

## EVALUATION OF THE MORTALITY EFFECT OF NATURAL ZEOLITE FORMULATION AND THE MODE OF ACTION IN THE PROTECTION OF DRY FRUITS FROM *EPHESTIA CAUTELLA* AND *STEGOBIUM PANICEUM* INFESTATION

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**ABSTRACT:** The almond moth, *Ephesia cautella* and the drugstore beetle, *Stegobium paniceum* are considered the most destructive pests of stored fruits. In the present study zeolite clinoptilolite (ultrafine < 20 µm) was applied at different rates to dried dates and apricots fruits to evaluate its insecticidal efficacy against different stages of both insects. Mortality was assessed after 3 and 7 days following exposure to zeolite powder. The mortality of *E. cautella* and *S. paniceum* exposed to treated dry date and apricot fruits with zeolite was significantly increased with increasing rates and exposure intervals. The adult progeny production and the number of laid eggs were significantly decreased by increasing the zeolite rate. No bad effects were observed in chemical analysis of date and apricot fruits. In the present study, a scanning electron microscope was used to observe the effect of zeolite powder on *E. cautella* larvae spiracles exposed to zeolite-treated dates. The spiracle openings were observed to be contracted to prevent zeolite powder entry. The results indicated that zeolite, which is considered nontoxic to the environment and safe for human consumption, can be effectively used as an integral part of programs to protect dried fruits from insect pests of stored products.

**Key words:** Store insects pest, safe agents, stored dry fruits, zeolite clinoptilolite.

### INTRODUCTION

Traditional dried fruits have been a staple of Mediterranean diets for millennia. Today, dried fruit consumption is widespread. Nearly half of the dried fruit sold throughout the world are raisins, followed by dates and apricots. Dates and apricots are some of the major dried fruits produced in the Mediterranean area (Ozer *et al.*, 2012).

The date palm (*Phoenix dactylifera* L.) is one of the oldest cultivated plants (Riad, 2006) and has been a staple food in the Middle East and North Africa for thousands of years. The largest date production in the world is concentrated in a few countries in this region, where Egypt occupied the first rank globally in the annual production of dates (FAO, 2020).

The apricot, *Prunus armeniaca* L. species is one of the most important Mediterranean fruits. This fruit is important in the diets of Asian and Mediterranean countries where it used as fresh

and dried fruit and is an important source of nutrient elements (Gómez *et al.*, 2021).

The almond moth or the tropical warehouse moth, *Ephesia cautella* (Walker) and the drugstore beetle, *Stegobium paniceum* (L.) are among the most common pests of stored products. They occur in both tropical and temperate regions and commonly attack grains, legumes, seed oils, dried fruits, and a wide variety of other stored products (Horak, 1994). However, certain species are specific pests on dried fruits and tree nuts as *E. cautella* and *S. paniceum* (Rajendran, 2003).

The search for alternatives to chemical insecticides and fumigants resulted in the proposal to employ inert dust to protect stored products (Allen, 2000, Fields and Korunic, 2002). Zeolites are crystalline hydrated aluminosilicates of alkali and alkaline earth metals (Christidis *et al.*, 2003; Sprynskyy *et al.*, 2005, Subramanyam and Roesli, 2000). Their usage in agriculture has risen after being deemed safe for humans by the FDA of the USA (FDA GRAS Listings, 2006) and non-toxic

by the WHO International Agency for Research on Cancer (IARC, 1997). Zeolite clinoptilolite is a mineral with special properties and its quality depends on its clinoptilolite content. While clinoptilolite is the most suitable type of natural zeolite for commercial and industrial applications, there have been few reports in the literature evaluating the effectiveness of natural zeolites for controlling storage insect pests. For the first time, Haryadi *et al.* (1994) reported the insecticidal potential of natural zeolites as insect development inhibitors for protecting stored products.

In addition, It was reported that natural zeolite from Indonesia completely controlled *Sitophilus zeamais* Motschulsky (Coleoptera: Curculionidae) during 3 months of storage (Haryadi *et al.*, 1994). In recent years, commercial formulations of zeolite have been developed and their insecticidal toxicity has been investigated. Subramanyam *et al.* (2015) evaluated the toxicity of synthetic zeolite (Odor-Z-Way, Phillipsburg, KS, USA) against seven stored-grain insect beetles. Rumbos *et al.* (2016) reported the insecticidal potential of three commercial zeolite formulations including Zeoprofeed Land 93 (ZeoProfit Hellas P.C., Thessaloniki, Greece), Zeofeed (ZeoProfit Hellas P.C., Thessaloniki, Greece) and a bulk (raw) zeolite (Volos, Greece) against adults of *Sitophilus oryzae*, *Tribolium confusum* and *Oryzaephilus surinamensis* in wheat. In other research, Lu *et al.* (2017) reported promising results on residual toxicity of synthetic zeolite (Odor-Z-Way, Phillipsburg, KS, USA) against *Callosobruchus maculatus* (F.) (Coleoptera: Bruchidae) when mixed with cowpeas seeds.

The current study aimed to investigate the efficacy of zeolite clinoptilolite powder to protect dates and apricots from *E. cautella* and *S. paniceum*. It incorporated scanning electron microscopy as a diagnostic technique to study the mode of action of tested powder. Additionally, a chemical analysis of its effect on dates and apricots was also conducted. A primary objective was to use safe and effective control methods that are easy to manipulate and utilize for local farmers rather than solely for large industrial users.

## MATERIALS AND METHODS

### 1. Test culture

This study was carried out using laboratory populations of the almond moth, *E. cautella* (Lepidoptera: Pyralidae) (eggs and larvae) and the drugstore beetle *S. paniceum* (Coleoptera: Anobiidae) (larvae and adults). The insects were obtained from existing cultures at the research laboratory of the Stored Products Department, Plant Protection Research Institute, Dokki, Giza, Egypt. They were reared on dried dates. The dates and apricot were sterilized before use by freezing (-10 °C) for 2 w to destroy any insect infestations. Cultures of these insects were kept in a darkened incubator maintained at 27°C ±2°C and 70% ±5% r.h. The colony was maintained at the same conditions for four generations before treatments.

### 2. Tested inert dust

The natural volcanic zeolite clinoptilolite mineral powder from Norway zeolite clinoptilolite (90%-92%, ultrafine <20 µm, very high cation exchange capacity and gray-green color) used in this study was purchased from Heiltropfen Lab, LLP, 27 Old Gloucester Street, WC1N3X London, UK. The inert dust was sieved through a 325-mesh sieve before use. Concentration response tests were conducted with immature stages (eggs, larvae), and larvae and adults of *E. cautella* and *S. paniceum*, respectively. Zeolite powder was applied to 1 kg of dates and apricots in 13 rates, 0, 0.18, 0.2, 0.4, 0.6, 1.0, 1.4, 1.6, 2, 4, 6, 8, 12, and 16g/kg of date and apricot fruits. The vials were shaken manually for 2 minutes to ensure uniform coverage of dates and apricots with zeolite powder before introducing any stages of both insects.

The eggs of *E. cautella* were carefully transferred to Petri dishes, then examined under a binocular microscope to eliminate injured or abnormal ones (malformed or pale white). The eggs were carefully transferred to vials (5 × 7.5 cm) at a density of 50 eggs/ vial. The number of hatched eggs was counted. The number of emerging adults from treated eggs was also recorded.

Fourth instar larvae of *E. cautella* and *S. paniceum* were separated from the medium with a camel brush. Twenty specimens were put into glass vials containing 1 kg of treated dried dates and apricots with rates of 0, 2, 4, 6, 8, 12, 16, and 0, 0.18, 0.2, 0.4, 0.6, 1.0, 1.4, and 1.6 g/kg of dates and apricots fruits, respectively. Mortality was evaluated on the 3<sup>rd</sup> and 7<sup>th</sup> d after exposure to the zeolite powder. The number of emerging adults from treated larvae was recorded.

For adults of *S. paniceum* the zeolite powder was tested at 0, 0.18, 0.2, 0.4, 0.6, 1.0, 1.4 and 1.6 g/kg dates and apricots. Twenty *S. paniceum* adults were separately introduced into each vial. Insect mortality was evaluated on the 3<sup>rd</sup> and 7<sup>th</sup> day after exposure to the zeolite powder.

All vials for all treatments were closed with muslin, secured tightly with rubber bands, and transferred to incubators at  $27^{\circ}\text{C} \pm 2^{\circ}\text{C}$  and 70%  $\pm 5\%$  r.h. Each rate was replicated three times. The same procedure was followed for the untreated dates and apricots that served as a control.

### 3. Chemical analysis of dry fruits

For both insect species' larvae, the mineral content of dried fruits post-exposure to zeolite powder applied at LC<sub>95</sub> on dried dates and apricots were estimated. Potassium and iron levels were determined using the flame photometric according to the methods of Toth and Prince (1949). Calcium and magnesium were determined using a PerkinElmer Atomic Absorption Spectrophotometer 2380 (PerkinElmer, Inc., Waltham, MA, USA). Total amino acids in the dates and apricots were determined using the method described by Lee and Takabashi (1966). Total carbohydrates were estimated in acid extracts of the samples using the phenol-sulphuric acid reaction of Dubois *et al.* (1956), followed by centrifugation and collection of the supernatant for analysis (Sadasivam and Manickam, 1991).

### 4. Scanning electron microscope study of spiracles of *E. cautella* larvae after zeolite powder treatment

The *E. cautella* larvae exposed to dried fruits treated with LC<sub>50</sub> of zeolite powders for 2 d were

examined using a scanning electron microscope (SEM). The zeolite powder-treated larvae after exposure to zeolite powder were allowed to air-dry for 5 d after exposure and without any other preparations for non-removal of zeolite from larvae, were placed onto adhesive stubs and coated with gold to improve the imaging of the samples. The classical dehydration method using graded alcohol series and critical point dryer was not used to preserve the zeolite particles on the larvae (Rumbos *et al.* 2016). The insects were next examined under a low vacuum (SEM) (Jeol-JSM-5600 LV in SEM) in the Electronic Microscope Unit, Central Laboratory, National Research Center, Cairo.

### 5. Data analysis

The data of *E. cautella* (eggs and larvae) and *S. paniceum* (larvae and adults) were transformed using arcsine before analysis. All data were subjected to one-way analysis of variance (ANOVA) using SPSS Statistics ver. 21.0 software (IBM Corp., Armonk, NY, USA). Duncan's Multiple Range Test (DMRT) was used to detect differences between mean values at the 0.05 significance level (Duncan 1955). To identify the lethal concentrations values (LC<sub>50</sub> and LC<sub>95</sub>); mortality percentages of both insect in different stages were used. The data were subjected to probit analysis as described by Finney (1971) and analyzed by computer program Ldp Line as described by Noack and Reichmuth (1978).

## RESULTS

### 1. Effect of zeolite powder rates on different stages of *E. cautella* and *S. paniceum*

The effect of inert dust zeolite on the number of hatched eggs and the number of adult survivorships from 2-day-old eggs of *E. cautella* after exposure to different rates of zeolite is presented in Table (1). The mean number of hatched eggs was directly related to the concentration rate, with hatched eggs in dates and apricot fruits and being significantly suppressed by zeolite powder ( $F= 23.3$ ;  $P < 0.0001$  and  $F= 49.12$ ;  $P < 0.0001$ ), respectively. All rates

generally caused a significant dose-dependent reduction in adult emergence from treated eggs relative to the control (F= 46.98; F= 121.00; P < 0.0001). Complete suppression of the hatchability and adult emergence was achieved after contact with date fruits infested by eggs of *E. cautella* at a rate of 20 g of zeolite powder /kg dry fruits. In case of apricot fruits, complete suppression of the hatchability and adult emergence was achieved at a rate 16 and 4 g zeolite powder /kg of dry fruits, respectively.

The mortality rate and the number of emerging adults from treated larvae of *E. cautella* post-exposure to different weights of zeolite powder are represented in Table (2). The mortality of *E. cautella* larvae was significantly affected (P < 0.0001) by the associated interactions of the exposure interval and the weight of the zeolite powder. The mortality of *E. cautella* larvae on zeolite-treated dates and apricot fruits increased with increasing weights and exposure time. Complete mortality was achieved at 8 g/kg after 7 days of exposure. On untreated dates and apricot (control), the number of emerging (mean) adults of *E. cautella* was 53.33 and 86.67 adult, respectively (Table 2). The number of emerging adults of *E. cautella* from treated larvae decreased with increasing zeolite powder rates. Differences

in the number of emerging adults of *E. cautella* from treated larvae were significant among zeolite rates in date fruits (F =81.44; P < 0.0001) and in apricot fruits (F =96.57; P < 0.0001). Adult progeny production occurred only at the rate of 2 and 4 g/kg of date fruits post-exposure to zeolite powder. Furthermore, strong, and complete suppression of *E. cautella* progeny was observed at 6, 8, 12 and 16 g/kg of zeolite powder. Nevertheless, in apricot fruits complete suppression of *E. cautella* progeny was observed in all rates.

The effect of zeolite powder on mortality rates of *S. paniceum* adults' post-exposure to different application rates is presented in Table (3). Mortality levels of *S. paniceum* adults were significantly affected by the exposure interval and zeolite rates. The mortality of *S. paniceum* adults on zeolite-treated dates increased with increasing rates and exposure time. Mortality of *S. paniceum* adults in treated dates was 23.33% - 96.67% and in treated apricots was 80%- 90% after 3 days of exposure to fruit treated with different rates of zeolite. Complete mortality was achieved at weights of 1.4, 1.0 and 0.6 g/kg after 7 days of exposure in the case of dates and after 3 days in apricots, respectively.

**Table 1. Number of hatched eggs and adult survivorships from 2 days old eggs (Mean± SE) of *Ephestia cautella* after exposure of dry date and apricot fruits to different rates of zeolite.**

Conc g/ kg	Dry dates				Dry apricot			
	% of hatched eggs mean± SE	Reduction % of egg hatch	No. emerging adults from treated eggs mean± SE	Reduction %emerging adults	%. of hatched eggs mean± SE	Reduction % egg hatch	No. emerging adults from treated eggs mean± SE	Reduction % emerging adults
0	32.00±0.58	-	30.67±1.5	-	67.33± 4.2	-	51.33±2.33	-
4	29.33±1.50	8.33	20.67±2.4*	32.62	16.0±1.15*	76.24	00±0.00*	100
8	20.00±2.3*	37.5	6.67±2.00*	78.28	8.67± 0.88*	87.14	0±0. 00*	100
12	14.00±1.5*	56.25	4.67±0.33*	84.8	4.67±0.88*	93.08	0±0.00*	100
16	5.33±0.67*	83.31	0.67±0.88*	97.85	0± 0.00*	100	0±0. 00*	100
20	0.00±0.0*	100	0±0.00*	100	0±0.00*	100	0. ±0.00*	100
F	23.3	-	46.98	-	49.12		121.00	
P	0.000	-	0.000	-	0.000		0.000	

\* The mean difference is significant at the 0.05 level compare with control.

**Table 2. Mortality and number of emerging adults from treated larvae of *Ephestia cautella* (Mean± SE) with different rates of zeolite powder on dry date and apricot fruits**

Conc. g/ kg	Dry dates				Dry apricot			
	Mortality of treated larvae Mean ± SE after indicated period (days)		No. emerging adults from treated larvae Mean ± SE	Reduction % of emerging adults	Mortality of treated larvae Mean ± SE after indicated period (days)		No. emerging adults from treated larvae Mean ± SE	Reduction % of emerging adults
	3	7			3	7		
0	0.0±0.0	0.0±0.0	53.33±0.33*	-	0.0±0.0	0.0±0.0	86.67 ±0.88*	
2	16.67±0.33	30.0±0.58	6.67±0.33*	87.43	20.00 ±0.58	56.67 ±0.33	0.00± 0.00*	100
4	20.0±0.58	33.33±0.33	3.33±0.33*	93.81	23.33 ±0.33	70.00 ±0.58	0.00 ± 0.00*	100
6	23.33±0.33	53.33±0.33	0.00±0.00*	100	40.00 ± 0.58	80.00 ±0.58	0.0±0.00*	100
8	53.33±0.33	100±0.00	0.00±0.00*	100	56.67 ± 0.58	100±0.00	0.0±0.00*	100
12	76.67±0.33	100±0.00	0.00±0.00*	100	76.67 ± 0.33	100±0.00	0.0±0.00*	100
16	80.0±0.58	100±0.00	0.00±0.00*	100	90.00± 0.58	100±0.00	0.0±0.00*	100
F	45.3	127.88	81.44	-	44.089	24.206	96.57	
P	0.000	0.000	0.000	-	0.000	0.000	0.000	

\* The mean difference is significant at the 0.05 level compare with control.

**Table 3. Effect of zeolite powder on mortality percent of *Stegobium paniceum* adults post exposure to different application rates of zeolite powder on dry date and apricot fruits**

Conc. g/ kg	Dry date fruits		Dry apricot fruits	
	Mortality% Mean± SE after indicated period (days)		Mortality% Mean± SE after indicated period (days)	
	3	7	3	7
0	0.00±0.00	00.00±0.00	0.00±0.00	00.00±0.00
0.18	23.33±0.33	63.33±0.33	80.00±0.00	100.0±0.00
0.2	43.33±0.33	80.00±0.58	83.33±0.88	100.0±0.00
0.4	50.00±0.58	86.67±0.33	90.00±0.58	100.0±0.00
0.6	63.33±0.88	100.0±0.00	100.0±0. 00	-
1	76.67±0.33	100.0±0.00	100.0±0. 00	-
1.4	96.67±0.33	100±0.00	100±0. 00	-
1.6	100±0.00	-	100±0. 00	-
F	35.976	25.667	4.9	-
P	0.000	0.000	0.007	-

The effect of zeolite powder on mortality rates and emerging adults of treated *S. paniceum* larvae post-exposure to different application rates is shown in Table (4). There was a significant treatment effect on overall *S. paniceum* larvae mortality over time ( $P < 0.0001$ ). The mortality of *S. paniceum* larvae on the zeolite-treated dates and apricots increased with increasing rates and exposure times. The mortality of *S. paniceum* larvae was 20.0% - 93.33% in the case of dates, and 43.33%- 100% in apricots after 3 d, respectively, post-exposure to treatment with different rates of zeolite powder. Complete mortality was achieved at weights of 1.4 and 1.6 g/kg after 7 days of exposure to zeolite in dates and in apricots in all rates. In contrast with mortality, progeny counts were significantly decreased ( $F=102.79$ ;  $P < 0.0001$ ) in date fruits. The parental (mean) survival rates of *S. paniceum* after exposure to 0.18 and 0.2 g/kg treated dates were 53.33 and 16.67, respectively, while it was 96.67 in the untreated control. Nevertheless, in apricot fruits complete suppression of *S.*

*paniceum* progeny was observed in all tested rates of zeolite powder.

The expected exposure rates of zeolite powder to cause 50 and 95% mortality after contact with the dried dates and apricots infested by *E. cautella* (eggs and larvae) and *S. paniceum* (adults and larvae) 3 days post-exposure are presented in Table (5). The sensitivity to zeolite powder treatments differs between stages of *E. cautella* and *S. paniceum* and the type of fruit used based on  $LC_{50}$  values. Overall, *E. cautella* was more tolerant species than *S. paniceum*, while *E. cautella* larvae was more susceptible stage to zeolite powder compared to *S. paniceum*. Probit analysis of zeolite powder concentration-response assessments indicated that *E. cautella* larvae needed 3 days of treatment to achieve 50% mortality at 7.1 g/kg after contact with the dried dates and apricots, respectively. In contrast, *S. paniceum* adults were more sensitive stage, needing 0.34 and 0.07 g/kg after contact with the dried dates and apricots fruits.

**Table 4. Effect of zeolite powder on mortality % and emerging adults of treated *Stegobium paniceum* larvae post exposure to different rates of zeolite powder on dry date and apricot fruits.**

Conc. g/ kg	Dry date fruits			Dry apricot fruits		
	Mortality % of treated larvae Mean $\pm$ SE after indicated period (days)		No. emerging adults from treated larvae Mean $\pm$ SE	Mortality of treated larvae Mean $\pm$ SE after indicated period (days)		No. emerging adults from treated larvae Mean $\pm$ SE
	3	7		3	7	
0	00.00 $\pm$ 0.0	0.0 $\pm$ 0.00	96.67 $\pm$ 0.33	00.00 $\pm$ 0.0	0.0 $\pm$ 0.00	90 $\pm$ 0.00
0.18	20.00 $\pm$ 0.00	40.0 $\pm$ 0.58	53.33 $\pm$ 0.33	43.33 $\pm$ 1.2	100 $\pm$ 0.00	0 $\pm$ 0.00
0.2	43.33 $\pm$ 0.33	66.67 $\pm$ 0.33	16.67 $\pm$ 0.88	56.67 $\pm$ 0.67	100 $\pm$ 0.00	0 $\pm$ 0.00
0.4	46.67 $\pm$ 0.66	70.00 $\pm$ 0.33	0.00 $\pm$ 0.00	76.67 $\pm$ 0.88	100 $\pm$ 0.00	0 $\pm$ 0.00
0.6	50.00 $\pm$ 0.00	73.33 $\pm$ 0.67	0.00 $\pm$ 0.00	93.33 $\pm$ 0.00	100 $\pm$ 0.00	0 $\pm$ 0.00
1	60.00 $\pm$ 1.00	86.66 $\pm$ 1.33	0.00 $\pm$ 0.00	100 $\pm$ 0.00	100 $\pm$ 0.00	0 $\pm$ 0.00
1.4	83.30 $\pm$ 0.88	100 $\pm$ 0.00	0.00 $\pm$ 0.00	100 $\pm$ 0.00	100 $\pm$ 0.00	0 $\pm$ 0.00
1.6	93.33 $\pm$ 0.33	100 $\pm$ 0.00	0.00 $\pm$ 0.00	100 $\pm$ 0.00	100 $\pm$ 0.00	0 $\pm$ 0.00
F	17.818	11.52	102.794	33.3	-	-
P	0.000	0.000	0.000	0.000	-	-

**Table 5. The expected exposure rates of zeolite powder (g/kg) caused 50 and 95% mortality for *S. paniceum* (adults and larvae) and *E. cautella* (larvae and egg) on dry date and apricot fruits after 3 days of exposure**

Developmental stages	Dry date fruits			Dry apricot fruits		
	LC <sub>50</sub> g /k (Lower-upper)	LC <sub>95</sub> g /k (Lower-upper)	Slope	LC <sub>50</sub> g /k (Lower-upper)	LC <sub>95</sub> g /k (Lower-upper)	Slope
Adults <i>S. paniceum</i>	0.34 (0.2-0.5)	1.8 (1.5-4.1)	2.3±0.2	0.07 (0.05-0.1)	0.4 (0.4-0.5)	2.1±0.3
Larvae <i>S. paniceum</i>	0.43 (0.2-0.7)	4.3 (4.9-29.8)	1.7±0.15	0.19 (0.1-0.2)	0.97 (0.6-2.8)	2.3±0.50
Larvae <i>E. cautella</i>	7.1 (4.8-9.6)	26.5 (23.59-53.97)	2.9±0.2	7.1 (6.6-7.7)	24.29 (18.64-37.32)	2.2±0.2
Eggs <i>E. cautella</i>	2.3 (6.4-12.2)	22.4 (20.8-47.3)	1.8±0.3	1.5 (0.8-2.4)	8.8 (7.3-10.9)	2.2±0.34

## 2. Effect of zeolite powder on components of dried fruits

The determination of the average values of the minerals (macro-elements) and certain component (total carbohydrates and free amino acids) content in dried dates and apricots post-treatment with 26.5 g/kg (LC<sub>95</sub> of *E. cautella*) of zeolite powder after 3 months are shown in Table (6). The application of zeolite powder on dried fruit resulted in a non-significant increase in total carbohydrates (658.0 and 516.33 mg/g) after 3 months of treatment compared with 549.67 and 424 mg/g in the control for dried dates and apricots, respectively. In contrast, a significant reduction was observed in free amino acids in dried dates with 665.0 µg/g compared with 768.33 µg/g in the control. This indicated that the used rate of zeolite powder decreased free amino acids percentage by only 13.45%. However, in dried apricots non-significant increase in free amino acids with 191.67 µg/g compared with 151.33 µg/g in the control. These results indicated that there was no significant effect of the application of zeolite powder at LC<sub>95</sub> level on the mineral

content of dried dates and apricots after 3 months of storage. The application of zeolite powder at the LC<sub>95</sub> level significantly increased the potassium level, which is considered the predominant element present in dates, 0.54%, compared with 0.45% in the control.

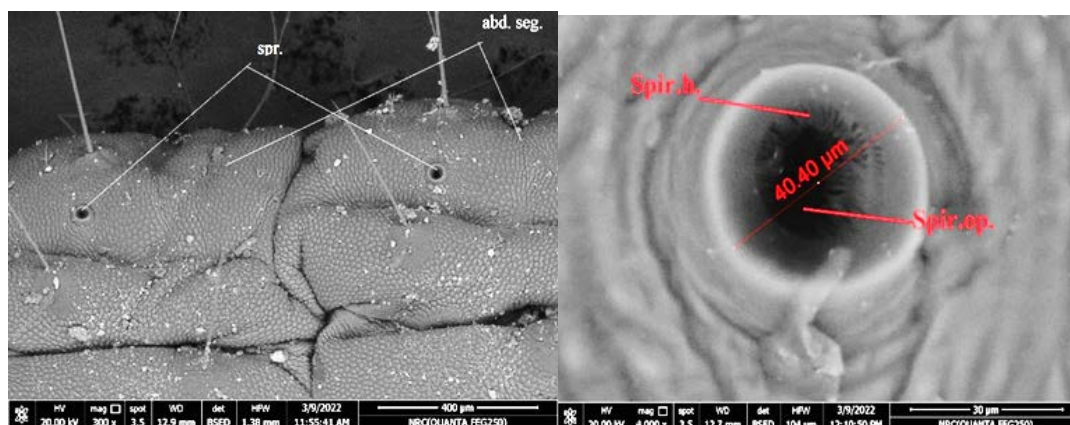
## 3. Effect of zeolite powder on the spiracles of *E. cautella* larvae

Examining the *E. cautella* larvae body, the spiracles appear on the side of the abdomen (The SEM shows very detailed 3-dimensional images at much higher magnifications than is possible with a light microscope). The open spiracles (breathing openings) are illustrated in Fig. (1 A and B). SEM showed the effect of zeolite powder on the spiracle openings which were closed Fig. (2 A) following treatment with zeolite powder. Zeolite powder particles appeared on the surface of the insect body and powder particles are seen closing the spiracle openings of *E. cautella* larvae in Fig. (2 B).

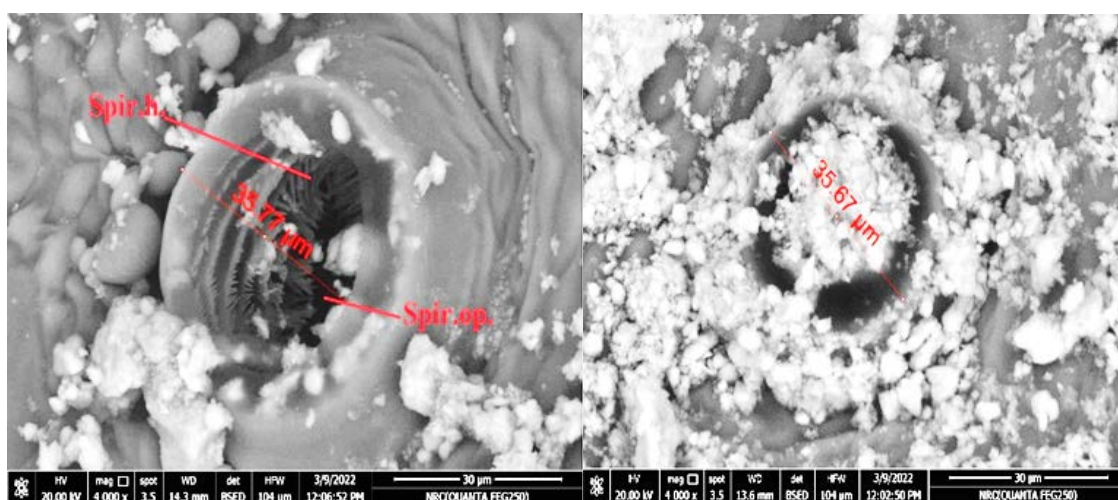
**Table 6. Chemical analysis the minerals content of dry date fruits**

Components	Dry date fruits		Dry apricot fruits	
	Treated± SE	Untreated± SE	Treated± SE	Untreated± SE
Total carbohydrates mg/g	568.0±10.4 <sup>a</sup>	549.67± 6.1 <sup>a</sup>	516.33±13.9a	424±18.15a
Free amino acids µg/g	665.0±12.1 <sup>a</sup>	768.33±26.2 <sup>b</sup>	191.67±4.4a	151.33±7.54a
Potassium K%	0.54±0.08 <sup>a</sup>	0.44±0.07 <sup>a</sup>	0.76±0.004a	0.68±0.05a
Calcium Ca%	1.25±0.1 <sup>a</sup>	1.5±0.06 <sup>a</sup>	0.98±0.00a	0.77±0.03a
Iron Fe mg/kg	289±2.0 <sup>b</sup>	312±1.5 <sup>a</sup>	64.33±0.35a	67.94±1.3a
Magnesium Mg%	0.15±0.01 <sup>a</sup>	0.15±0.01 <sup>a</sup>	0.29±0.01a	0.28±0.02a

Means followed by the same letter are not significantly different using Tukey's HSD test, (P= 0.05).



**Fig. 1A, B normal spiracle of *E. cautella* larvae**



**Fig. 2 A, B treated spiracle of *E. cautella* larvae**



#### 4. Discussion

Inert dust, such as zeolite, are promising alternatives to conventional chemical insecticides and have been extensively researched and used in practice due to their low mammalian toxicity and slight or no effects on product quality when treated with commercially applicable concentrations (Korunic, 1998; Andric *et al.*, 2012). Zeolites are crystalline, hydrated aluminosilicates of alkali and alkaline earth (Christidis *et al.*, 2003). As mentioned, there have been few reports concerning the effectiveness of natural zeolites for controlling storage pests. To our knowledge, this is the first published report on the insecticidal efficacy of zeolites against *E. cautella* and *S. paniceum*. The data of the present study showed that natural zeolite powder effectively protected dried fruits against *E. cautella* (egg and larvae) and *S. paniceum* (larvae and adults). Mortality rates were confirmed to increase gradually with increasing concentrations of zeolite powder and with increasing exposure periods. Complete mortality (100%) of *E. cautella* (larvae) and *S. paniceum* (larvae and adults) was observed on the 7<sup>th</sup> day of exposure to 8, 1.4, and 0.6 g/kg of zeolite, respectively. In another study, the mortality of adult *Sitophilus oryzae*, *Rhyzopertha dominica*, and *Tribolium castaneum* was 100, 96, and 82%, 21 d after exposure to 1.0 g/kg of natural zeolite products, respectively (Kljajic *et al.*, 2010b). Recently, Bohinc *et al.* (2018) evaluated the insecticidal activity of natural zeolites against the maize weevil, *S. zeamais*, and complete mortality (100%) of adults was observed after 21 days of exposure to 900 ppm. Also, Ali *et al.* (2021) investigated the insecticidal activity of zeolite (ZeoFeed) against *Trogoderma granarium* larvae on wheat, rice and maize and founded maximum mortality in *Tribolium granarium* larvae was observed on wheat at dose rate of 750 ppm at 35°C and 55% R.H. after 14 days of treatment which was 45.90%.

It is often more important in practical cereal storage conditions to prevent progeny formation than to concentrate on obtaining direct lethal effects from zeolite against parent insects. The present results showed that the natural zeolite

suppressed progeny production of *E. cautella* (larvae) and *S. paniceum* (larvae and adults) and inhibited the egg-to-adult emergence of *E. cautella* eggs exposed to zeolite-treated fruit. The present experiments with both insect species achieved high levels of progeny inhibition (97.85–100%) after intervals of parental exposure to zeolite-treated dry fruits. This difference in exposure required to achieve the same hatchability and adult emergence rates may be attributed to a difference between the two types of dried fruits (Riudavets *et al.*, 2009). Subramanyam and Roesli (2000) reported that the prevention of massive progeny production in substrate treated with inert dust, as an indirect action, was of greater importance than direct action against parents. Previous studies have recorded 94%–100% mortality and >80% progeny suppression of *S. zeamais*, *S. oryzae*, and *T. castaneum* exposed to natural zeolites at varying doses (Haryadi *et al.*, 1994, Kljajic *et al.*, 2010a, Andric *et al.*, 2012). However, natural zeolite modified by treatment with NH<sub>4</sub><sup>+</sup> ions, applied at 1g/kg showed much lower insecticidal potential, with 36%–56% mortality and 62%–71% progeny reduction in *S. oryzae* and *T. castaneum* (Andric *et al.*, 2012).

Based on a literature review, the application of zeolites may reduce infestations of stored products by insects (Ziaee *et al.*, 2021). Generally, the effectiveness of inert dust may vary depending on grain type (Kavallieratos *et al.*, 2005), insect species, their developmental stage (Korunic, 1997; Korunic, 1998; Fields and Korunic, 2000), silicon dioxide content and structure (amorphous or crystalline), particle size distribution, geographic origin of the dust (Vayias *et al.*, 2009), concentration, exposure time, and grain temperature and moisture content (Arthur, 2000, 2002; Athanassiou *et al.*, 2005). However, the insecticidal potency of zeolites is influenced by several factors. For instance, insect species indicate different levels of susceptibility to these aluminum silicates. According to the calculated LC<sub>50</sub> values, *E. cautella* larvae and eggs were stages which were the most tolerant to zeolite powder compared to *S. paniceum* adults and larvae post-exposure to different application rates

of zeolite powder on dried dates and apricots at 3 days post-exposure. This result agrees with Sabbour *et al.* (2012) who reported that *E. cautella* and *E. kuehniella* larvae were the most tolerant species to test DEs at 5%. These *E. cautella* were reared on dates, and this may have increased their tolerance. It was difficult to know how the infestation was distributed relative to the portion of dates being treated, and this could have influenced insect responses. The location of the insect within the dates may also influence its susceptibility to dust. This suggestion is also supported by the finding of Rumbos *et al.*, 2016 who reported that *O. surinamensis* was the most susceptible species to zeolite application, whereas *T. confusum* was the most tolerant. In the present study, high zeolite dosages are required to obtain satisfactory control of the *E. cautella* larvae that live within dates. Long periods of exposure and high zeolite dosages are required to obtain satisfactory control of the insect pests (Kljajic *et al.*, 2010a, 2010b). Andric *et al.* (2012) reported that natural zeolite at a concentration of 1.0 g/kg resulted in 96 and 100% mortality of *S. oryzae* and *T. castaneum* 21 days after exposure. Another natural zeolite from Slovenia at 900 ppm showed promising insecticidal potential against adult *S. zeamais* (Trdan *et al.*, 2015). This result concurs with those reported by Popoola and Adenuga (2013) who found that the application of palm ash at 1.25 g/25 g within 5 weeks of storage control *O. surinamensis* infestations on palm dates in storehouses. These data are similar to those reported by Subramanyam *et al.* (2015) who observed that an application of zeolite at 1 g/kg achieved protection in cowpea seeds from *C. maculatus* with a significant decrease in progeny production on zeolite-treated cowpea seeds compared with untreated seeds, moreover, zeolite at 5 g/m<sup>2</sup> showed high effectiveness with 100% mortality of *C. maculatus* adults after 36 h of exposure to treated surfaces (Subramanyam *et al.*, 2015).

Since sugars are the predominant constituent of dates, there is an association between calorific value and sugar content. This makes dates a preferable main foodstuff for the general population, especially during the holy fasting month (Ramadan) and for those involved in

strenuous work. Overall, this study showed that there was increased in total carbohydrates and potassium which are considered the predominant elements present in dates and apricots after 3 months of treatment with zeolite powder. In another study, packaging material was combined with zeolite, diatomite, and calcium carbonate to improve the storage quality of peaches (Kim *et al.*, 2022). Zeolite combines aluminum oxide and silicic acid oxide (Kim, 2016) and it is known to exhibit a high cation exchange capacity and high selective adsorption capacity for gases (Park and Ha, 1994). Though numerous studies have been conducted on the adsorption of zeolite (Lee *et al.*, 2011; Otker *et al.*, 2005). Zeolite adsorption of ethylene, which is continuously generated by peaches during storage, enables respiration and limits ripening and thus delays softening (Kim *et al.*, 2022). In fact, zeolite is known to remove ammonia nitrogen rapidly and stably (Bernal and Lopez-Real, 1993). Furthermore, zeolite, in which some portion of the cations is substituted with anions, has been used as a food quality maintenance or freshness retention agent because of its antibacterial properties. Thus, food freshness and preservation can be improved by capitalizing on the excellent moisture adsorption and gas-binding properties of proposed zeolite-based functional packaging (Kim *et al.*, 2022). Additionally, zeolites did not alter the evolution of reducing sugars during tomato storage in a previous study which showed that the main free sugars of commercial tomato varieties are reducing sugars with a negligible amount of sucrose (Raiola *et al.*, 2018). Increased fruit shelf life was the result of maintaining quality until consumption observed by Kim *et al.* (2022) which is similar to the observations in the present study.

Most insects breathe through the trachea which usually leads to the opening of the spiracles (Kemabonta and Falodu, 2013). Spiracles are important structures at the boundary between atmospheric air and the gas-filled tracheal system. Therefore, a significant problem that spiracles must address is the conflict between gas exchange and transpiratory water loss. These spiracles might have been blocked by zeolite powders thus impairing respiration and leading to suffocation and subsequent death (Owoade, 2008, Abdullahi

*et al.*, 2012). Gerolt (1965 and 1969) found that the spiracle was the most important penetration site for applied dieldrin. Applying powder was observed to result in some morphological changes to spiracle shape (Fawki *et al.*, 2014). Spiracles of *E. cautella* larvae have an internal closing mechanism and are annular (simple), uniforous (one-opening), and atriate (spiracle chamber) (Fig. 1). The atrium's inside wall is covered in countless tiny cuticular hairs (Fig. 1). Concerning the response of *E. cautella* larvae to zeolite powder, it seems that the spiracle openings were contracted to prevent the entry of zeolite powder. This appeared as decreasing atrium size (depth) but normal appearance of the inner valve parts (Fig. 2). The insect cuticle shows some amount of shrinkage due to the contraction of the spiracle's closing muscles to prevent zeolite powder particles from entering and prevent respiration via the trachea (Shukla *et al.*, 2007).

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تقييم التأثير المميت لتركيبية الزيوليت الطبيعية وطريقة عملها لحماية الثمار الجافة من  
الإصابة بدودة البلح العامري *Ephestia cautella* وخنفساء العقاقير  
*Stegobium paniceum*

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المخلص العربي

تعتبر دودة البلح العامري وخنفساء العقاقير من أكثر الآفات تدميرا للفواكة المجففة المخزنة. في هذه الدراسة تم استخدام الزيوليت كلينوبتيلوليت (متناهي الصغر >20 ميكروملايتر) بمعدل جرعات مختلفة على ثمار التمور و المشمش المجففة والمخزنة لتقييم فاعليتها الابادية ضد الاطوار المختلفة من الحشرات تحت الدراسة . تم تقييم نسبة الموت بعد 3 و7 أيام من المعاملة بمسحوق الزيوليت. زادت نسبة الموت لكلا الحشرتين زيادة معنوية مع زيادة الجرعة او فترة المعاملة. انخفض معدل خروج الحشرات الكاملة وعدد البيض الموضوع بشكل معنوي بزيادة جرعة الزيوليت. لم يلاحظ أي تأثير سلبي في التحليل الكيميائي لثمار البلح والمشمش. في هذه الدراسة تم استخدام الميكروسكوب الإلكتروني الماسح لملاحظة تأثير مسحوق الزيوليت على فتحات الثغور في يرقات دودة البلح العامري. أشارت النتائج إلى أن الزيوليت، الذي يعتبر غير سام للبيئة وآمن للاستهلاك البشري، يمكن استخدامه بشكل فعال كجزء لا يتجزأ من برامج حماية الفواكه المجففة من الآفات الحشرية للمنتجات المخزنة.