



Biological studies and toxicity experiments of AEROSIL 200 nanoparticles on adults and larvae of some stored grain insects

Nilly AH Abdelfattah¹, Doaa M Zein²

^{1,2} Department of Stored Products and Grains, Plant Protection Res. Institute, Agric. Res. Center., Dokki, Giza, Egypt

Abstract

Current study was conducted to investigate the toxicity of fumed silica, Aerosil 200 nano particles (99.8% SiO_2) against adults and larvae of three stored grain insects. Khapra beetle, *Trogoderma granarium*, drugstore beetle, *Stegobium paniceum* and red flour beetle, *Tribolium castaneum*, residual toxicity, seeds germination % and seeds components % for 6 months storage. Results showed that @ highest conc. 1.5 and 2 gm/kg fumed silica was sufficient to cause 100% mortality with *T. granarium* and *S. paniceum* after 5 and 2 days of treatment while for *T. castaneum* which known by its highly resistance for most alternatives, 4 gm/kg recorded 83% mortality after 21 days from treatments. Fumed silica @ lower limit LC_{90} can protect wheat seeds from insect infestation until the 4th month storage. The crude protein %, total carbohydrate%, crude fiber% and total ash% were increased while crude fat % and moisture % decreased in treated grains when compared with untreated grains. Germination % of seeds stored for 6 months after treatment with lower limit LC_{90} of fumed silica slightly affected comparing with untreated seed. Also chlorophyll analysis showed that fumed silica slightly decreased chlorophyll % in treated seeds. Nano particles by transmission Electron Microscope (TEM) indicates that the original morphology of particles approximately spherical with the diameter varying between 5 to 20 nm.

Keywords: stored grain insects- aerosil 200- germination- toxicity

Introduction

In recent years, consumer awareness of the health hazard from Fumigants and residual insecticides toxicity which commonly used to combat stored grain pests and the growing problem of insect resistance to these conventional insecticides have led the researchers to look for alternative strategies for stored grains protection ^[1]. World-wide scientists are providing clean and safe food to all human beings and live-hoods by adapting many green technologies such as nanotechnology. Nanotechnology is a promising field of interdisciplinary research. It opens up a wide array of opportunities in various fields like insecticides, pharmaceuticals and agriculture. Considering overall merits of nanomaterials, investigators recommend the application in various spheres of sustainable agriculture because nanotechnology will be helpful to meet the food security challenges; targeted delivery of pesticides, promote the seed germination and plant growth, increase crop yield, improve food quality, control of pestiferous insects that destroy crops and their products in the field as well as in storage ^[2]. According to relevant specialized encyclopedias, the definition of NPs is 'Nanoparticles are solid colloidal particles ranging in size from 1 to 100 nm ^[3, 4]'. More recently, materials including diatomaceous earth, silica aerogels and silica nano particles have been increasingly finding use in commercial storage in the developed world replacing conventional chemicals ^[5]. AEROSIL 200 nano particles (size is 5–50 nm) which is synthetic amorphous silica composed of (99.8% SiO_2) also known as pyrogenic silica is available in hydrophilic and hydrophobic (coated) forms. It is used as a matting agent, thickener, and filler in many adhesives and coatings. Fumed silica has also been used as a desiccating agent to kill insects ^[6]. According to the International Agency for the Research of Cancer (IARC), amorphous silica belongs

to group 3; it is classified as not carcinogenic. United States Department of Agriculture (USDA) has already approved the use of amorphous silica as safe ^[7].

The aim of this study was to investigate the entomotoxicity of fumed silica, Aerosil 200 nanoparticles against adults and larvae of three stored grain insects which are serious pests of stored grains and its products. These insects are, Khapra beetle, *Trogoderma granarium*, drugstore beetle, *Stegobium paniceum* and red flour beetle, *Tribolium castaneum* at $30 \pm 2^\circ\text{C}$ and $65 \pm 5\%$ R.H.

Khapra beetle, *Trogoderma granarium* (Everts) is one of the world's most destructive stored-product pests. In fact, it has been recognized as an A2 quarantine organism for EPPO ^[8] and ranked as one of the 100 worst invasive species worldwide ^[9]. Losses caused by *T. granarium* (Everts) have been reported to range from 0.2 to 2.9% over a period of 1 to 10.5 months ^[10].

Tribolium castaneum is a polyphagous, cosmopolitan pest, feeding mostly on stored flour and other milled cereal products, broken wheat and farm stored products. In severe infestation the flour turns grayish and mouldy and has pungent, disagreeable odor making it un-fit for human consumption ^[11].

Drugstore beetle, *Stegobium paniceum* (L.) is the most widely encountered insect causing serious damages and huge economic losses to stored products ^[12].

2. Materials and methods

2.1 Insects

Adults and larvae of Khapra beetle, *Trogoderma granarium*, drugstore beetle, *Stegobium paniceum* and red flour beetle, *Tribolium castaneum* were reared in the laboratory of Department of Stored Products and Grains Pests, Plant Protection Research Institute, Agriculture Research Centre,

Dokki, Giza, Egypt. The insects reared for several generations on wheat and wheat flour media. The wheat grains were sterilized at a temperature of 55°C for 6 hrs. In order to eliminate any hidden infestation before using.

2.2 Biological studies

Ten replicates were used for each treatment. For observing the incubation period of eggs and the developmental periods for the successive stages, single one (0-1 day old) egg was placed in a glass tube (10 x 3 cm) covered with muslin secured by rubber band. The incubation period for every egg was recorded. Duration of immature stages was recorded. Each replicate contained one of newly hatched larvae putted with 1 gm of wheat for *T. granarium* while flour for *S. paniceum* and *T. castaneum* (0-1 day old) and pupae (0-1 day old). each putted individually in test tubes (10 x 3 cm) covered with muslin secured by rubber band.

For adult longevity, five replicates were used per insect. A pair of freshly emerged unmated males and females (0-1 day old) was placed in a Petri dish (6 x 1 cm) surrounded with rearing medium for *T. granarium* and *S. paniceum*. In case of *T. castaneum*, the pair was in a glass test tube (10 x 3 cm) contained 1 gm of flour and covered with muslin secured by rubber band. These pairs were examined daily until the death of adults. The experiment conditions were at 30±2 °C and 65±5 % R.H.

2.3 Tested material

Aerosil 200 nanoparticles (fumed silica) obtained from Taiba Company for scientific services, Egypt.

2.4 Bioassay test

Adults of *T. granarium* & *S. paniceum* (0-24 hrs old), *T. castaneum* adults (1-2 weeks old) and larvae of all (10-15 days old) were used for the experiments. Effect of Aerosil 200 nano particles were determined by contact toxicity assay at different concentration of two stages. The experiments were carried out in completely randomized design with 3 replications each consisted of 20 insect adults or larvae in small plastic screw capped jars containing 10 g of wheat grains for *T. granarium* and crashed wheat for *S. paniceum* & *T. castaneum*. Wheat in each jar was treated individually with aerosil 200. Then, the jars were shaken manually for approximately 1 min to achieve equal distribution. In one additional set no Aerosil 200 was mixed with wheat and this set served as untreated for comparison. All bioassays were performed at 30±2°C and 65±5% RH. Adult mortality was recorded after 2, 3, 5 and 7 days for adult of *T. granarium*, 1, 2, 3, 4 and 5 days for adult of *S. paniceum* and 2, 4, 6, 8, 10, 14 and 21 days for adult of *T. castaneum*. Reduction percentage in *F*₁ progeny was calculated according to equation [13] after 45-60 days. Larvae mortality for *T. granarium* was recorded after 2, 4, 6, 8 and 11 days. *T. castaneum* larva mortality checked after 2, 4, 6, 8, 10, 14 and 21 days. While larva of *S. paniceum* were incubated until adult emergence. The aqueous suspensions of the studied nano particles have been previously characterized by the Transmission Electron Microscope (TEM) [14].

2.5 Residual toxicity experiment

A laboratory experiment was conducted to study the residual efficiency of Aerosil 200 nanoparticles against *T. granarium* larvae, lower limit LC₉₀ conc. was mixed with 250 gm of wheat and stored for 6 months under laboratory conditions.

250 gm of untreated seeds were used for control. Data recorded every month in which 3 replicates were used for treated and untreated seeds, each replicate contain 10 gm of seeds and infested with 10 pairs of larvae at 30±2°C and 65±5% R.H. The mortality (%) of larvae after 7 days of treatment was recorded. The used concentration was 6 gm/kg. Used conc. in residual toxicity experiment is the lower limit of LC₉₀ = 6 gm/kg for the 10th day of treatment.

2.6 Seeds components analysis

This experiment was carried out at Soil, Water and Environment Research Institute to study the effect of lower limit LC₉₀ conc. of Aerosil 200 nanoparticles on wheat seeds components (crude protein%, crude fats%, carbohydrates %, moisture %, and ash %) after 6 months from storage, control grains was used for comparison.

2.6.1 Crude Protein

According to [15] crude protein of each sample was calculated by multiplying the total nitrogen by the factor 6.25.

2.6.2 Crude Fat (Ether Extract)

Ten g of each powdered sample were extracted using a continuous extraction apparatus (Soxhlet) with a solvent of petroleum ether (b.p.60-80°C) for sixteen hours. Each extract was dried over anhydrous Na₂SO₄, and evaporated to dryness. The residue was dried at 80°C for ten minutes, cooled, weighed and expressed as percent lipid [15].

2.6.3 Crude fiber contents

Two g of the defatted powder of each sample were boiled with 200 ml of 1.25% sulphuric acid under reflux for 30 minutes and filtered. The Residue was washed with distilled water, then transferred back to the flask with 200 ml of 1.25% NaOH Solution. Boiling for 30 minutes under reflux, rapidly filtered and washed with distilled water. The residue was dried at 100°C to constant weight> the difference between the weight of residue after drying at 110°C and the powder represents the weight of crude fiber [15].

2.6.4 Moisture Contents

Five g of each air-dried powder sample were accurately weighed in a porcelain crucible, and then dried in an oven at 105°C until constant weight was obtained. The loss in weight was calculated and reported as percent moisture [15].

2.6.5 Total ash content

Two g of sample were added into previously weighed porcelain crucible, place in muffle furnace at 600°C for 2 hours, and then placed in desiccators, cool and weigh. The weight of the residue was calculated and expressed as percent ash [15].

2.6.6 Determination of total carbohydrate

Percentage carbohydrate was given by: 100 - (percentage of ash + percentage of moisture + percentage of fat + percentage of protein) [16].

2.7 Germination test

The germination test was carried out under laboratory conditions to evaluate the effect of Aerosil 200 nanoparticles at lower limit LC₉₀ on wheat seeds after 6 months from storage. 30 seed were randomly picked from treated and untreated groups and placed on a surface layer of cotton wool

in petri dish (6×1 cm) which wetted carefully with a tap water at the rate of 10 seeds per plate. Seeds were arranged in a completely randomized design with 3 replicates. 7 days later the germination percentage was calculated thus [17, 18].

2.8 Chlorophyll estimation

Experiment was carried out at Soil, Water and Environment Research Institute at the end of germination test to study the Chlorophyll content (%) of the seedlings leaves [19].

2.9 Data analysis

The mortality (%) of *T.granarium* larvae after 10th day from treatment was probit analysis using a computer program named LDP-line according to [20]. From which the toxicity value (LC₉₀) was estimated. Slope values and toxicity index were also estimated.

Insect mortality data were subject to analysis of variance and differences using ANOVA test (a computer program costate). Mean values were adjusted by Duncan's Multiple Range test [21] at 0.05% level of significance with Statistical software version 6.3.0.3.

3. Results and Discussion

3.1. Structural study of AEROSIL 200

The shape and size of AEROSIL 200 were checked by transmission Electron Microscope (TEM). Figure (1) indicates that the original morphology of particles approximately spherical with the diameter varying between 5 to 20 nm.

3.2 Biological studies

Results recorded in Table (1) for duration of eggs, larvae, pupae stages, adults longevity and life span of *T. granarium*, *S. paniceum* and *T. castaneum* insects reared at 30±2 °c and 65±5 % R.H. showed that incubation period of egg were 6.1, 5 and 5.5 days and larval duration recorded 33.7, 35 and 23.2 days while pupal duration were 6.5, 4 and 7 days for *T. granarium*, *S. paniceum* and *T. castaneum* insects respectively. Longevity of adults was 10 and 10 days for *T. granarium* and *S. paniceum* while for *T. castaneum* was over one year. Meanwhile life span (from egg to adult mortality) was 46.3, 44 and 35.7 days for *T. granarium*, *S. paniceum* and *T. castaneum* insects respectively. This finding goes on the same harmony with results [22] who studied the effect of different temperature on biology of some stored grains insects.

3.3 Activity of toxic effect

3.3.1 Adult stage

The toxic effect of AEROSIL 200 NPS against adults of tested insects, *T. granarium*, *S. paniceum* and *T. castaneum* and reduction % in F₁- progeny was showed in Tables (2, 3 and 4). 1.5 and 2 gm/kg fumed silica was sufficient to cause 100% mortality with *T. granarium* and *S. paniceum* after 5 and 2 days of treatment while for *T. castaneum* which known by its highly resistance for most alternatives, 4 gm/kg recorded 83% mortality after 21 days from treatments.

Reduction % in F₁- progeny ranged from 93.7-100 %, 90-100% and 56.5-100% with *T. granarium*, *S. paniceum* and *T. castaneum* respectively.

These results are in harmony with [23] revealed that fumed silica, AEROSIL 200 NPS exhibited significant strong toxic effect (P<0.05) @ the highest conc., 1gm/kg for *C.maculatus*, 1.5 gm/kg for *R.dominica* and 2.5gm/kg for *S.orzae* and

reduce the progeny by 100% at the highest conc. Also many previous researches experimented the toxicity of silica and silicon oxide NPS against stored grain insects such as [24] recorded that, silica NPS applied at 0.5 and 0.25 g kg⁻¹ dosages against cigarette beetle, *Lasioderma serricorne* Fabricius showed the superior performance of the over other treatments at one day after treatment which caused higher mortality, reduced oviposition and adult emergence and has a great promise in cigarette beetle pest management. Another finding by [25] reported that, two silicon dioxide NPS of Aerosil ® and Nanosav have high toxicity on *R. dominica* and *T. confusum* adults. At low concentrations, Aerosil® was more effective than Nanosav. It has been revealed that the control efficacy against adult *Tribolium castaneum* was about 80%, presumably due to the slow and persistent release of the active components from the nano-particles [26].

3.3.2 Larval stage

Toxicity of fumed silica on larvae of *T. granarium*, *S. paniceum* and *T. castaneum* was presented in Tables (5 and 6). The mortality % of *T. granarium* and *T. castaneum* larvae were 83.3 %, 65.4 % after 11 and 21 days from treatment at conc. 4 gm/kg. This indicated that larvae of both tested insects are more resistance than its adults while larva of *S. paniceum* showed 100 % reduction in F₁-progeny at conc. 2.5 gm/kg. Larvae of *T. confusum* was more susceptible to Nano-Diatomaceous earth than *T. castaneum* larvae [26].

Aerosil 200 NPS known by its absorption characters when applied the insect began to lose water due to damage of the water barrier [1]. This hypothesis for the physical mode of action makes the case in the use of nanocides stronger and the nanocides can be removed by conventional milling process unlike sprayable formulations of conventional pesticides, leaving residues on the stored grain. Therefore, silica NPS have a good potential to be used as grain protecting agent and alternatives to chemical insecticides [27].

3.3 Residual toxicity experiment

Previous results indicated that larvae of *T. granarium* was more tolerance than other tested adults and larvae insects so the residual toxicity at lower limit LC₉₀(6 gm/kg) was tested for six months storage. As recorded in Table (7), data indicated that, fumed silica can protect wheat seeds from insect infestation until the 4th month. [28] Revealed that, egg production was highly suppressed by nano-DE under stored conditions. The mean number of deposited eggs per female (the fertility) and % of adult emergence (F₁) of each tested insects were greatly affected with highly significant differences. Nano-DE strongly suppressed the number of deposited eggs of *T. confusum* more than *T. castaneum* after 20, 90 and 120 storage interval days, respectively. The persistent effect of nanoparticles displayed several different modes of action by reducing oviposition, adult emergence (F₁) and infestation percentages of tested insects [29]. Found that two aerogels, Gasil and Aerosil prevented F₁-progeny of *Prostephanus truncatus* (Horn) more than 6 months after treatment.

3.4 Seeds components analysis

Table (8) represented wheat components % (crude protein, crude fat, total carbohydrate, crude fiber, moisture contents and Total ash) of treated wheat grains with lower limit LC₉₀ of fumed silica after 6 months storage. Untreated seeds were used for comparison.

Results showed differences between the components of treated and untreated seeds in which the crude protein %, total carbohydrate%, crude fiber% and total ash% were increased while crude fat % and moisture % decreased in treated grains when compared with untreated seeds. Decrease of seed moisture content was useful for safe storage, this may be due to characters of silica which known with absorbent moisture material. Prior of storage, the moisture content of cultivars seed as good quality based on certification standard cited by [30] have recommendation which moisture content for long duration storage seed does not above 14 or below 5 %. Seeds store at moisture content above 14% begin to exhibit increased respiration, heating and fungal invasion that destroy seed viability more rapidly, while below 5 % cause seed membrane structure, seed deterioration, thus improve that it had no significant effect on seeds viability.

3.5 Germination test and Chlorophyll estimation

According to data presented in Table (9) germination % of seeds stored for 6 months after treatment with lower limit LC₉₀ of fumed silica slightly affected comparing with untreated seed. Also chlorophyll analysis showed that fumed silica slightly decreased chlorophyll % in treated seeds. These results agreed with that of [31] recoded that, a slight effect in germination of wheat grains with the silica oxide (SiO₂) and aluminum oxide Al₂O₃ nanoparticles was observed as compared with control [27]. Found that, application of mesoporous silica NPS (MSNs) as a smart delivery system to agricultural crops do not have any negative impacts on seeds germination, plant growth or development.

Table 2: Percent mortality (mean \pm SE) of *T.granarium* adults treated with Aerosil 200.

Conc. (gm/kg)	Adult mortality after indicated days				% reduction in F ₁ progeny
	2	3	5	7	
1.5	61.7 \pm 9.2 ^a	98.3 \pm 1.6 ^a	100 \pm 0 ^a	100 \pm 0 ^a	100
1	58.3 \pm 4.4 ^a	96.7 \pm 1.6 ^a	100 \pm 0 ^a	100 \pm 0 ^a	100
0.5	50 \pm 7.6 ^{ab}	88.3 \pm 4.4 ^{ab}	96.7 \pm 3.3 ^{ab}	100 \pm 0 ^a	100
0.25	38.3 \pm 7.2 ^{bc}	83.3 \pm 6 ^{bc}	96.6 \pm 3.3 ^{ab}	100 \pm 0 ^a	100
0.12	25 \pm 7.3 ^c	76.7 \pm 1.6 ^c	88.3 \pm 1.7 ^b	100 \pm 0 ^a	95.3
0.06	21.7 \pm 3.3 ^c	56.7 \pm 4.4 ^d	76.7 \pm 4.4 ^c	96.7 \pm 3.3 ^a	93.7
LSD 5%	17.9	11.5	8.4	4.2	

Means within a column followed by the same lower case letter are not significantly different (P<0.05)

Table 3: Percent mortality (mean \pm SE) of *S.paniceum* adults treated with Aerosil 200.

Conc. (gm/kg)	Adult mortality after indicated days					% reduction in F ₁ progeny
	1	2	3	4	5	
2	78.3 \pm 1.7 ^a	100 \pm 0 ^a	100 \pm 0 ^a	100 \pm 0 ^a	100 \pm 0 ^a	100%
1.5	61.5 \pm 1.7 ^b	100 \pm 0 ^a	100 \pm 0 ^a	100 \pm 0 ^a	100 \pm 0 ^a	100%
1	41.5 \pm 7.3 ^c	96.5 \pm 3.3 ^a	100 \pm 0 ^a	100 \pm 0 ^a	100 \pm 0 ^a	100%
0.5	38.3 \pm 4.4 ^c	83.3 \pm 1.7 ^b	95 \pm 2.9 ^{ab}	100 \pm 0 ^a	100 \pm 0 ^a	100%
0.25	5 \pm 2.8 ^d	55 \pm 2.8 ^c	93 \pm 1.6 ^b	96.5 \pm 1.6 ^a	100 \pm 0 ^a	93%
0.2	3.3 \pm 3.3 ^d	31.5 \pm 4.4 ^d	81.5 \pm 3.3 ^c	96.5 \pm 3.3 ^a	100 \pm 0 ^a	90%
LSD 5%	12.4	8.1	5.9	4.7	0	

Means within a column followed by the same lower case letter are not significantly different (P<0.05).

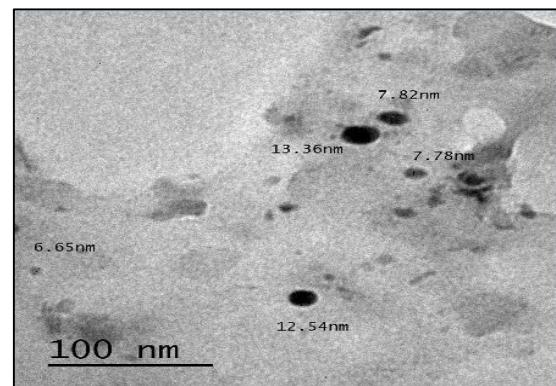
Table 4: Percent mortality (mean \pm SE) of *T.castaneum* adults treated with Aerosil 200.

Conc. (gm/kg)	Adult mortality after indicated days							% reduction in F ₁ progeny
	2	4	6	8	10	14	21	
4	8.3 \pm 3.3 ^a	25 \pm 2.9 ^a	43.3 \pm 3.3 ^a	61.6 \pm 4.4 ^a	73.3 \pm 4.4 ^a	81 \pm 4.4 ^a	83 \pm 7.6 ^a	100%
3	5 \pm 2.9 ^{ab}	23.3 \pm 4.4 ^a	36.6 \pm 6 ^a	40 \pm 2.9 ^b	46.6 \pm 1.7 ^b	62 \pm 3.3 ^b	71.7 \pm 6 ^a	100%
2.5	0 \pm 0	6.6 \pm 1.7 ^b	23.3 \pm 1.7 ^b	36.6 \pm 4.4 ^{bc}	43.3 \pm 4.4 ^{bc}	51.7 \pm 3.3 ^{bc}	66 \pm 5.8 ^a	76%
2	0 \pm 0	0 \pm 0 ^b	8.3 \pm 1.7 ^c	23.3 \pm 6.6 ^c	31.6 \pm 6 ^c	36.2 \pm 6.6 ^c	37.8 \pm 7.6 ^b	56.5%
LSD 5%	7.2	9	11.5	15.6	14.4	15.1	23.5	

Means within a column followed by the same lower case letter are not significantly different (P<0.05)

This paper results revealed that, AEROSIL 200 NPS can provide as a good alternative to protect stored seeds from insect infestation during storage without affecting germination.

4. Tables and Figures



X: magnification power

Fig 1: The TEM images of Aerosil 200 nano particles (x=40)

Table 1: Biological duration of different stages of the three insects reared at 30 \pm 2°C and 65 \pm 5 % R.H.

Parameter	Egg (day)	Larva (day)	Pupa (day)	Adult (day)	Life span (day)
<i>T. granarium</i>	6.1	33.7	6.5	10	46.3
<i>S. paniceum</i>	5	35	4	10	44
<i>T. castaneum</i>	5.5	23.2	35.7	Over year	35.7

Table 5: Percent mortality (mean \pm SE) of *T.granarium* larvae treated with Aerosil 200.

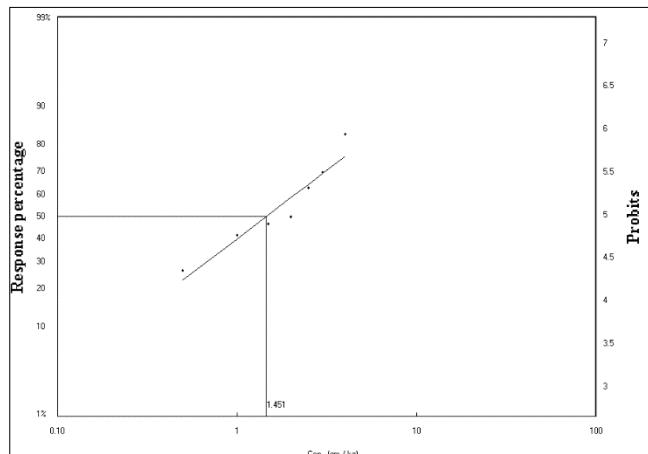
Conc. (gm/kg)	(%) Larva mortality after indicated days				
	2	4	6	8	11
4	18.3 \pm 4.4 ^a	28.3 \pm 4.4 ^a	53.3 \pm 1.7 ^a	61.6 \pm 1.7 ^a	83.3 \pm 1.6 ^a
3	11.6 \pm 1.6 ^{ab}	21.6 \pm 1.7 ^{ab}	45 \pm 2.9 ^{ab}	53.3 \pm 1.7 ^a	70 \pm 4.9 ^b
2.5	10 \pm 2.9 ^b	21.6 \pm 2.9 ^b	41.6 \pm 4.4 ^{bc}	51.6 \pm 4.4 ^a	63.3 \pm 4.3 ^b
2	6.6 \pm 1.7 ^{bc}	15 \pm 2.9 ^{bc}	31.6 \pm 1.7 ^{cd}	40 \pm 2.9 ^b	50 \pm 2.9 ^c
1.5	5 \pm 2.9 ^{bc}	11.6 \pm 3.3 ^c	30 \pm 2.9 ^d	36.6 \pm 4.4 ^{bc}	46.6 \pm 6 ^c
1	0 \pm 0 ^c	8.3 \pm 1.7 ^c	26.6 \pm 4.4 ^d	33.3 \pm 4.4 ^{bc}	41.6 \pm 4.4 ^c
0.5	0 \pm 0 ^c	0 \pm 0 ^d	13.3 \pm 4.4 ^e	23.3 \pm 4.4 ^c	26.6 \pm 4.4 ^d
LSD 0.05	7.4	8.3	10.3	10.9	13.1

Means within a column followed by the same lower case letter are not significantly different (P<0.05)

Table 6: Percent mortality (mean \pm SE) of *T.castaneum* larvae treated with Aerosil 200.

Conc. (gm/kg)	Larva mortality after indicated days						
	2	4	6	8	10	14	21
4	0 \pm 0	0 \pm 0	11.6 \pm	16.6 \pm 3.3 ^a	41.6 \pm 8.7 ^a	51.6 \pm 6 ^a	65.4 \pm 5.7 ^a
3	0 \pm 0	0 \pm 0	0 \pm 0	15 \pm 2.9 ^{ab}	28.3 \pm 1.7 ^{ab}	35 \pm 4.4 ^{ab}	44.2 \pm 4.4 ^b
2.5	0 \pm 0	0 \pm 0	0 \pm 0	8.3 \pm 1.7 ^b	21.6 \pm 4.4 ^b	30 \pm 5 ^b	32.6 \pm 4.4 ^b
2	0 \pm 0	0 \pm 0	0 \pm 0	1.6 \pm 1.7 ^c	13.3 \pm 3.3 ^b	20 \pm 2.9 ^c	21 \pm 3.3 ^c
LSD 5%	-	-	-	8.2	17.2	15.4	14.8

Means within a column followed by the same lower case letter are not significantly different (P<0.05)

**Fig 2:** Toxicity line of *T. granarium* larva treated with aerosil 200 nano particles.**Table 7:** Residual toxicity of aerosil 200 nanoparticles at lower limit LC₉₀ against *T.granarium* larvae for 6 months storage.

Month	% mortality
1	100
2	100
3	98.3
4	96.6
5	85
6	70

Table 8: Effect of LC₉₀ of aerosil 200 nanoparticles on wheat seeds components after 6 months storage.

Components %	Treated seeds	Untreated seeds
Crude protein %	11.41	9.83
Crude fats %	2.12	2.69
Total carbohydrate%	76.05	75.23
Crude fiber %	5.78	4.95
Moisture %	9.63	11.12
Ash %	1.79	1.13

Table 9: Effect of AEROSIL 200 NPS @ LC₉₀ on germination % and chlorophyll % of wheat seeds after 6 months storage.

	Mean germination%	Mean Chlorophyll%
Treated seeds	93.3 \pm 6.6	0.45
Untreated seeds	96.6 \pm 3.3	0.49

5. Conclusion

AEROSIL 200 NPS can provide as effective alternative against stored grains insects which can protect seeds stored for several months without any harmful effect on seeds contents % or germination %.

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