics (df: 4, 10)		F: 211.559; P: 0.000	F: 235.454; P: 0.000	F: 194.136; P: 0.000
Number of damaged grains	Polyethylene, opaque	3	3.333 ± 3.333 a	11.333 ± 6.119 a	18.000 ± 9.291 a
	Polyethylene, transparent	3	0.000 ± 0.000 a	0.000 ± 0.000 a	0.000 ± 0.000 a
	Polypropylene	3	2.667 ± 2.667 a	8.000 ± 8.000 a	15.333 ± 10.333 a
	Polyvinylchloride	3	0.000 ± 0.000 a	0.000 ± 0.000 a	0.000 ± 0.000 a
	Control (unpackaged grains)	3	127.33 ± 8.667 b	227.00 ± 10.693 b	321.00 ± 9.238 b
	Total (grand means)	15	26.667 ± 13.556	49.267 ± 23.909	70.867±33.604
	Statistics (df: 4, 10)		F: 169.774; P: 0.000	F: 229.318; P: 0.000	F: 352.358; P: 0.000
Weight of undamaged grains	Polyethylene, opaque	3	29.877 ± 0.133 a	29.557 ± 0.223 a	29.383 ± 0.312 a
	Polyethylene, transparent	3	30.030 ± 0.011 a	30.030 ± 0.011 a	30.030 ± 0.011 a
	Polypropylene	3	29.910 ± 0.100 a	29.653 ± 0.357 a	29.343 ± 0.423 a
	Polyvinylchloride	3	30.033 ± 0.013 a	30.033 ± 0.013 a	30.033 ± 0.013 a
	Control (unpackaged grains)	3	26.050 ± 0.208 b	23.563 ± 0.199 b	21.193 ± 0.370 b
	Total (grand means)	15	29.180 ± 0.421	28.567 ± 0.675	27.997 ± 0.919
	Statistics (df: 4, 10)		F: 214.671; P: 0.000	F: 181.088; P: 0.000	F: 176.190; P: 0.000
Number of undamaged grains	Polyethylene, opaque	3	792.00 ± 9.291 a	784.00 ± 10.598 a	777.33 ± 12.914 a
	Polyethylene, transparent	3	782.00 ± 10.440a	782.00 ± 10.440 a	782.00 ± 10.440 a
	Polypropylene	3	793.33 ± 0.667 a	788.00 ± 6.000 a	780.67 ± 8.452 a
	Polyvinylchloride	3	784.67 ± 2.186 a	784.67 ± 2.186 a	784.67 ± 2.186 a
	Control (unpackaged grains)	3	675.33 ± 7.965 b	575.67 ± 10.682 b	481.67 ± 8.413 b
	Total (grand means)	15	765.47 ± 12.406	742.87 ± 22.588	721.27 ± 32.212
	Statistics (df: 4, 10)		F: 48.516; P: 0.000	F: 116.166; P: 0.000	F: 212.251; P: 0.000

[a]Means in a column followed by different lowercase letters are significantly different ($P \leq 0.05$; ANOVA and Tukey HSD test). N denotes numbers of replicates per treatment.

The primary reason for damage to packaging and for insect penetration could be the thickness of the packaging material. Opaque polyethylene and polypropylene were thinner than transparent polyethylene and polyvinylchloride (i.e., thickness of packaging materials were 0.020 for opaque polyethylene, 0.023 for polypropylene, 0.026 for transparent polyethylene, and 0.036 for polyvinylchloride). This is consistent with previous research that attributed thickness of packaging materials to be a major factor in resistance of packaging to penetration by storage insect pests. For example, Li et al. (2014) assessed penetration by larvae of Indian meal moth, *Plodia interpunctella* Hübner (Lepidoptera: Pyralidae) through packaging of different types and thicknesses. Their results showed that larvae generally penetrated thinner packaging

Table 3. Weight of frass and percentage weight loss of packed grains (means)[a] by *Trogoderma granarium* in relation to different plastic packaging compared to unpacked grains.

Damage type	Packing type	N	30 d (mean ± SE)	60 d (mean ± SE)	90 d (mean ± SE)
Weight of frass	Polyethylene, opaque	3	0.040 ± 0.040 a	0.103 ± 0.052 a	0.127 ± 0.064 a
	Polyethylene, transparent	3	0.000 ± 0.000 a	0.000 ± 0.000 a	0.000 ± 0.000 a
	Polypropylene	3	0.037 ± 0.037 a	0.097 ± 0.097 a	0.137 ± 0.084 a
	Polyvinylchloride	3	0.000 ± 0.000 a	0.000 ± 0.000 a	0.000 ± 0.000 a
	Control (unpacked grains)	3	0.440 ± 0.032 b	0.667 ± 0.041 b	0.820 ± 0.047 b
	Total (grand means)	15	0.103 ± 0.046	0.173 ± 0.070	0.217 ± 0.084
	Statistics (df: 4, 10)		F: 44.986; P: 0.000	F: 28.533; P: 0.000	F: 44.214; P: 0.000
Percent weight loss	Polyethylene, opaque	3	0.003 ± 0.002 a	0.877 ± 0.890 a	0.499 ± 0.266 a
	Polyethylene, transparent	3	0.000 ± 0.000 a	0.000 ± 0.000 a	0.000 ± 0.000 a
	Polypropylene	3	0.044 ± 0.044 a	0.050 ± 0.050 a	0.056 ± 0.085 a
	Polyvinylchloride	3	0.000 ± 0.000 a	0.000 ± 0.000 a	0.000 ± 0.000 a
	Control (unpackaged grains)	3	3.099 ± 0.431 b	9.382 ± 0.999 b	16.100 ± 1.322 b
	Total (grand means)	3	0.629 ± 0.338	2.062 ± 1.007	3.331 ± 1.722
	Statistics (df: 4, 10)	15	F: 50.641; P: 0.000	F: 47.091; P: 0.000	F: 139.606; P: 0.000

[a]Means in a column followed by different lowercase letters are significantly different ($P \leq 0.05$; ANOVA and Tukey HSD test). N denotes numbers of replicates per treatment.

Table 4. Mean mortality of *Trogoderma granarium*[a] at 24 and 48 h due to phosphine fumigation as affected by packaging type.

		Mortality (mean ± S.E)	
Packaging	N	24 h	48 h
Polyethylene, transparent	4	89.868 ± 2.045 a	100.000 ± 0.000
Polyvinylchloride	4	52.311 ± 14.869 b	100.000 ± 0.000
Statistics		*t*: 2.502; *P*: .046	No test statistics presented in output due to 0 SD

[a]Means in a column followed by different lowercase letters are significantly different (Sig ≤ 0.05; t-test). N denotes numbers of replicates per treatment. SD: Standard deviation.
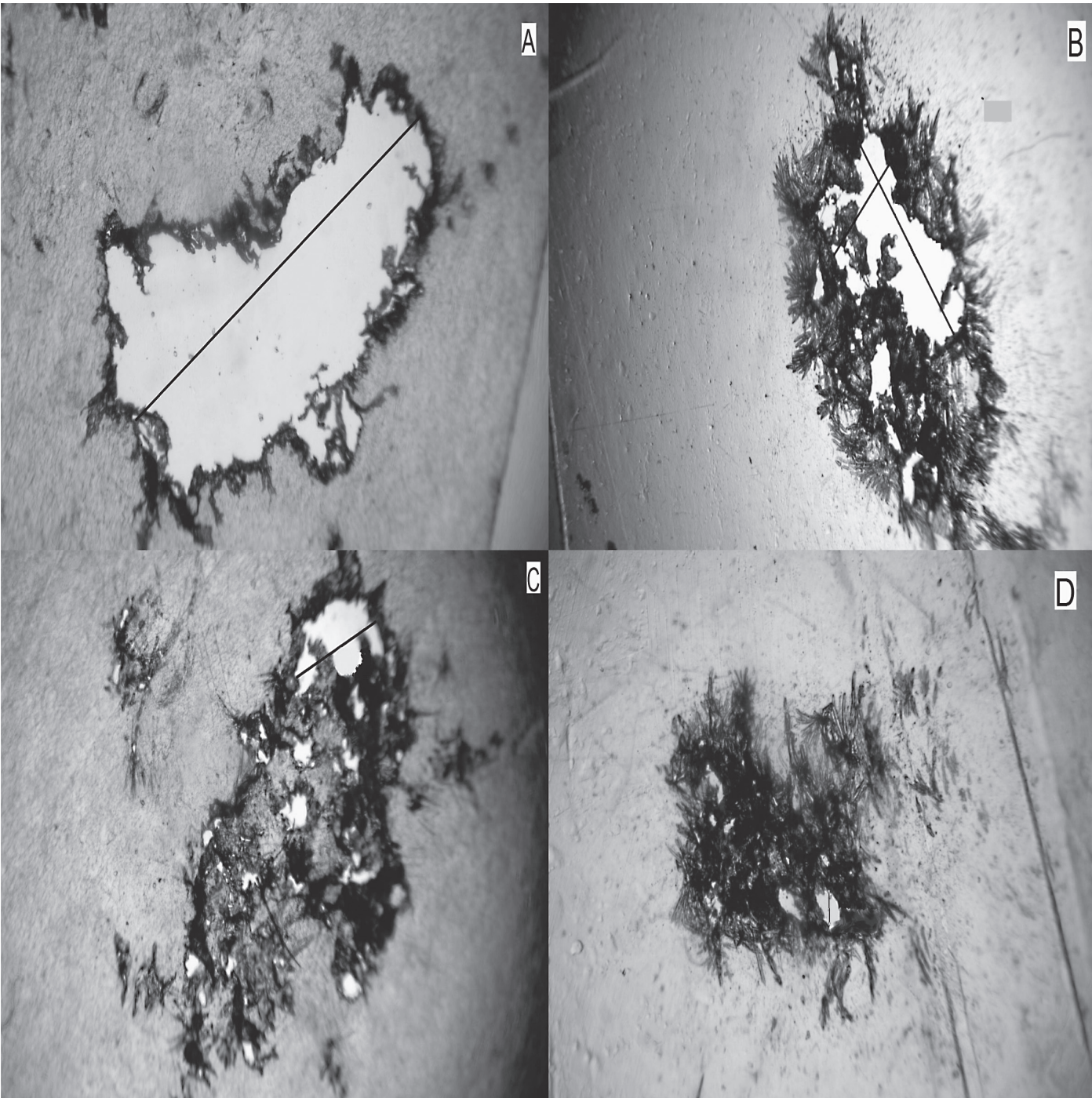

Fig. 1. Damage (holes and scratches) with openings created by *Trogoderma granarium* in plastic packaging: Lengths of openings in holes, (A) measurement line = 2,495 **μm** in opaque polyethylene, and (B) measurement line = 1,943 **μm** in polypropylene. Lengths of openings in scratches, (C) measurement line = 688 **μm** in opaque polyethylene, and (D) measurement line = 146 **μm** in polypropylene.

compared to thick packaging, and that polypropylene and polystyrene packaging were completely penetrated when used at thinner thicknesses. Our results are in agreement with our recent findings showing that damage was more pronounced in thin packaging than in thicker packaging (Hassan et al. 2016), and our more recent studies showed that transparent polyethylene packaging became particularly susceptible for packed foodstuff against *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae) when used at 0.02 mm thickness followed by polypropylene at 0.02 mm; no damage occurred in polyvinylchloride packaging at 0.03 mm thickness (Yar et al. 2017). Our results also are consistent with the findings of Shukla et al. (1993), who reported that khapra beetles can penetrate packaging of less than 0.08 mm thickness. In our results, *T. granarium* only created holes and entered packaging when the packaging was ≤ 0.023 mm in thickness.

The packagings that were most resistant to penetration by *T. granarium* were evaluated further in terms of phosphine fumigation efficacy, because this is a component of the decision as to which packaging is superior. After 24 h of treatment, *T. granarium* mortality was higher in transparent polyethylene packaging compared to polyvinylchloride packaging. After 48 h 100% mortality occurred in both types of packaging. These results show that transparent polyethylene packaging is superior to the polyvinylchloride packaging. These results are in agreement with our previous findings that showed mortality of pest species was lower in polyvinylchloride packaging than in transparent polyethylene or polypropylene packaging (Hassan et al. 2016; Akram et al. 2018).

Overall, our results show that transparent polyethylene and polyvinylchloride packaging were better choices than the other products evaluated for packaging of grains. However, when phosphine fumigation efficacy was factored in, transparent polyethylene was revealed to be the best packaging for prevention of damage by *T. granarium*. Thus, we recommend transparent polyethylene as a safe choice for packaging of commodities that are susceptible to *T. granarium*.

Acknowledgments

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