Egypt. Acad. J. Biolog. Sci., 12(1):125-136(2020)

Egyptian Academic Journal of Biological Sciences
G. Microbiology

ISSN: 2090-0872
https://eajbsg.journals.ekb.eg/

Chemical Composition of Five Botanical Powders and Their Insecticidal Activity Against the Rice Weevil, *Sitophilus oryzae* on Wheat Crop

Rady Shawer*, Mohamed Mahrous El-Shazly, Adel Mohamed Khider and Ahmed Mohamed Kordy
Plant Protection Department, Faculty of Agriculture (Saba Basha), Alexandria University, Alexandria 21531, Egypt
E.Mail: rady.shawer@alexu.edu.eg

ARTICLE INFO
Article History
Received: 2/5/2020
Accepted:30/6//2020

***Keywords*:**
Triticum aestivum, rice weevil, *Sitophilus oryzae*, botanical powder, GC-MS.

ABSTRACT

The rice weevil, *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae) is a key pest of stored cereals in Egypt and elsewhere. The extensive use of synthetic insecticides in its control programs for several decades has led to undesirable effects on humans, the environment and non-targeted organisms. Several plant-based materials have been found to be effective against *S. oryzae.* The insecticidal performance of botanical powders made from certain parts of Guava, *Psidium guajava*, Pomegranate, *Punica granatum* L., Snow thistle, *Sonchus oleraceus*, Thyme, *Thymus vulgaris* L. and Purslane, *Portulaca oleracea* at rates of 1, 2, 4, 6, 8 and 10 g/50 g wheat grains were evaluated against *Sitophilus oryzae* adults at 3, 7, 10 and 14 days after treatment (DAT). The chemical compositions of the oils extracted from these plant powders were characterized by gas chromatography coupled with mass spectrometry (GC-MS). The major constituents identified in the isolated oils of *P. guajava, P. granatum* L., *S. oleraceus, T. vulgaris* L. and *P. oleracea* powders were caryophyllene (24.34%), 2-furancarboxaldehyde, 5-(hydroxymethyl)- (50.76%), 9-Hexadecenoic acid (13.47%), phenol, 2-methyl-5-(1-methylethyl)- (24.28%) and 9,12-Octadecadienoic acid (Z,Z)- (13.04%), respectively. All the evaluated powders significantly caused *S. oryzae* adult mortality compared with controls at 3 DAT, even if at the lowest used application rate (1 g powder/50 g wheat grains). The application of *P. guajava* and *T. vulgaris* powders at 10 g l/50 g wheat grains showed the highest adult activity, giving the same excellent mortality (93%) at 14 DAT. This was followed by the treatments of *S. oleraceus, P. oleracea* and *P. granatum* powders, providing 91, 86 and 70% adult mortality at 14 DAT. These effective plant-based materials could be helpful in lowering chemical pesticides use and should be considered an effective IPM strategy for controlling *S. oryzae*.

INTRODUCTION

Wheat (*Triticum aestivum* L.) is one of the most important cultivated crops in Egypt and worldwide (Derbalah & Ahmed, 2011). It ranks second after maize in the world cereal outputs (ASA, HM Tayeb, A Masoud, & AA Shawer, 2011). There are many factors that limit its high production (ASA *et al.*, 2011) and cause considerable losses in the field and storage (Mahmoud & Zedan, 2018; Mehta & Kumar, 2020). Stored grain insect pests are the major reason for stored grains damage. They can cause postharvest losses in weight grains estimated at 9% in developed countries and more than 20% in developing countries (Mehta & Kumar, 2020; Phillips & Throne, 2010). The rice weevil, *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae) is the most widespread and destructive major insect pest of stored cereals all over the world, especially weight (Atwal & Dhaliwal, 1997; Mehta & Kumar, 2020). Their adults and larvae feed on the internal pulp of weight grains, affecting their quality and quantity (Gowda *et al.*, 2019; Suleman, Aslam, & Riaz, 2000). As a result, the temperature of the grain heaps in storage is increased. Consequently, the injured grains become then highly susceptible to the development of mold diseases (Soujanya, Sekhar, & Kumar, 2016). The use of synthetic chemical insecticides has been the main method to control *Sitophilus oryzae* on weight grains, as it is the simplest and most cost-effective means of dealing with such stored product pests (Derbalah & Ahmed, 2011; Khanal *et al.*, 2021; Mehta & Kumar, 2020). However, their extensive repeated applications for several decades have led to unfavorable effects regarding the development of insecticide resistance, the exceeding of maximum residue limits (MRLs) on weight grains and adverse action on the environment and beneficial micro-organisms (Govindan & Nelson, 2009; Gowda *et al.*, 2019; Khanal *et al.*, 2021; Shawer, Tonina, Tirello, Duso, & Mori, 2018). As a result, great attention has been recently focused on developing safe and non-traditional alternatives for such hazardous chemicals**.** Therefore, many approaches have targeted the investigation of plant-based materials for their biological performance against stored grains pests including *Sitophilus oryzae* (Mehta & Kumar, 2020)**.** Several plant-based materials have been found to have insecticidal actions as antifeedants, repellents, or toxic properties with little or no mammalian toxicity (Isman, 2006; Khanal et al., 2021; Kulkarni & Joshi, 1998). Many studies reported that different plant powders and extracts had strong effects including toxicity and inhibition of reproduction against stored grain insects (Asawalam, Ebere, & Emeasor, 2012; Demeter *et al.*, 2021; Huang, Ho, Lee, & Yap, 2002; Ismail *et al.*, 2021; Isman, 2006; Kemabonta & Falodu, 2013; Rajendran & Sriranjini, 2008; Ziaee & Moharramipour, 2013). The essential oil of pomegranate, *Punica granatum* L. extracted from fruit peels showed an effective antifeedant against *S. oryzae* (Wahba, 2020). In Nigeria, botanical powders of *Curcuma longa* L., *Dennettia tripetala* Baker, *Piper guineense*, and *Zingiber officinale* caused an acceptable adult mortality and suppressed the adult emergence of *S. oryzae* under in laboratory studies (Asawalam et al., 2012). The extracts of Snow thistle, *Sonchus oleraceus* had anti-microbial activity against some bacteria such as *Pseudomonas aeruginosa, Escherichia coli* and *Salmonella typhi* (Kaundal, Sharma, & Kulshrestha). The anti-microbial effect depended on the extract concentration. Several studies confirm that the oil of thyme, *Thymus vulgaris* L. has carminative, antimicrobial and antioxidant activities (Boskabady, Aslani, & Kiani, 2006; Lee, Umano, Shibamoto, & Lee, 2005; Schwarz, Ernst, & Ternes, 1996). In the current study, the insecticidal activity of botanical powders made from certain parts of Guava, *Psidium guajava,* Pomegranate, *Punica granatum* L., Snow thistle, *Sonchus oleraceus,* Thyme, *Thymus vulgaris* L. and Purslane, *Portulaca oleracea* were investigated against *Sitophilus*

oryzae. The chemical compositions of the oils extracted from these plant powders were identified as well.

MATERIALS AND METHODS

1. Insects Colony:

The insects of *S. oryzae* used in the present study were provided from susceptible strains reread in the laboratory of the Plant Protection Department, Faculty of Agriculture (Saba-Basha), Alexandria University, Egypt. Four hundred adults (mixed sexes and ages) were added into 2 L glass jars with 500 g sterilized rearing medium (wheat grains var. Sakha 68 in case of *S. oryzae* and wheat flour, bran and dry yeast at a rate of 17:5:1, respectively in case of *T. castaneum*) (Saad *et al.,* 2018; E. H. Tayeb and Metraw 2017). The inside upper part of jars (8-10 cm) was coated with a thin layer of Vaseline to prevent adults from getting out. Jars were wrapped with pieces of muslin to ensure good aeration (Mackled *et al.*, 2019). After one week, adults were taken away using 10-mesh sieves and discarded. The jars were secured in a controlled environmental condition (28ºC ± 2ºC and 70±5% RH.) till adults emerged (about 5 to 6 weeks) (Mehta & Kumar, 2020). The homogenized emerging Weevils (2-3 weeks age) were used in further bioassays (Abdelgaleil, Badawy, Shawir, & Mohamed, 2015; Mehta & Kumar, 2020).

2.Botanical Powders Preparation:

Fresh plant materials of *Psidium guajava, Thymus vulgaris, Sonchus oleraceus, Punica granatum and Portulaca oleracea* were collected from distinctive farms located in the Abees region, Alexandria governorate, Egypt and transferred to the laboratory of Plant Protection Department, Faculty of Agriculture (Saba Basha), Alexandria University. Botanical powders were prepared from the leaves of *P. guajava, T. vulgaris* and *P. oleracea,* the fruit peels of *P. granatum* and the aerial parts of *S. oleraceus*. The plant materials were washed with distilled water, dried in the open air and thereafter, put in an oven at 60°C for complete dryness. The dried parts of each plant were ground by an electric mill and were sieved in 100-mesh sieves (Mahmoud & Zedan, 2018; Mehta & Kumar, 2020). The powders were sieved by 150-mesh sieves and the powders that remained over the sieve (>150 µm) were preserved in dry bags to be used in toxicity bioassays or the extraction of the essential oils.

3. Chemical Compositions of Plant Materials:

The chemical composition of the used plant materials was estimated in their oil extracts (Khanal *et al.,* 2021). About 250 g of each prepared powder was sent to hydro-distillation for 6 h using the Clevenger type apparatus (Nenaah & Ibrahim, 2011). The yielded oils were filtrated and dried over anhydrous sodium sulfate. The chemical composition of obtained oils was estimated by employing a thermo logical gas chromatograph GC Follow 1300 coupled with an El-Mass spectrometer ISQ7000 show (Thermo Logical USA) prepa red with Thermo TR-50 MS capillary column (30 m in length x 250 µm in breadth x 0.25 µm in thickness of film) (Tapondjou, Adler, Bouda, & Fontem, 2002). Spectroscopic location by GC-MS included an electron ionization frame work the utilized tall vitality electrons (70eV), MS exchange line temperature of 300°C. Unadulterated helium gas (99.995%) was utilized as the carrier gas with a stream rate of 1 ml/min. the beginning temperature was set at 60°C for 2 min, at that point expanded to 100 °C at a rate of 10°C/min kept for 5 min, at that point with 10°C/min to 150 °C and kept for 5 min, at that point with 10°C/min to 200°C and kept for 5 min, and kept for 20 min. One at that point with 10°C/min to 250°C and kept for 20 min. One microliter of the arranged extricates was infused in a part less mode.

4. Bioassay Technique:

The efficacy of botanical powders of the leaves of *Psidium guajava, Thymus vulgaris* and *Portulaca oleracea*, the fruit peels of *Punica granatum* and the aerial parts of *Sonchus oleraceus* at rates of 1, 2, 4, 6, 8 and 10 g/50 g wheat grains were evaluated

against *S. oryzae* adults. Plant powders were added in 2 L glass jars and well-mixed with 50 g. Twenty homogenized adults of *S. oryzae* (same age, mixed-sex) were placed in each jar containing 50 g sterilized weight grains well-mixed with the botanical powders (Mehta & Kumar, 2020). Treatments were replicated fifth. A treatment without plant powders acted as control. Jars were daily checked and adult mortality was recorded at 3, 7, 10 and 14 days after treatment (DAT) and was corrected according to Abbott's formula (Abbott 1925) as follows:

$$\text{Corrected mortality\%} = \frac{(\text{Mortality\% of treated insects} - \text{Mortality\% of control})}{(100 - \text{Mortality\% of control})} \times 100$$

5. Statistical Analysis:

Data of the adult mortality were statistically analyzed by the generalized linear model (GLM) using one-way analysis of variance (ANOVA) to compare the significance of differences between treatments. The percentages of adult mortality were first transformed to arcsine (sqrt (%mortality)) before analysis to stabilize variance and resulted in means being back-transformed to percentages to be presented. Means were compared by determining the Duncan's least significant difference (LSD) test between treatments (Duncan, 1955) using CoStat software. Differences were considered significant at $\alpha = 0.05$.

RESULTS AND DISCUSSION

1. Chemical Compositions of Plant Materials:

The chemical constituents identified in the isolated oils of *Psidium guajava*, *Punica granatum* L., *Thymus vulgaris* L., *Sonchus oleraceus*, *Portulaca oleracea* powders are listed in Table 3. The major constituent found in *P. guajava* was Caryophyllene (24.34%), followed by 1,6,10-Dodecatrien-3-ol, 3,7,11-trimethyl- (7.67%), Aromadendrene (7.43%), 1H-Benzocycloheptene, 2,4a,5,6,7,8,9,9a-octahydro-3,5,5-trimeth yl-9-methylene (5.57%), Globulol (5.55%) and 9-Eicosyne (4.47%). In a recent study, limonene, 1,8-Cineole and caryophyllene at concentrations of 54.7, 32.14 and 2.91% were identified, respectively in the oils extracted from guava leaves (Kumar et al., 2021). The main compounds characterized in the fruit peels of *P. granatum* were 2-furancarboxaldehyde, 5-(hydroxymethyl)- (50.76%), furfural (14.43%), 9-oxabicyclo[6.1.0]non-6-en-2-one (6.73%), 4h-pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl (5.65%) and oleic acid (3.11%). (Wahba, 2020) identified linoleic-acid (29.33%), D-Limonene (13.79%), Caryophyllene (13.9%), cis-Vaccenic acid (12.66%), and squalene (9.12%) in the fruit peels of *P. granatum*. While, the main compounds found in the oil of *S. oleraceus* were 9-Hexadecenoic acid (13.47%), phytol (12.5%), caryophyllene (7.54%), 1,2-15,16-Diepoxyhexadecane (7.42%) and cyclohexene, 1-methyl-4-(5-methyl-1-methylene-4-hex enyl)-, (S)- (5.41%). The extracts of *S. oleraceus* were effective against some bacteria including *Klebsiella pneumoniae*, *Staphylococcus aureus, Pseudomonas aeruginosa, Escherichia coli* and *Salmonella typhi*. This anti-bacterial activity has been attributed to the presence of alkaloids, flavonoids, protein, carbohydrates, tannins, phenol, terpenoids and saponins in these extracts (Kaundal et al., 2021). The major constituents of *T. vulgaris* extracts were phenol, 2-methyl-5-(1-methylethyl)- (24.28%), 1,5-Heptadien-4-one, 3,3,6-trimethyl- (15.28%), 2-Naphthalenemethanol, decahydro-à, à,4a-trimethyl-8-methylene-, [2R-(2à,4aà,8aŕ)]- (11.88%) and Ledol (11.05%). Many previous studies proved the carminative, antimicrobial and antioxidant effects of *T. vulgaris* (Al-Asmari, Athar, Al-Faraidy, & Almuhaiza, 2017; Boskabady et al., 2006; Lee et al., 2005; Schwarz et al., 1996). A previous study reported the presence of caryophyllene at a concentration of 0.91%; however, it was identified in the current study at 0.87% (Al-Asmari et al., 2017; Dahham et al., 2015). The main compounds characterized in the oil of *P. oleracea* were 9,12-Octadecadienoic acid (Z, Z)- (13.04%), n-Hexadecanoic acid (11.89%),

Oleic Acid (10.77%), 8,11,14-Eicosatrienoic acid, (Z,Z,Z)- (6.15%) and 2-Pentadecanone, 6,10,14-trimethyl- (5.23%). It has been reported that oxalic and citric acids are the most abundant organic acids in purslane (Petropoulos *et al.,* 2015).

Table 1. The main constituents identified in oils extracted from certain botanical parts of *P. guajava, P. granatum, S. oleraceus, T. vulgaris* and *P. oleracea.*

RT[a]	Compound[b]	Area %				
		P. guajava	***P. granatum***	***S. oleraceus***	***T. vulgaris***	***P. oleracea***
3.13	1,2-Ethanediol, monoformate	-	1.58	-	-	-
5.17	Bicyclo[2.1.1]hexan-2-ol, 2-ethenyl-	-	-	-	-	1.01
6.15	Furfural	-	14.43	-	-	-
7.27	Limonene	3.14	-	-	-	-
7.91	Benzene, 1-methyl-2-(1-methylethyl)-		-	-	2.39	-
7.92	Eucalyptol	1.34	-	-	-	-
8.9	1,5-Heptadien-4-one, 3,3,6-trimethyl-	-	-	-	15.28	-
10.91	Desulphosinigrin	-	-	-	-	2.34
11.89	Benzeneethanamine, à-methyl-	-	-	-	-	1.47
13.57	Furyl hydroxymethyl ketone	-	1.76	-	-	-
14.76	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl	-	5.65	-	-	-
15.24	2-Hexadecanol	-	-	-	-	1.36
15.45	Hepta-2,4-dienoic acid, methyl ester	-	2.31	-	-	-
16.09	2-Myristynoyl pantetheine	-	-	-	-	1.14
16.28	Copaene	1.33	-	-	-	-
16.34	7-Methyl-Z-tetradecen-1-ol acetate	-	-	1.13	-	-
16.36	Cholestan-3-ol, 2-methylene-, (3ƒ,5à)-	-	-	-	-	1.2
16.39	Pyrimidine, 4-chloro-5-ethoxy-2-methyl-	-	2.79	-	-	-
17.31	Phenol, 2-methyl-5-(1-methylethyl)-	-	-	-	24.28	-
17.79	6-Acetyl-ƒ-d-mannose	-	1.23	-	-	-
17.79	Caryophyllene	-	-	7.54	-	-
17.81	Caryophyllene	24.34	-	-	-	-
18.12	Aromadendrene	-	-	3.29	-	-
18.14	Aromadendrene	7.43	-	-	-	-
18.21	2-Furancarboxaldehyde, 5-(hydroxymethyl)-	-	-	-	1.5	-
18.46	2-Furancarboxaldehyde, 5-(hydroxymethyl)-	-	50.76	-	-	-
18.99	5,8,11,14-Eicosatetraenoic acid, methyl ester, (all-Z)-	-	-	1.7	-	-
19	à-Caryophyllene	3.52	-	-	-	-
20.11	Cyclohexene, 1-methyl-4-(5-methyl-1-methylene-4-hex enyl)-, (S)-		-	5.41	-	-
20.14	1H-Benzocycloheptene, 2,4a,5,6,7,8,9,9a-octahydro-3,5,5-trimeth yl-9-methylene	5.57	-	-	-	-
20.21	cis-à-Bisabolene	3.67	-	-	-	-
20.71	ç-Elemene	1.69	-	-	-	-
22.02	Epiglobulol	-	-	-	1.15	-
22.16	Cedran-diol, 8S,14-	-	-	-	1.6	-
22.39	6,9-Octadecadiynoic acid, methyl ester	-	-	1.12	-	-
22.58	1,6,10-Dodecatrien-3-ol, 3,7,11-trimethyl-	7.67	-	-	-	-
22.91	Epiglobulol	1.09	-	-	-	-
23.83	Globulol	5.55	-	-	-	-
24.05	trans-Z-à-Bisabolene epoxide	1.15	-	-	-	-
24.09	1H-Cycloprop[e]azulen-7-ol, decahydro-1,1,7-trimethyl-4-methylene-, [1ar-(1aà,4aà,7ƒ,7aƒ,7bà)]-	-	-	-	1.4	-
24.21	Caryophyllene oxide	-	-	1.82		-
24.22	Caryophyllene oxide	-	-	-	1.34	-
24.22	Caryophyllene oxide	2.37	-	-		-
25.36	.tau.-Muurolol	-	-	-	2.21	-
25.53	Tetracyclo[6.3.2.0(2,5).0(1,8)]tridecan-9- ol, 4,4-dimethyl	-	-	-	2.23	-
25.54	Tetracyclo[6.3.2.0(2,5).0(1,8)]tridecan-9- ol, 4,4-dimethyl	3.39	-	-	-	-

25.66	2-Butyl-5-methyl-3-(2-methylprop-2-enyl)cyclohexanone	-	-	1.8	-	-
25.66	2(4H)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4,7a-trimethyl-, (R)-	-	-	-	-	2.53
25.89	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	-	-	-	-	2.42
25.9	Phytol	-	-	12.5	-	-
25.91	9-Eicosyne	4.47	-	-	-	-
26.23	Ledol	-	-	-	11.05	-
26.43	Ethanol, 2-(9-octadecenyloxy)-, (Z)-	-	-	2.23	-	-
26.71	9-Eicosyne	-	-	3.81	-	-
27.07	[1,1'-Bicyclopropyl]-2-octanoic acid, 2'-hexyl-, methyl ester	-	-	1.31	-	-
27.07	2-Pentadecanone, 6,10,14-trimethyl-	-	-	-	-	5.23
28.63	(3-Methyl-1,4-diphenylbicyclo[2.2.0]hex2-yl)methano	-	-	1.17	-	
28.95	Hexadecanoic acid, methyl ester	-	-	-	-	3.49
29.52	Desulphosinigrin	-	-	-	2.91	-
29.66	[1,1'-Bicyclopropyl]-2-octanoic acid, 2'-hexyl-, methyl ester	-	-	-	2.28	-
30.14	Diepicedrene-1-oxide	-	-	-	1.77	-
30.46	2-Naphthalenemethanol, decahydro-à,à,4a-trimethyl-8-methylene-, [2R-(2à,4aà,8aF)]-	-	-	-	11.88	-
30.52	Oleic Acid	-	-	2.35	-	-
30.57	n-Hexadecanoic acid	-	-	-	-	11.89
30.58	n-Hexadecanoic acid	-	1.84	-	-	-
30.71	Cholestan-3-ol, 2-methylene-, (3F,5à)-	-	-	-	1.5	-
31.45	1-Isobutyl-7,7-dimethyl-octahydro-isoben zofuran-3a-ol	-	1.4	-		-
31.51	Hydrazinecarboxamide, 2-(2-methylcyclohexylidene)-	-	1.33	-		-
31.76	2,5,5,8a-Tetramethyl-4-methylene-6,7,8,8 a-tetrahydro-4H,5H-chromen-4a-yl hydroperoxide	-	-	-	1.09	-
32.72	2,5,5,8a-Tetramethyl-4-methylene-6,7,8,8 a-tetrahydro-4H,5H-chromen-4a-yl hydroperoxide	-	-	2.13	-	-
32.73	2,5,5,8a-Tetramethyl-4-methylene-6,7,8,8 a-tetrahydro-4H,5H-chromen-4a-yl hydroperoxide	-	-	-	-	2.06
33.18	1,2-15,16-Diepoxyhexadecane	-	-	7.42	-	
33.48	7-Hexadecenoic acid, methyl ester, (Z)-	-	-	-	-	3.81
33.51	10-Octadecenoic acid, methyl ester	-	1.39	-	-	-
33.79	Cyclopropaneoctanoic acid, 2-[[2-[(2-ethylcyclopropyl)methyl]cyclopr opyl]methyl]-, methyl ester	-	-	-	-	4.35
33.85	Acetic acid, 3-hydroxy-6-isopropenyl-4,8a-dimethyl-1 ,2,3,5,6,7,8,8a-octahydronaphthalen-2-yl ester	-	-	-	1	-
34.28	9,12,15-Octadecatrienoic acid, 2,3-dihydroxypropyl ester, (Z,Z,Z)-	-	-	1.2	-	-
34.28	9,12,15-Octadecatrienoic acid, methyl ester, (Z,Z,Z)-	-	-		-	4.03
34.66	9-Octadecenoic acid, (2-phenyl-1,3-dioxolan-4-yl)methyl ester, cis	-	-	1.55	-	
34.72	Oleic Acid	-	-	-	-	10.77
34.73	Oleic Acid	-	3.11	-	-	
34.99	9,12-Octadecadienoic acid (Z,Z)-	-	-	-	-	13.04
35.4	Nonanoic acid, 9-(3-hexenylidenecyclopropylidene)-, 2-hydroxy-1-(hydroxymethyl)ethyl ester, (Z,Z,Z)-	-	-	3.31	-	-
35.44	8,11,14-Eicosatrienoic acid, (Z,Z,Z)-	-	-	-	-	6.15
35.47	10-Methyl-8-tetradecen-1-ol acetate	-	-	-	2.02	-
36.15	2-Myristynoyl pantetheine	-	-	1.52	-	-
36.54	5,8,11,14-Eicosatetraenoic acid, methyl ester, (all-Z)-	-	-	-	-	1.11
37.15	9-Oxabicyclo[6.1.0]non-6-en-2-one	-	6.73	-	-	-
37.75	9,12-Octadecadienoyl chloride, (Z,Z)-	-	-	-	-	1.41
38.05	12-Methyl-E,E-2,13-octadecadien-1-ol	-	-	-	-	1.59

38.43	10-Methyl-8-tetradecen-1-ol acetate	-	-	-	-	1.19
40.03	Pseudosolasodine diacetate	-	-	2.44	-	-
42.25	2,5-Octadecadiynoic acid, methyl ester	-	-	1.27	-	-
47.5	Squalene	2.16	-	-	-	-
48.85	2(1H)Naphthalenone, 3,5,6,7,8,8a-hexahydro-4,8a-dimethyl-6-(1-methylethenyl)-	1.65	-	-	-	-
48.94	Acetic acid, 3-hydroxy-7-isopropenyl-1,4a-dimethyl-2 ,3,4,4a,5,6,7,8-octahydronaphthalen-2-yl ester	1.23	-	-	-	-
48.98	Androstan-17-one, 3-ethyl-3-hydroxy-, (5à)-	-	-	-	1.17	-
49.84	9-Hexadecenoic acid	-	-	13.47	-	-
53.46	Cholesta-8,24-dien-3-ol, 4-methyl-, (3F,4à)-	-	-	1.25	-	-
54.23	Hexadecane, 1,1-bis(dodecyloxy)-	-	-	1.55	-	-

[a] Retention time.

[b] Compounds are listed in the order of their elution.

2. Bioassay results

All the tested application rates of *Psidium guajava* leaves powder were significantly effective against *S. oryzae* adults throughout the whole experiment compared with controls even when the powder was used at a rate of 1 g (Table 2). The powder caused significant adult mortality than the untreated from the third day after treatment. The lower application rates of the powder (1, 2 and 4 g) showed moderate mortalities ranging from 22 to 69% at 3, 7, 10 and 14 DAT. While the highest two application rates (8 and 10 g) provided excellent adult mortality (83-93%) at 7, 10 and 14 DAT. Overall, the highest activity was caused when the powder was used at 10 g/50 g weight grains, giving 65, 84, 87 and 93% adult mortality at 3, 7, 10 and 14 DAT, respectively.

The *Punica granatum* powder was significantly able to cause *S. oryzae* adult mortality even if at a low application rate (1 g/ 50g weight grains) at 3 DAT (Table 3). All treatments showed significant mortality at 3, 7, 10 and 14 DAT compared with control treatments. The highest adult mortality (70%) was achieved at 14 DAT by using the botanical powder at a rate of 10 g/ 50g weight grains.

The mixing of *Sonchus oleraceus* powder with weight grains significantly reduced the number of *S. oryzae* adults at all the evaluated application rates compared with the controls throughout the whole experiment (Table 4). At 3 DAT, all treatments caused significant mortality compared with the control treatment. The first three-application rates (1, 2 and 4 g) showed almost low and moderate adult mortality (10 -67%) throughout the experiment. The most effective treatment was performed by the application rate of 10 g, giving 35, 60, 80 and 91% adult mortality at 3, 7, 10 and 14 DAT, respectively.

The powder of *Thymus vulgaris* leaves significantly reduced the number of *S. oryzae* adults on all recorded days after treatments (Table 5). In comparison with controls, there were significant differences between all treatments at 3, 7, 10 and 14 DAT ($P< 0.05$). The most effective results were significantly caused by the application of 10 g/ 50 g weight grains, giving moderate mortality at 3 DAT (55%) and an excellent activity (88-93%) at 7, 10 and 14 DAT.

The powder of *Portulaca oleracea* leaves significantly caused *S. oryzae* adult mortality compared with the untreated throughout the whole trial (table 6). There were significant differences between all treatments and controls ($P< 0.05$) at 3, 7, 10 and 14 DAT. The lowest-three treatments (1, 2 and 4 g) gave a weak activity (10-21%adult mortality) at all recorded DAT; however, they significantly reduced adult numbers over the controls. The best results were shown by the application of the highest rate (10 g), giving 57, 76, 80 and 86 % mortality at 3, 7, 10 and 14 DAT, respectively.

The obtained results confirmed the significant activity of all evaluated botanical powders against *S. oryzae* adults even if at a low application rate (1 g/50 g wheat grains). These powders had a quick adult mortality

action as well (at 3 DAT). Those results are with the same trend as those previously obtained (Sharaby, 1988; Wahba, 2020). The contact toxicity of different essential oils extracted by hydrodistillation from different plants including *P. guajava* was studied against *S. oryzae* adults (Saad, El Gendy, Elkhateeb, & Abdelgaleil, 2022). The oil extract of *P. guajava* showed acceptable contact toxicity against *S. oryzae*, causing a lethal concentration for 50% of the tested insects (LC50) value of 0.12 mg/cm2 at 1 DAT. It is reported that plant extract of *P. guajava* leaves had excellent repellent effects against *S. oryzae* (Akhtar *et al.*, 2013). Other results proved the strong antimicrobial properties of *T. vulgaris* essential oil (Borugă et al., 2014; El-Refai, Sharaf, Azzaz, & El-Dengawy, 2020; Lee *et al.*, 2005; Rota, Herrera, Martínez, Sotomayor, & Jordán, 2008) and *S. oleraceus* (Kaundal *et al.*, 2021; Li *et al.*, 2015; Xia, Yu, Zhu, & Zou, 2011) against different bacteria. However, its antimicrobial performance depends on its chemical compositions (Borugă *et al.*, 2014).

Table 2. Mortality of *S. oryzae* adults treated with the powder of *Psidium guajava* leaves

Treatment (g/50 g grains)	**Mortality % ± SD**			
	Days after treatment			
	3	7	10	14
1	22.0±1.34^{c}	31.0±2.17^{d}	33.0±1.67^{d}	40.0±2.83^{c}
2	28.0±1.34^{c}	40.0±1.22^{d}	48.0±0.54^{c}	56.0±0.45^{bc}
4	37.0±1.34^{b}	53.0±2.51^{c}	59.0±3.11^{b}	69.0±3.65^{ab}
6	42.0±1.34^{b}	68.0±0.89^{b}	84.0±1.30^{a}	93.0±0.55^{a}
8	60.0±1.22^{a}	83.0±1.52^{a}	87.0±1.22^{a}	93.0±0.55^{a}
10	65.0±1.22^{a}	84.0±1.10^{a}	87.0±1.22^{a}	93.0±0.55^{a}
Control	0.00±0.00^{d}	0.00±0.00^{e}	0.00±0.00^{e}	0.00±0.00^{d}

Means within each column followed by the same letter are not significantly different (Duncan's LSD test; p=0.05).

Table 3. Mortality of *S. oryzae* adults treated with the powder of *Punica granatum* fruit-peels

Treatment (g/50 g grains)	**Mortality % ± SD**			
	Days after treatment			
	3	7	10	14
1	7.00±0.54^{e}	15.0±0.70^{e}	20.0±0.70^{f}	27.0±0.70^{e}
2	11.0±1.10^{de}	19.0±0.71^{e}	30.0±0.70^{e}	40.0±0.70^{d}
4	15.0±0.71^{d}	25.0±0.71^{d}	35.0±0.71^{d}	42.0±0.71^{d}
6	26.0±0.70^{c}	40.0±0.71^{c}	45.0±.0.84^{c}	50.0±0.71^{c}
8	35.0±0.71^{b}	51.0±0.71^{b}	55.0±0.71^{b}	60.0±0.55^{b}
10	40.0±0.70^{a}	60.0±0.55^{a}	65.0±0.85^{a}	70.0±0.71^{a}
Control	0.00±0.00^{f}	0.00±0.00^{f}	0.00±0.00^{g}	0.00±0.00^{f}

Means within each column followed by the same letter are not significantly different (Duncan's LSD test; p=0.05).

Table 4. Mortality of *S. oryzae* adults treated with the powder of *Sonchus oleraceus* aerial parts

Treatment (g/50 g grains)	Mortality % ± SD			
	Days after treatment			
	3	7	10	14
1	10.0±0.70d	20.0±2.24e	37.0±0.71e	50.0±0.71d
2	11.0±0.83d	24.0±0.71e	50.0±0.71d	63.0±0.84c
4	20.0±2.24c	35.0±2.32d	60.0±1.22c	67.0±1.22c
6	25.0±1.34b	42.0±0.71c	70.0±0.70b	80.0±1.10b
8	32.0±1.79a	55.0±0.71b	76.0±0.71b	85.0±1.22b
10	35.0±2.35a	60.0±1.22a	80.0±1.10a	91.0±0.55a
Control	0.00±0.00^{e}	0.00±0.00^{f}	0.00±0.00^{f}	0.00±0.00^{e}

Means within each column followed by the same letter are not significantly different (Duncan's LSD test; p=0.05).

Table 5. Mortality of *S. oryzae* adults treated with the powder of *Thymus vulgaris* leaves

Treatment (g/50 g grains)	Mortality % ± SD			
	Days after treatment			
	3	7	10	14
1	15.0±0.71^{e}	35.0±2.35^{e}	55.0±0.71^{e}	70.0±0.70^{d}
2	25.0±0.71^{d}	50.0±0.71^{d}	65.0±1.22^{d}	75.0±0.70^{c}
4	35.0±2.35^{c}	65.0±0.71^{c}	70.0±0.70^{c}	75.0±0.70^{c}
6	40.0±0.71^{b}	70.0±0.84^{b}	75.0±0.70^{b}	80.0±1.10^{b}
8	55.0±0.70^{a}	88.0±1.22^{a}	93.0±0.55^{a}	93.0±0.55^{a}
10	55.0±0.70^{a}	88.0±1.22^{a}	93.0±0.55^{a}	93.0±0.55^{a}
Control	0.00±0.00^{f}	0.00±0.00^{f}	0.00±0.00^{f}	0.00±0.00^{e}

Means within each column followed by the same letter are not significantly different (Duncan's LSD test; p=0.05).

Table 6. Mortality of *S. oryzae* adults treated with the powder of *Portulaca oleracea* leaves

Treatment (g/50 g grains)	Mortality % ± SD			
	Days after treatment			
	3	7	10	14
1	10.0±0.70^{d}	12.0±0.54^{d}	14.0±0.83^{c}	16..0±0.83^{c}
2	12.0±0.45^{d}	15.0±0.83^{d}	15.0±0.70^{c}	19.0±1.30^{c}
4	12.0±0.45^{d}	16.0±0.84^{d}	17.0±0.84^{c}	21.0±1.20^{c}
6	25.0±0.70^{c}	55.0±0.70^{c}	65.0±0.70^{b}	75.0±0.70^{b}
8	40.0±0.71^{b}	70.0±0.71^{b}	75.0±0.71^{a}	80.0±1.60^{b}
10	57.0±0.45^{a}	76.0±0.84^{a}	80.0±0.71^{a}	86.0±0.84^{a}
Control	0.00±0.00^{e}	0.00±0.00^{e}	0.00±0.00^{d}	0.00±0.00^{d}

Means within each column followed by the same letter are not significantly different (Duncan's LSD test; p=0.05).

REFERENCES

Abdelgaleil, S., Badawy, M., Shawir, M., & Mohamed, M. (2015). Chemical composition, fumigant and contact toxicities of essential oils isolated from Egyptian plants against the stored grain insects; Sitophilus oryzae L. and Tribolium castaneum (Herbst). *Egyptian Journal of Biological Pest Control*, *25*(3), 639.

Akhtar, M., Arshad, M., Raza, A. B. M., Chaudhary, M. I., Iram, N., Akhtar, N., & Mahmood, T. (2013). Repellent effects of certain plant extracts against

rice weevil, Sitophilus oryzae L.(Coleoptera: Curculionidae). ***International Journal*** of ***Applied*** Agricultural Sciences, *Vol, 5*(1).

Al-Asmari, A. K., Athar, M. T., Al-Faraidy, A. A., & Almuhaiza, M. S. (2017). Chemical composition of essential oil of Thymus vulgaris collected from Saudi Arabian market. *Asian Pacific Journal of Tropical Biomedicine, 7*(2), 147-150.

ASA, S., HM Tayeb, E., A Masoud, M., & AA Shawer, R. (2011). Comparative performance of wheat post-emergence herbicides in relation to their effect on wheat yield. *Alexandria Science Exchange Journal*, 32(OCTOBER-DECEMBER), 442-452.

Asawalam, E., Ebere, U., & Emeasor, K. (2012). Effect of some plant products on the control of rice weevil Sitophilus oryzae (L.) Coleoptera: Curculionidae. *Journal of Medicinal Plants Research,* 6(33), 4811-4814.

Atwal, A., & Dhaliwal, G. (1997). Agricultural pests of South Asia and their management. *Journal Of Entomological Research,* 21, 389-390.

Borugă, O., Jianu, C., Mişcă, C., Goleţ, I., Gruia, A., & Horhat, F. (2014). Thymus vulgaris essential oil: chemical composition and antimicrobial activity. *Journal of medicine and life, 7*(Spec Iss 3), 56.

Boskabady, M. H., Aslani, M., & Kiani, S. (2006). Relaxant effect of Thymus vulgaris on guinea-pig tracheal chains and its possible mechanism (s). *Phytotherapy Research: An International Journal Devoted to Pharmacological and Toxicological Evaluation of Natural Product Derivatives,* 20(1), 28-33.

Dahham, S. S., Tabana, Y. M., Iqbal, M. A., Ahamed, M. B., Ezzat, M. O., Majid, A. S., & Majid, A. M. (2015). The anticancer, antioxidant and antimicrobial properties of the sesquiterpene β-caryophyllene from the essential oil of Aquilaria crassna. *Molecules*, 20(7), 11808-11829.

Demeter, S., Lebbe, O., Hecq, F., Nicolis, S. C., Kenne Kemene, T., Martin, H., . . . Hance, T. (2021). Insecticidal Activity of 25 Essential Oils on the Stored Product Pest, Sitophilus granarius. *Foods*, 10(2), 200.

Derbalah, A., & Ahmed, S. (2011). Oil and powder of spearmint as an alternative to Sitophilus oryzae chemical control of wheat grains. *Journal of plant protection research*,51(2),145-150.

Duncan, D. B. (1955). Multiple range and multiple F tests. *Biometrics,* 11(1), 1-42.

El-Refai, A., Sharaf, A., Azzaz, N., & El-Dengawy, M. (2020). Antioxidants and Antibacterial Activities of Bioactive Compounds of Clove (Syzygium aromaticum) and Thyme (Tymus vulgaris) Extracts. *Journal of Food and Dairy Sciences,* 11(9), 265-269.

Govindan, K., & Nelson, S. J. (2009). Insecticidal activity of twenty plant powders on mortality, adult emergence of Sitophilus oryzae L. and grain weight loss in paddy. *Journal of Biopesticides,* 2(2), 169-172.

Gowda, G. B., Patil, N. B., Adak, T., Pandi, G. P., Basak, N., Dhali, K., . . . Jena, M. (2019). Physico-chemical characteristics of rice (Oryza sativa L.) grain imparting resistance and their association with development of rice weevil, Sitophilus oryzae (L.)(Coleoptera: Curculionidae). *Environmental Sustainability*, 2(4), 369-379.

Huang, Y., Ho, S.-H., Lee, H.-C., & Yap, Y.-L. (2002). Insecticidal properties of eugenol, isoeugenol and methyleugenol and their effects on nutrition of Sitophilus zeamais Motsch.(Coleoptera: Curculionidae) and Tribolium castaneum (Herbst)(Coleoptera: Tenebrionidae). *Journal of stored products research, 38*(5), 403-412.

Ismail, I. A., Qari, S. H., Shawer, R., Elshaer, M. M., Dessoky, E. S., Youssef, N. H., . . . Behiry, S. I. (2021). The Application of Pomegranate, Sugar Apple, and Eggplant Peel Extracts Suppresses Aspergillus flavus Growth and Aflatoxin B1 Biosynthesis Pathway. *Horticulturae*, 7(12), 558.

Isman, M. B. (2006). Botanical insecticides, deterrents, and repellents in modern agriculture and an increasingly regulated world. *Annu. Rev. Entomol.*, *51*, 45-66.

Kaundal, S. P., Sharma, A., & Kulshrestha, S. (2021). Analysis of phytochemical constituents, anti-microbial activity and molecular characterization of medicinal weed sonchus oleraceus l. using rapd marker. *IJPSR*, 12(12), 6453-6463.

Kemabonta, K., & Falodu, B. (2013). Bioefficacy of three plant products as post-harvest grain protectants against Sitophilus oryzae Linnaeus (Coleoptera: Curculionidae) on stored wheat (Triticum aestivum). *International Journal of Science and Nature*, 4(2), 259-264.

Khanal, D., Neupane, S. B., Bhattarai, A., Khatri-Chhetri, S., Nakarmi, N., Sapkota, S., . . . Sharma, V. (2021). Evaluation of botanical powders for the management of rice weevil (Sitophilus oryzae L. Coleoptera: Curculionidae) in Rupandehi, Nepal. *Advances in Agriculture, 2021*.

Kulkarni, N., & Joshi, K. (1998). Against maharukha webworm, atteva fabriciella (lepidoptera: yponomeutidae). *Journal of Tropical Forest Products*, 4(1), 11-16.

Kumar, M., Tomar, M., Amarowicz, R., Saurabh, V., Nair, M. S., Maheshwari, C., . . . Singh, S. (2021). Guava (Psidium guajava L.) leaves: nutritional composition, phytochemical profile, and health-promoting bioactivities. *Foods*, 10(4), 752.

Lee, S.-J., Umano, K., Shibamoto, T., & Lee, K.-G. (2005). Identification of volatile components in basil (Ocimum basilicum L.) and thyme leaves (Thymus vulgaris L.) and their antioxidant properties. *Food Chemistry*, 91(1), 131-137.

Li, W., Sun, H., Zhou, J., Zhang, Y., Liu, L., & Gao, Y. (2015). Antibacterial activities, antioxidant contents and antioxidant properties of three traditional Chinese medicinal extracts. *Bangladesh Journal of Pharmacology*, 10(1), 131-137.

Mahmoud, M., & Zedan, O. (2018). Essaying some Plant Powders against Maize Weevil, Sitophilus zeamais (Motsch.), for Protecting Maize Grains from Damage. *Journal of Plant Protection and Pathology*, 9(11), 729-734.

Mehta, V., & Kumar, S. (2020). Influence of different plant powders as grain protectants on Sitophilus oryzae (L.)(Coleoptera: Curculionidae) in stored wheat. *Journal of Food Protection*, 83(12), 2167-2172.

Nenaah, G. E., & Ibrahim, S. I. (2011). Chemical composition and the insecticidal activity of certain plants applied as powders and essential oils against two stored-products coleopteran beetles. *Journal of Pest Science*, 84(3), 393-402.

Petropoulos, S. A., Karkanis, A., Fernandes, Â., Barros, L., Ferreira, I. C., Ntatsi, G., . . . Khah, E. (2015). Chemical composition and yield of six genotypes of common purslane (Portulaca oleracea L.): An alternative source of omega-3 fatty acids. *Plant foods for human nutrition*, 70(4), 420-426.

Phillips, T. W., & Throne, J. E. (2010). Biorational approaches to managing stored-product insects. *Annual review of entomology*, *55*, 375-397.

Rajendran, S., & Sriranjini, V. (2008). Plant products as fumigants for stored-product insect control. *Journal of*

stored products research, 44(2), 126-135.

Rota, M. C., Herrera, A., Martínez, R. M., Sotomayor, J. A., & Jordán, M. J. (2008). Antimicrobial activity and chemical composition of Thymus vulgaris, Thymus zygis and Thymus hyemalis essential oils. *Food control*, 19(7), 681-687.

Saad, M. M., El Gendy, A. N. G., Elkhateeb, A. M., & Abdelgaleil, S. A. (2022). Insecticidal properties and grain protective efficacy of essential oils against stored product insects. *International Journal of Tropical Insect Science*, 1-10.

Schwarz, K., Ernst, H., & Ternes, W. (1996). Evaluation of antioxidative constituents from thyme. *Journal of the Science of Food and Agriculture*, *70*(2), 217-223.

Sharaby, A. (1988). Evaluation of Some Myrataceae Plant Leaves as Protectants Against the Infestation by Sitophilus oryzae L. and Sitophilus granarius L. *International Journal of Tropical Insect Science*, *9*(4), 465-468.

Shawer, R., Tonina, L., Tirello, P., Duso, C., & Mori, N. (2018). Laboratory and field trials to identify effective chemical control strategies for integrated management of Drosophila suzukii in European cherry orchards. *Crop protection*, *103*, 73-80.

Soujanya, P. L., Sekhar, J., & Kumar, P. (2016). Efficacy of plant extracts against rice weevil Sitophilus oryzae (L.) in stored maize. *Indian Journal of Entomology*, 78(4), 342-345.

Suleman, N., Aslam, M., & Riaz, A. (2000). Evaluation of resistance in some wheat cultivars to Sitophilus oryzae Linnaeus.(Coleoptera: Curculionidae) under laboratory conditions. *Pakistan Journal of biological sciences*, 3(6), 1029-1032.

Tapondjou, L., Adler, C., Bouda, H., & Fontem, D. (2002). Efficacy of powder and essential oil from Chenopodium ambrosioides leaves as post-harvest grain protectants against six-stored product beetles. *Journal of stored products research*, 38(4), 395-402.

Wahba, T. (2020). Antifeedant activity of three essential oils and their nanoemulsions against antifeedant activity of three essential oils and their nanoemulsions against the rice weevil Sitophilus oryzae (L.). *Egypt. Sci. J. Pestic*, 2, 19-31.

Xia, D.-Z., Yu, X.-F., Zhu, Z.-Y., & Zou, Z.-D. (2011). Antioxidant and antibacterial activity of six edible wild plants (Sonchus spp.) in China. *Natural product research*, 25(20), 1893-1901.

Ziaee, M., & Moharramipour, S. (2013). Effectiveness of medicinal plant powders on Sitophilus granarius and Tribolium confusum. *Journal of Crop Protection*, 2(1), 43-50.