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Development, Survival, and Reproduction of the Khapra Beetle, *Trogoderma granarium* Everts (Coleoptera: Dermestidae) in Response to different Stored Diets and their Chemical Composition

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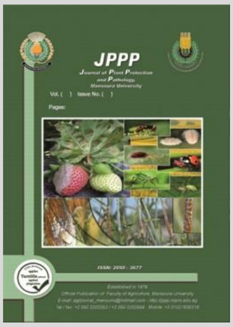

ABSTRACT

Many stored grains and by-products become infested by the polyphagous pest, *Trogoderma granarium* Everts (Coleoptera: Dermestidae). The effect of eight leguminous commodities seeds (e.g. cowpea, faba bean, lentil, and kidney bean) and fruit nuts (e.g. cashew, almond, hazelnut, and walnut) on development, survival and reproduction of *T. granarium* was studied at conditions of 30 ± 1 °C, 65 ± 5% RH and a photoperiod of 14L: 10 D. Furthermore, all chemical components of the tested products were analyzed. The results implied that the examined commodities affected biological characteristics of *T. granarium*. The proportions of the internal components of the tested commodities varied, and the proximate analysis (%) varied among them. Biologically, the longest period of life cycle (egg-adult) of *T. granarium* was estimated on almonds (72.1±1.65 d) and the shortest one was on cowpea seed (45.4±1.47 d). The total oviposition period of *T. granarium* was the highest on cowpea seeds (12.9 d) and the lowest on almond (8.8 d). The highest fecundity and adult emergence rates were on cowpea seeds and the lowest was on almonds. The highest survival rate of larval and pupal stages was on faba bean seeds, and the lowest was on almond. Results revealed that both hazelnuts and walnuts were unsuitable food for the development of *T. granarium* beetles.

Keywords: Development, chemical composition, reproduction proximate, survival.

INTRODUCTION

The khapra beetle, *Trogoderma granarium* Everts (Coleoptera: Dermestidae) is one of the most significant insects infesting stored products and subjects to quarantine globally (Hagstrum *et al.*, 2012 and Castañé *et al.*, 2020). This beetle is included in the list of the 100 most invasive species in the world (Lowe *et al.* 2000). Apart from its significance as a quarantine species, *T. granarium* might be thought of as a species that "travels" across nations with ease because it has a very wide range of dietary requirements and can use cracks and crevices or other refuges with relative ease (Aitken, 1975 and Hagstrum *et al.*, 2012). In hot and dry conditions, almost most of stored-product insect species cannot develop, however the larvae of khapra beetle can live in a wide range of abiotic environments (Hill *et al.* 2002). However, human health may be at risk if cast larval skin and hair are present (Pruthi and Singh, 1950 and Bell, 1984). Chemical control has been the main strategy for managing pests associated with stored products; however, its use is not without its drawbacks, including high production costs, insect resistance, and hazards to consumer health and environmental (Han *et al.*, 2017 and Kavallieratos and Boukouvala, 2018).

Because it is both economically feasible and safe for humans, the use of resistant crops is a part of insect pest management (Subramanyam, 1995). Nutritional indicators are significant metrics used to assess plant-insect interactions and the vulnerability or resistance of various host plants to insect pests (Naseri and Borzoui, 2016 and Hashem *et al.* 2021). To accomplish this objective, it may be necessary to find secondary compounds in host grains that are inhospitable to *T. granarium* by studying the feeding and growth behaviors of stored product pests on various host cultivars (Borzoui *et al.* 2017 and Naseri *et al.* 2017). The quantity and quality of food consumed by larvae has a significant impact on the storage of resources with a non-feeding adult stage, such as *T. granarium* (Awmack and Leather, 2002). Some chemical characteristics of the host plants, as well as the nutrient levels of the food taken, are related to an insect's capacity to grow on different diets (Joern and Behmer,1997 and Karasov *et al.* 2011). Growth, survival, and reproduction of an insect species can be affected by the nutritional content of its food type (Tsai and Wang, 2001).

One of the main components of an integrated pest control program has been proposed in recent years: assessing the nutritional performance and digestive physiology of khapra beetle larvae that fed with various grain cultivars (Naseri and Borzoui, 2016 and Majd-Marani *et al.*, 2018). Aheer and Ahmad (1993) examined how well *T. granarium* able to feed on twelve different wheat cultivars and found that 86299 was a cultivar that relatively inhibited the pest well. According to study performed by Aheer and Ahmad (1993) on population dynamics of *T. granarium* on different wheat cultivars, seed hardness

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significantly affected the *T. granarium* population. According to earlier research, *T. granarium* can consume a variety of foods, both plant and animal-based (Lindgren 1955 and Lindgren and Vincent 1959). It also seems to have similar nutritional requirements for development compared to other stored insects that use cereal byproducts (Nawrot *et al.* 1985 and Rajendran 2005).

It is generally accepted that grains and related amylaceous materials, like flour, are the preferred diets for this species (Lindgren *et al.*, 1955; Bhattacharya and Pant, 1968 and Viljoen, 1990).

This beetle infests spices, nuts, oilseeds, dried fruits, and dried vegetables in addition to cereals and their by-products (Degri and Zainab 2013 and Hagstrum and Subramanyam, 2017). For instance, Davey *et al.* (1960) found that this species might significantly alter the qualitative characteristics of peanuts. Furthermore, *T. granarium* can only grow on lentil and French bean flours when they are supplemented with artificial diets, which suggests that these products lack certain nutrients or contain growth inhibitors (Bhattacharya and Pant, 1969). Furthermore, (Rao, *et al.* 2004) evaluated various wheat types, and they discovered that specific traits—such as kernel hardness and kernel size—were critical for the expansion of *T. granarium* population. In a different study, (Rajput, *et al.*, 2015) discovered a negative correlation between rate of population increase and kernel size of various wheat genotypes. The majority of the published data focused on grains, compared various intact grain commodities, however no recent data are available regarding effect of legume seeds (*i.e.* cowpea, faba bean, lentil, and kidney bean) or nuts (*i.e.* cashew, almond, hazelnut, and walnut) on development and reproduction of *T. granarium* or the relationship between grain condition and chemical kernel containment. Consequently, this study examined these parameters to know how these variables affect *T. granarium* population growth.

MATERIALS AND METHODS

Sources of grains and insects

The tested legume seeds (cowpea, faba bean, lentil and kidney bean) and nuts (cashew, almond, hazelnut and walnut) were collected and were dried in an oven at 50 °C (Athanassiou *et al.*, 2017) to ensure that they were free from any infection and pesticide residues. Adults of *T. granarium* were obtained from Plant Protection Research Institute, the Stored Products and Grain Pests Department at the Sakha Station, Kafr El-Sheikh, Egypt. *Trogoderma granarium* was reared on wheat (*Triticum aestivum* L.) at conditions of 30 ± 1 °C, 65 ± 5% relative humidity, and a photoperiod of 14L: 10D h Experiments were conducted in laboratory of Economic Entomology Department, Faculty of Agriculture, Mansoura University Three-day-old, mated females were used for the experiments. Two to three days later, females started to oviposit.

Development and reproduction

Roughly 10 g of each seed were distributed in Petri dishes measuring 6 cm in diameter and 1 cm in depth., and each Petri dish was then filled with 10 freshly hatched *T. granarium* larvae (Borzoui *et al.*, 2015). The Petri dishes were kept in an incubator adjusted to physical conditions similar to those previously described. Five replicates were performed. Survival during the immature stages (larval and pupal stages) was recorded every day.

After adult eclosion, a pair of male and female was confined within a plastic tube measuring 6 cm in diameter and 2 cm in depth until death. And 10 replicates were used once female started to lay eggs, male was excluded. The number of eggs laid daily by each female was counted. This study recorded the following: fecundity, adult longevity, oviposition period, total preoviposition period and adult preoviposition period

Grain chemical properties

According to (Al-Mentafji, 2016) , the crude protein, crude fat, ash, and crude fiber content of the tested commodities in this study were measured. In brief, for each commodity, 200 mg of pulverized seeds were homogenized in 35 ml of distilled water and brought to a boil. Each sample was weighed out to be 100 milliliters. An equal volume of iodine reagent (0.02% I2 and 0.2% KI) was added, and the absorbance was measured at 580 nm. Two grams of each ground product were weighed, dried for three hours at 100 degrees Celsius in an oven, and then weighed again to ascertain the moisture content of the tested cultivars. Using the Ienway Flame photometer model Corning 400, total magnesium, calcium, and potassium were determined flame photometrically, according to (Peterburgski, 1968). Zn and Fe were extracted from the samples by use of the microwave digestion technique.

Methanol was used to extract the total phenol content of the 2g of powdered plant material during an overnight room-temperature extraction. The methanol extracts were combined and concentrated on a rotating evaporator with the pressure reduced. Following the specified protocol, the total phenolic content of each plant extract was calculated using Folin-Ciocalteu's reagent (FCR). Each sample (0.5 ml) was mixed with 2.5 ml of FCR (diluted 1:10, v/v), and then 2 ml of Na_2CO_3 (7.5%, v/v) solution was added. After incubating for 90 minutes at 30°C, the absorbance was then measured at 765 nm. The results were stated as the equivalent of gallic acid (mg Gallic acid /100g dry extract) (Slinkard and Singleton, 1977). One hundred grams of grain were taken in to determine the oil content (%) of the examined cultivars. The grains were ground and sent to a Soxhlet machine for a 12-hour oil extraction process at 60˚C, along with solvent (hexane) washings (Al-Mentafji, 2016). Following the attainment of uniform weight, the extracted cultiv oil's hexane was evaporated using a boiling water bath and rotating vacuum, allowing the oil percentage to be determined.

Data analysis

Prior to analysis, the Kolmogorove-Smirnov test was used to check all the data for normalcy. The data of development and reproduction of *T. granarium* in response to different stored products were tested using one-way ANOVA using SPSS 28.00 (IBM SPSS statistics 2021). In case of significance, Tukey test was used to separate means at level of α=0.05 .

RESULTS AND DISCUSSION

Results

Development

The immature life history of the khapra grain beetle, *T. granarium* on eight prducts is showed in Fig. (1). The incubation period was constant (3.6 d), and the pupal period was also quite stable (within a range of 7.1 - 7.8 d) in all diets tested. The longest period of the larval stage was 60.6±1.41 d on almonds and the shortest one was on cowpea (34.7±1.37 d). The longest developmental period (egg-adult) of *T. granarium* was recorded on almonds (72.1±1.65 d) and the shortest was on cowpea (45.4±1.47 d).

Survival rate of *T. granarium* on different diets is shown in Fig. (2). The survival rate of both larvae and pupae was the highest on faba bean, while the lowest e was when feeding on almond.

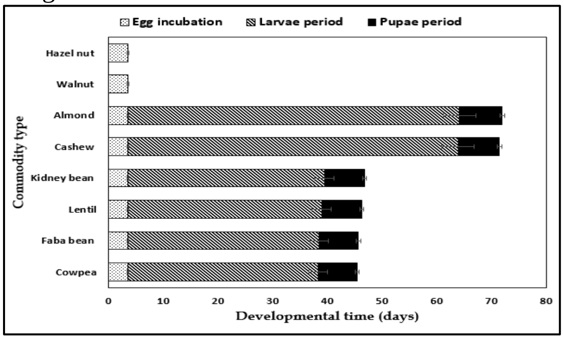

Fig. 1. Developmental periods of the Khapra grain beetle, *Trogoderma granarium* stages that reared on various stored leguminous diets.

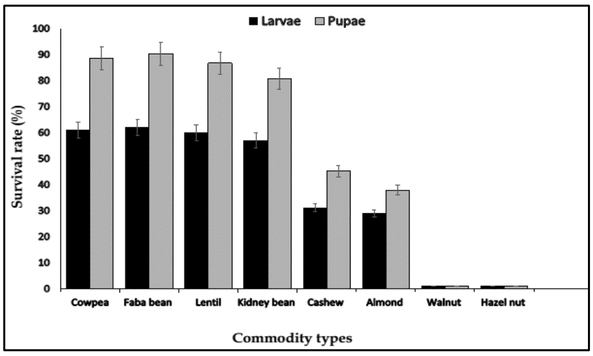

Fig. 2. Mean (±SE) survival rate of the larval and pupal stages of *Trogoderma granarium* that larvae fed on various diets

Reproduction

Table (1) show the reproductive parameters of *T. granarium* on the diets t used.

Table 1. Longevity (mean ± SE), fecundity (mean ± SE) and adult emergence (%) of the Khapra grain beetle, *Trogoderma granarium* reared on various cultivars of legume seeds and nuts

Diets	Types	Oviposition periods (days)			Female longevity (days)	Adult emergence rate (%)
		Pre-Oviposition	Oviposition	Post- Oviposition		
seeds	Cowpea	5.5±0.41 a	6.3±2.45 a	1.1±0.74 a	12.9 ± 3.6 a	0.54 a
	Faba bean	5.4±0.37 a	6.1±0.66 a	1.1±0.23 a	12.6 ± 1.26 a	0.56 a
	Lentil	5.3±0.34 a	5.7±0.63 ab	1.1±0.21 a	12.1 ± 1.18 a	0.52 a
	Coomon bean	5.6±0.45 a	5.4±0.52 ab	1.0±0.26 a	12.0 ± 1.23 a	0.46 ab
Nuts	Cashew	6.6±0.51 a	2.6±0.51 b	0.4±0.25 a	9.6 ± 1.27 b	0.14 b
	Almond	6.2±0.37 a	2.2±1.31 b	0.4±0.25 a	8.8 ± 1.93 b	0.11 b
	Walnut	-	-	-	0	0
	Hazel nut	-	-	-	0	0

Mean values in a column followed by different lowercase letters are significantly different on the basis of ANOVA with Tukey test (P < 0.05).

Significant differences were observed in the ovipositional periods, fecundity and adult emergence (%) of *T. granarium*; however, no significant differences were seen in pre-oviposition and post- oviposition periods of the khapra beetle on different stored diets. The total oviposition period of *T. granarium* was the highest on cowpea (12.9 d) and the lowest on almond (8.8 d). Statistical analysis revealed that there were significant differences in female longevity of khapra beetle on all diets, but it did not differ among seeds (Table 1). The highest fecundity was estimated on cowpea (11.9±1.1 eggs), and the lowest was on almonds (4.1±0.71 eggs) (Fig. 3). Furthermore, the adult emergence rate was the highest on cowpea (0.54), and the lowest on almonds (0.11) (Table. 1). Our results, it can suggest that rate was suitable diet for development and reproduction of the khapra beetle is cowpea seeds, but almonds seem to be rejected by the larvae as a feeding diet.

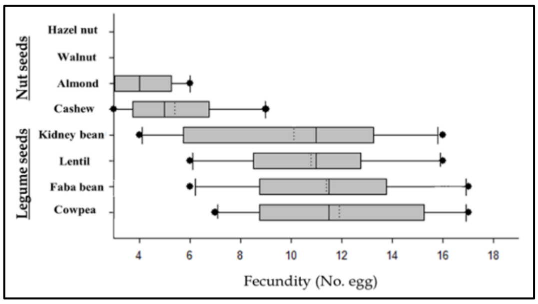

Fig. 3. Mean (±SE) of fecundity of the Khapra grain beetle, *Trogoderma granarium* that reared on various feeding diets.

Chemical analysis

Chemical analysis of the collected products is illustrated in Table (2).

Table 2. Biochemical properties in different leguminous diets used in feeding khapra beetle, *Togoderma granarium*.

Group	Seeds				Nuts			
Cultivar	Cowpea	Faba bean	Lentil	Kidney bean	Cashew	Almond	Hazelnuts	Walnuts
Proximate analysis (%)								
Moisture	11.81	7.18	8.91	9.11	7.18	3.91	2.78	6.94
Protein	23.18	31.06	32.11	21.11	20.81	19.36	17.46	19.21
Fat	3.11	1.46	0.91	3.03	35.18	64.18	50.17	44.18
Ash	3.22	3.06	2.43	4.11	2.74	3.18	1.48	3.47
Fiber	1.53	5.18	3.46	6.43	3.11	11.21	6.24	10.26
Total carbohydrates	58.68	57.24	55.64	62.64	34.09	9.37	28.11	26.20
Minerals (mg. / 100 gm.)								
P	15.61	62.11	284.61	131.78	13.88	18.41	306.26	189.11
K	21.63	713.32	863.43	711.03	28.64	249.14	491.33	357.21
Ca	21.82	65.36	122.18	514.47	22.16	20.88	551.72	71.17
Mg	7.91	7.11	73.29	374.81	18.94	27.23	483.67	171.18
Fe	0.81	14.48	2.94	4.73	0.59	0.66	7.18	4.33
Zn	0.46	5.03	3.98	3.82	0.78	0.98	2.23	1.93
Total phenol	113.77	13.54	95.58	58.85	176.19	112.92	109.84	2386.16
Oil %	-	-	-	-	48.91	62.53	63.61	51.76

The proportions of the internal components of the tested seeds and nuts varied, and the proximate analysis (%) varied among them. It is noted that the proportions of fat and fiber increased in almonds and the proportions of ash and total carbohydrates in kidney beans, while the proportions of moisture, protein, and ash were lowest in hazelnuts. Regarding minerals analysis (mg. / 100 gm.), the highest rates in total phenol and oil (%) were in walnuts and hazelnuts, respectively.

Discussion

Food compatibility and resistance to herbivorous insects can be impacted by dietary nutritional qualities(Lee *et al.*, 2002) Reduced growth, delayed development, low survivability, fecundity, and fertility have all been linked to phytophagous insects consuming a poor-quality diet (Lee, 2007; Borzoui *et al.*, 2015 and Borzoui *et al.*, 2017) Our findings showed that *T. granarium's* biological characteristics and life table parameters were influenced by the quality of the diet. A few recent research have looked into *T. granarium's* demographics (Golizadeh and Abedi, 2017 and Papanikolaou *et al.* 2019). The observed discrepancies in the results could be the consequence of several factors, including variations in the characteristics of the insect life table brought about by the diets, insect strains, and experimental procedures used (Jalali *et al.,* 2009 and Borges *et al.,* 2013). The mean generation time, or the average amount of time needed for a population to grow by a factor equal to the net reproductive rate, was lower when *T. granarium* fed on barley as opposed to the other commodities (Borges *et al.,* 2013). Food macronutrients, particularly those high in protein and carbohydrates, might modify the digestive physiology of post-harvest insects like *T. granarium*, hence affecting the appropriateness of the food for them (Bernardi *et al.*, 2012). In order to successfully develop and reproduce, *T. granarium* larvae employ a variety of methods to determine the macronutrient composition of various diets and modify the amounts of digestive enzymes (Naseri and Borzoui, 2016).

Although these parameters may occasionally play a role, the data that is currently available makes it abundantly evident that there was no consistent and direct relationship between population growth and grain size, protein, or oil content (Rao *et al.,* 2004). Regarding the nutritional variations across grains, *T. granarium* depends on protein, lipids, and carbs for growth and development. For instance, *T. granarium* development was adversely affected when essential amino acids in the diet were substituted with artificial ones, even though it has been demonstrated that members of this species are capable of synthesizing both essential and non-essential amino acids, such as proline, tyrosine, glycine, aspartic acid, alanine, and glutamic acid (Bhattacharya and Pant 1968). When lipids were absent from meals, larvae either never reached adulthood or just a small percentage of them did so after a protracted developmental period (Pant and Pant, 1961).

In this study, the effects of several hybrids on the *T. granarium* immature stages period were substantial; values ranged from 72.1±1.65 d on almond seed to 45.4±1.47 d on cowpea seed. (Golizadeh and Abedi, 2016) discovered that when *T. granarium* was raised on several wheat cultivars, there was a variation in its immature period. According to their findings, cultivar Gaskojen had the quickest development period from egg to adult (40.83 6 0.70 d), whereas cultivar Bezostaya had the longest (58.40 6 0.73 d). PL 472 showed the highest immature survival rate of *T. granarium* among the various hybrids of maize. Similar findings were evaluated by (Awadalla, *et al.* 2005), who identified Sakha 69 (a wheat cultivar) and Tri-H 324 (a maize cultivar) as the two most ideal hosts for raising *T. granarium* and showed substantial differences in survival rates among five maize hybrids. According to (Naseri and Borzoui, 2016) , *T. granarium* raised on nutritive diets (poor in proteinaceous inhibitors) exhibited a high survival rate of almost 90%. According to research by Borzoui *et al*., (2015) , inhibitors contained in the examined grains—barley, rice, rye, and wheat—as well as seed hardness were found to have a significant impact on larval survival.

The mating process of *T. granarium* can begin immediately after eclosion (Lindgren *et al.,* 1955), but because of its adults' brief lifespan, it is more quickly associated to other stored products Coleoptera (Benelli *et al.,* 2017). Any resource that inhibits or delays this process—for example, by changing the pattern of mating behavior or reducing the lifetime of adults—could prove to be an invaluable asset in the effective management of this species. Additional research is required to realize the accurate developmental potential of *T. granarium* by introducing additional grain species and varieties, as recent study (Papanikolaou *et al*., 2019) have shown under various cracking condition situations and abiotic components, specific temperature levels and commodities accelerate the pest's rapid population expansion.

CONCLUSION

This findings demonstrate potential adaptability of *T. granarium* following feeding on several stored products. The development plasticity observed provides information on the pest's capacity to adjust to changes in its diet. The appropriateness of the various products for *T. granarium* varied, and additional traits of the seeds (such as a-amylase, lipids, carbohydrates, protease inhibitors, and so on) must be investigated in order to elucidate the causes of resistance. This study indicates that the insects who consumed almonds had the longest immature period. Consequently, among the products examined, hazel nut and walnut seems to be inappropriate for development and hence reproduction of khapra beetle, and it can be advised that it be grown in locations where *T. granarium* is more likely to occur. By identifying a product resistant to *T. granarium*, this research will aid in the development of more workable plans for the long-term control of this pest.

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نمو وبقاء وانتاجية خنفساء الخابرا(خنفساء الصعيد) بالنسبة للمواد الغذائية المخزونة المختلفة وتركيبها الكيماوي

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الملخص

أصبحت العديد من الحبوب المخزونة والمنتجات الثانوية تصاب بحشرة خنفساء الخابرا عديدة العوائل. تم دراسة تأثير مجموعة من البذور البقولية (اللوبيا – الفول – العدس – الفاصوليا) وكذلك المكسرات (الكاجو – اللوز – عين الجمل – البذور) على نمو وبقاء وانتاجية خنفساء الخابرا على درجات الحرارة 30 م ، 65 % رطوبة نسبية وفترة اضاءة اليوم الطويل. علاوة على ذلك تم تحليل كل المكونات الكيميائية للمنتجات المختبرة. أوضحت النتائج أن المنتجات المختبرة أثرت على الخصائص البيولوجية لخنفساء الخابرا حيث اختلفت نسب المكونات الداخلية للمنتجات المختبرة طبقا للتحليل الكيماوي لها. ومن الناحية البيولوجية أتضح ان أطول فترة لدورة حياة خنفساء الخابرا(بيضة – حشرة كاملة) تم تقديرها على اللوز (72.1 ± 1.65 يوم) وان أقصر فترة كانت على بذور اللوبيا (45.4 ± 1.47 يوم) أظهرت النتائج أن أطول فترة لوضع البيض لخنفساء الخابرا كانت على بذور اللوبيا (12.9 يوم) وان أقل فترة كانت على اللوز (8.8 يوم) كما أوضحت النتائج أيضا أن أعلى معدلات الخصوبة الكلية وخروج الحشرات الكاملة كانت على بذور اللوبيا وان اقل معدلات كانت على اللوز في حين أن أعلى معدل بقاء لطور اليرقة و العذراء كان على بذور الفول بينما أقل معدل كان على اللوز وقد أشارت النتائج الى أن كلا من البندق وعين الجمل كانت أغذية غير مناسبة لنمو خنفساء الخابرا.