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The insecticidal effect of silica nanoparticles on *Callosobruchus maculates* (Coleoptera: bruchidae) and its side effects

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ABSTRACT

Stored product insect pests were responsible for considerable quantitative and qualitative losses of agricultural products mainly cereals and legumes. Laboratory studies were carried out to evaluate the insecticidal potential based on contact and residual effect of the inert dust of silica nanoparticles (35±5nm) against the adult of stored pulse beetle *Callosobrachus maculatus,* in addition its effect on germination of the test seeds. Concerning the contact treatment, mortality assessed after 7 and 14 days from exposure to the three treated seeds (cow pea, kidney bean and chick pea) with silica nanoparticles (SNPs) among the different concentrations of SNPs 200 ppm was proved to be superior by recording highest mortality, lowest adults emerged, least seed damage in both cow pea and chick pea seeds up to 14 days of storage. Regarding the residual assay, the treated seeds storage for 7 and 15 days and exposure to *C. maculatus* was accomplished a complete mortality (100%) after 168 h on the three seeds varieties post treatment for 7 days (one week) concluded that (SNPs) was not proper effect and less efficient with long period. Treated seeds of these three varieties of pulses revealed no effect on the germination and the growth rate of root and shoot of cow pea and chick pea, except the kidney bean was decreased the germination compared to respective control. Scanning electron microscope (SEM) was used to observe the adult insect exposed to silica nanoparticles treated seeds. The particles attached all over the insect body of the bruchid beetle with scratches and splits on the cuticle that lead to the loss of water through dehydration which eventually led to death. Silica is promising material for controlling the tested insect as safe alternative to synthetic insecticides.

Keywords: *C. maculatus*, insecticidal potential, stored products, silica nanoparticles, the germination

1. Introduction

Post-harvesting crops exposed to many insect pests causing extensive damage in yield, grain damage arising from direct feeding of insects on endosperm and grain embryos increases the exposure of grain to rot because scratches lead to unpleasant odors which cannot be accepted by humans and animals (Ismail, 2014 and Kalpna *et al*., 2022).This situation necessitated application of some control measures in order to minimize the losses caused by this stored product pests. The control of stored grain pests stands mostly on broad action insecticides and fumigants unfortunately, this leads to contamination of food with toxic pesticide residues (Debnath *et al*.,2011) thus, it is crucial to find new alternative control methods for stored products, environmentally safe and convenient methods such as the use of inert dusts, plant extracts, oils, leaf powders, pressurized carbon dioxide and temperature management techniques (low and high temperature) are the growing interest to replace synthetic pesticides (Yuya *et al.*, 2009).

Nano technology has become one of the most promising new approaches for pest control in recent years. Materials including diatomaceous earth, silica aerogels, silica nanoparticles natural zeolite and zeolite in the nanoform have been evaluated against some stored product insect pests.

Nanoparticles have helped to produce new pesticides, insecticides and repellents Owolade *et al*. (2008) and Yang *et al*. (2009) found that nanoparticles loaded with garlic essential oil is efficacious against

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Tribolium castaneum, also, Stadler *et al.* (2010) showed that nano alumina can be successfully used to control stored grain pests.

On other hand, some researchers believe that nanoparticles could inhibit plant growth Yang and Watts, (2005) indicated that the alumina nanoparticles in ground water inhibited the growth of some vegetables (cabbage, cucumber and soybean).

Silica nanoparticles (SNPs) have been evaluated against the stored grain insect the rice moth *corcyra cephalonica* (Vani and Brindhaa, 2013), the lesser grain borer beetle *Rhyzopertha dominica* and red flour beetle *Tribolium castaneum* (El-Samahy *et al*., 2014) and against the stored pulse beetle, *Callosobruchus maculatus* (Arumugam *et al*.,2016).

The cow pea beetle *callosobruchus maculatus* (Col., Bruchidae) is one of the major pest of many stored products such as cow pea, chick pea, green, black and red gram, lentil and soy bean (Edde and Amatobi, 2003).The development of a single larva in a grain seeds can lead to weight losses of 8to 22% of weight (Credland *et al*., 1986).

The eggs are cemented to the surface of pulses and are smooth, domed structures with oval and flat bases. The larvae and pupae are normally only found in cells bored within the seeds of pulses (Chavan *et al*., 1997). The treatment of hydropholic silica nano particles (SNPs) with the pulse seeds against this pest (*Calloso bruchus* spp.) revealed a significant reduction in oviposition, adult emergence and seed damage potential with no effect on seeds germination.

Wazid *et al*.,(2020) indicated that the 1500 ppm of silica nano particles proved to be superior by recording highest mortality, lowest number of eggs, lowest adults emerged, least seed damage (Sorghum and chick pea) up to five months. Comparative insecticidal activity of nano and coarse silica (natural) on the Chinese beetle *C. chinesis* (L) found that NPS had more insecticidal activity against the pest than coarse silica with respect to the exposure time and concentration increased in both type Mesbah *et al*., (2017).

The present investigation was conducted to determine the toxicity of silica nano particles (SNPs) against the *C. maculatus* through the three various of seed varieties (cow pea, kidney bean and chickpeas seeds).The delayed effect of silica nano particles on progeny production and development test pest during the first generation post-parent exposure, in addition to evaluate the residual toxicity of nano silica particles against the adult of the pest by examining under scanning electron microscopic (SEM) photo graphs of SNP-treated and untreated pests. The effect of nano silica on the three varieties of seed germination and development of seedling growth was also observation. Percentage of germination, shoot length and root length were recorded.

2. Material and Methods

2.1. Commodity.

Three grains, cow pea seeds (*Vigna unguiculata*) kidney bean (*Phoseolus vulgaris*) and chickpeas (*Cicer arietinum*) were purchased from local market and placed in the freezer (-4°C) for at least three days in order to kill any alive insects present in products.

2.2. Silica nanoparticles (SNPs)

Silica nanoparticles material used in the experiments was obtained from Nanotech Egypt for photo-Electroncs-The product formula is (Sio_2NPs) and name silica dioxide nanoparticles with physical properties illustrated in table (1): for size and shape characterizations were checked by Transmission Electron Microscope (TEM) performed on JEOl-TEM-2100 high resolution TEM at an accelerating voltage of 200KV(Fig:1).

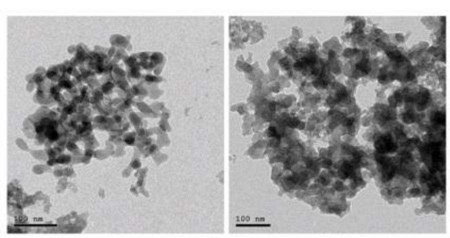


Fig.1: TEM micrograph of hydrophilic SiO_2 NPs

Table 1: Some physical properties of nano silica used in the experiments

Appearance (color):	White
Appearance (form):	Powder
Avg-Size (TEM):<100 nm	(-35±5nm)
Shape (TEM)	Quasi-spherical-like shape
Solubility	Dispersion into water or ethanol

2.3. Insect rearing:

C. maculatus was obtained from laboratory culture in National Research Center of Egypt (NRC); colony was maintained in the laboratory for several generations free from insecticides in laboratory of pests and plant protection department, NRC center. *C. maculatus* reared on sterilized intact cow pea seeds in glass jar (10.9&18cm) covered with fine muslin cloth for ventilation. All experiments were carried out at 30±2ºC and 65±5 RH in continuous darkness.

2.4. Bioassay:

2.4.1. Insecticidal efficiency of silica nano against *C. maculatus*.

The efficacy nano silica particles against *C. maculatus was* determined by direct contact assay (Broussalis *et al*.,1999).The three tested varieties of seeds was performed in small plastic cups, each cup had a radius of 6 cm and height of 6.5 cm. The cups were perforated to allow aeration, 40gm of each variety seeds was placed in each cup. The nano silica dust was mixed at different concentration (50,100 and 200 ppm) in the rate of (2 mg, 4mg and 8mg /40g) respectively, another group served as control (without nano silica dust) and to achieve equal distribution of nano particles on all seeds cups were shaken manually for two minutes, then *C. maculatus* 15 unsexed newly emerged adults (3- days old) were introduced into each cup containing dust. Three replicates were carried out for each variety of seeds treated and untreated was used. Insect mortality % of treated and untreated was recorded after 2,7 and 14 days post-treatment by the number of dead or alive were recorded and the effect of dust on insect progeny production were evaluated (cumulative number of first offspring).

2.4.2. The residual effects of silica nano against *C. maculatus*

Infestation free 40gm of different varieties of cow pea, kidney bean and chickpeas seeds were sun dried and later kept in hot air oven for one h.at50ºC to sterilize to ensure that sample is free from previous infestation and such seeds were treated with the three concentration of silica nano particles powder (200,100,and 50ppm) and kept in plastic cups for one week,10 individual, were introduced into each cup ,the cumulative mortality was recorded after 2,5 and 7days from exposure to the tested varieties. The same steps were recorded after treated the seed varieties with three concentrations of silica nano particles and storage these for two weeks (15days), another different untreated varieties and adult insects were introduced in the each plastic cup used as control insects. Three replicates of treated and untreated varieties were used.

2.4.3. Seeds treatment and germination (phytotxicity test)

Germination and seedling growth of treated the three varieties of seed were evaluated 40 gm from each type of seeds variety (cowpea, kidney bean and chickpeas) were presoaked in water for 24h, then left for 2h in air for dried. 20 seeds from each type of variety treated with different concentration of nano silica particles (200,100and 50 ppm) using contact toxicity assay. The treated seeds were transferred on a piece of moistened cotton inside the petri-dishes (20cm diameter) as well as the untreated seeds (without dust) from each type of variety used control. After treatment, the dishes placed in room temperature.

Percentage of seed germination, shoot length and root length were recorded after 10 days post-treatment. Each treatment in addition the control seeds were repeated three times.

D-Scanning electron microscope (SEM) *C. maculatus* adults exposed to cowpea seeds treated with 200ppm concentration of silica nanoparticles for 7 days were examined under scanning electron microscope (Jeol-TSM-5600LvM) in the Electronic Microscope Unites central, Cairo.

2.5. Statistical analysis

The results obtained were analyzed by using Student's t-tests ($p < 0.05$) to compare between the means of treated and untreated larvae.

3. Results

3.1. Insecticidal efficiency of silica nanoparticles against *C. maculatus*:

The insecticidal activity of silica nanopaticles (NSP) was determined by direct contact application. The results obtained in (table 2) indicated that cumulative mortality % increased with increased concentrations and exposure periods. Mortality due to the treatment of hydrophilic silica mixed with all varieties of seed that both doses of 200 ppm and 100ppm were most effective concentrations against *C. maulatus*, specially after 7 days from exposure the treated kidney bean seeds (*Phaseolus vulgaris*) were more susceptible and sensitive among the tested varieties and nano silica totally controlled the adults after 14days from exposure accordingly insect progeny production in f1 generation of *C. maculatus* reproduction none occurred in all cases of different concentrations and achieved high inhibition rate (IR%) (100%) reduction rates (table 2) while, the offspring production on treated cow pea (*Vigna unguiculata*) and chickpeas *(Cicer arietinum*) ranged from (35.9-87.3%) and (64.2-80.4%) respectively.

Table 2: Mortality percent of *Callosobruchus maculatus* adults exposed for seeds treated with hydrophilic silica-nano particles.

Silica nanoparticles Dose (ppm)	Periods in Days (Mean±SE)				IR%
	(Vigna unguiculata)				
	2	**7**	**14**	**F1**	
200	86.67±13.33***	100±0.00***	-------	8.00±4.62**	87.3
100	26.67±6.67*	88.90±5.88 ns	100±0.00**	38.33±19.53 ns	39.2
50	24.43±4.43**	86.67±13.33ns	100±0.00**	40.33±20.27ns	35.98
Control	8.88±2.21	71.13±8.02	86.67±7.71	63.00±11.68	
	(*Phaseolus vulgaris*)				
200	86.67±6.67***	100±0.00***	--------	----	
100	86.67±6.67***	100±0.00***	100±0.00	----	
50	71.10±19.76***	62.2±17.77ns	100±0.00	-----	
Control	6.67±6.67	66.67±3.84	100±0.00	-----	
	(*Cicer arietinum*)				
200	84.45±9.69***	100±0.00***	-------	5,00±1.00ns	80.4
100	64.47±9.68***	91.13±4.43***	100±0.00***	7,00±2.00 ns	72.5
50	60.00±3.87***	80.00±17.63**	100±0.00***	9.00±1.00ns	64.7
Control	4.43±4.43	64.43±8.02	91.13±4.43	25.50±6.70	

* Significant of differences from "un-treated" control (taking into account the Student`s t-test): p=0.05. Ns: denotes not significant.

3.2. Residual effect of silica nanoparticles against *C. maculatusa* adults:

After treatment the three varieties of seeds with different concentrations of silicanano particles and storage for 7days and another set storage for 15days.The results of the present study elucidated the variation in mortality of *C. maculatus* due to different concentrations used of silica nanoparticles in addition the time of exposure. At 48h from exposure at 200ppm of(NSP) exhibited 87.2,90.0 and 80.0% adults mortality in different seeds post treatment (7days storage) were as 120h after exposure significantly highest mortality (100%) was recorded in three varieties compared to control, while the accumulative mortality of *C. maculates* adults was recorded in silica nanoparticles at 200ppm showed 100% after 168hin seeds (cow pea and kidney been) post treatment at 15 days (storage) except chickpeas pea was 95.5% (table 3). It was found that when the adults *C. maculatus* were exposed to (SNP) at different concentrations (200,100 and 50) and different time intervals on the three seeds variety post-treatment for 7days (one week) more effect than 15 days (2weeks)silica nano particles was not proper effect and less efficient with long period use

Table 3: Residual toxicity of hydrophilic silica-nano particles were evaluated after 7 and15 -day post treatment of seeds.

Seeds	Silica nanoparticles Dose (ppm)	7 (days)			15 (days)		
		Mortality percent of *Callosobruchus maculatus* adults post exposure time (H)					
		48	**120**	**168**	**48**	**120**	**168**
Vigna unguiculata	**200**	87.2±3.65**	100±0.00**		51.13±8.02**	95.57±4.42**	100±0.00**
	100	76.67±6.67^{ns}	93.33±6.67^{ns}	100±0.00^{ns}	60.0±3.87**	82.23±2.23**	97.77±2.23**
	50	60.90±6.57^{ns}	80.33±6.04^{ns}	90.00±5.77^{ns}	40.0±3.87**	62.23±2.23**	77.77±2.23^{ns}
	Control	55.0±10.41	71.1±6.75	86.67±6.67	15.57±8.02	24.42±12.37	71.1±5.88
Phaseolus vulgaris	**200**	90.0±5.77**	100±0.0**		62.2±16.02**	82.23±9.68**	100±0.00**
	100	76.67±8.82**	96.67±3.33**	100±0.0**	40.00±5.77^{ns}	80.0±11.55**	95.00±5.00*
	50	70.0±5.77**	83.3±6.67**	96.67±3.33**	23.3±8.82^{ns}	53.33±14.52^{ns}	90.0±5.77*
	Control	23.33±8.82	53.33±3.33	76.67±3.33	28.9±5.88	53.33±3.84	71.13±4.42
Cicer arietinum	**200**	80.0±5.77**	100±0.0**		64.33±5.89**	84.4±5.92**	95.57±4.43*
	100	73.33±8.82**	83.33±8.82*	95.0±5.00**	46.67±7.71*	68.9±5.88**	84.47±2.23*
	50	66.67±3.33**	80.0±5.77*	96.67±3.33**	37.8±5.88^{ns}	55.6±5.88*	75.53±2.22^{ns}
	Control	30.0±5.77	56.67±8.82	76.67±3.33	14.36±11.18	33.33±7.71	68.87±4.42

* Significant of differences from "un-treated" control (taking into account the Student`s t-test): p=0.05. ns: denotes not significant

3.3. Effect of silica nanoparticles on germination and growth of the three varieties seed

The seed germination provides a suitable foundation for plant growth, development and yield. In this study, seed germination was not affected by the treatment of SNP and had no phytotoxic accordingly proved best giving the highest value for germination % in chickpeas seeds, the % of germination was 75% compared with control 85% at the highest application rate (200 ppm) of SNP and had no significant effect on shoot and root growth (table 4 and Fig.2), while in kidney bean application of 100, 200 ppm of SNP was very weak and decrease the germination by 35and 30% compared the respective control (80%) at the same time had no significant effect on the shoot and root growth .On the other hand germination of cow pea seeds was consider moderate affected by the treatment of 100and 200ppm concentration that showed significantly decreased in shoot and root length were reached to less than 50% compared to control after 10 days from treatment.

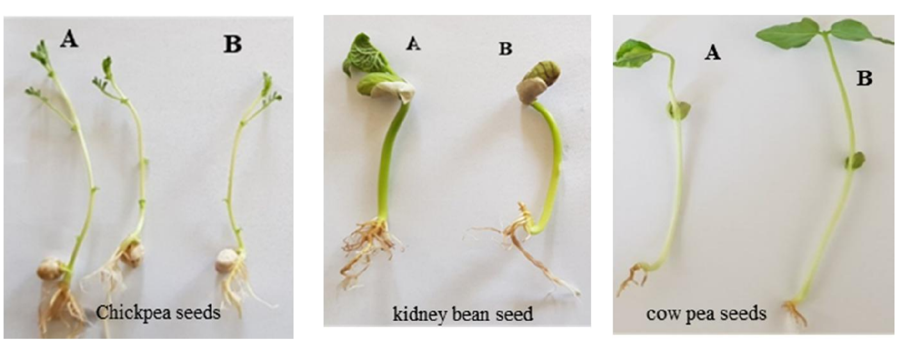


Fig. 2: Effect of $NSiO_2$ on germination and growth of the three varieties seeds
(A) Untreated seeds, (B) treated seeds

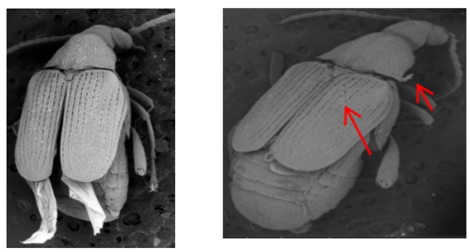
Table 4: Effect of hydrophilic silica-nano particles treated seeds on germination and growth of some various seeds.

	Concentration of SNPs (ppm)	Seed varieties		
		Vigna unguiculata	***Phaseolus vulgaris***	***Cicer arietinum***
Germination (%)	Control	95	80	85
	50	80	65	80
	100	35	35	80
	200	65	30	75
Shoot length (cm)	Control	15.100± 0.458	3.600± 0.620	8.167±0.601
	50	13.167± 1.682^{ns}	3.600± 1.005 ^{ns}	5.80±5.80*
	100	6.667± 0.333***	9.200± 1.497**	7.417±0.554^{ns}
	200	8.400± 1.461**	5.100± 1.166 ^{ns}	7.700±1.044^{ns}
Root length (cm)	Control	5.600± 0.678	2.700± 0.436	6.583±0.757
	50	3.583± 1.114^{ns}	1.700± 0.255 ^{ns}	7.200±0.490^{ns}
	100	3.00± 0.58*	2.700± 0.561 ^{ns}	5.857±1.392^{ns}
	200	2.400± 2.400**	2.900± 0.510 ^{ns}	5.750±0.512^{ns}

* Significant of differences from "un-treated" control (taking into account the Student`s t-test): p=0.05. ns: denotes not significant

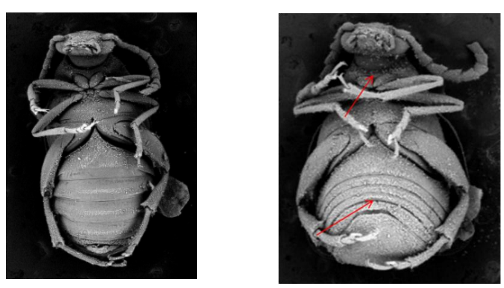
3.4. Scanning electron microscope (SEM) :

Scanning electron examination revealed that the image of untreated *C .maculatus* adults, the insect is retaining its antennae and legs (Fig 3A &Fig 4A). Images analysis showed that silica nanoparticles adhered to all body parts, including head, thorax, abdomen and caused abrasion like elytra and legs the insect appeared lost its mouse parts elytra and



(A) **(B)**

Fig. 3: Scanning electron microscope photographs of *C. maculatus* adults exposed to untreated (A) cowpea (dorsal view). Dorsal view, silica-nanoparticles (200 ppm) aggregate on insect surface, aggregate between thorax and abdomen joints (arrow) silica particles (arrow) attached on the elytra (B).



(A) **(B)**

Fig. 4: Scanning electron microscope photographs of *C. maculatus* adults exposed to cowpea seeds. (A) ventral view of *C. maculatus* adults exposed to untreated cowpea. (B) Ventral view, silica-nanoparticles attached on abdomen and distributed near the rostrum (arrow) aggregate between head and thorax (arrow).

Legs and distributed on the near rostrum (Fig 3B& Fig 4B) silica nanoparticles homogeneously distributed on the outer surface of the insect. The obtained images revealed that the reduction of silica nanoparticles size could possibly increase dust even further when particles applied to cow pea seeds to control *C. maculates*.

4. Discussion

An attempt was made in the present study to test the efficacy of silica nanoparticles (~35±5) to protect the three pulse seed varieties (cow pea, kidney bean and chickpeas) from the infestation of the stored product insect pest, *C. maculatus* through insecticidal and residual effect and its effect on germination of the different varieties of seed used.

It was observed from the present study that seeds treated with SNPs increased the percentage of mortality levels of *C. maculatus* adults and reached to maximum rate at concentration (200ppm) after 7days, data show 7 days was sufficient exposure period for significant effects due to desiccation and spiracles blockage, this could have prevented mating and reduced the fecundity, reduced egg laying could be due to lower insects surviving. (Voigt *et al*., 2009). The silica nanoparticles were desiccation or blockage of spiracles, but also the surface enlargement of the integument as consequence of dehydration (Arumugam *et al*., 2016).

The current researches indicated that *C. maculatus* was more sensitive to natural inert dust or its formulations than other stored product, this may be attribute to inner or gentle wax coats (Mesbah *et al*., 2017). It could be concluded that the main cause of insect death is the dehydration caused by silica nanoparticls that affect or absorb, the wax layer from the body of the insect (Debnath *et al*., 2011, Arumugam *et al*., 2016 and Lu *et al*., 2017).

In the present study the three varieties seed post treatment with silica nanoparticles at high application rate (200ppm) and storage for 7and 15 days. Adults' mortality after 168h of exposure, remarkably residual toxicity of silica nanoparticles was achieved after post storage for 7days more than 15days post storage. The silica nanoparticles (SNPs) became less effect and not proper these results agree with (Boraei *et al*.,2015) noticed that the complete mortality (100%) was occurred on the 6th day from treatment with fumed silica against *C. maculatus* and complete reduction in f1-progeny was recorded at the highest concentration (1gm/ Kg) and are most effective under conditions of low humidity causing desiccation of water is lost due to destruction of waxy layer of cuticle by absorption (Haroun *et al*.,2019) on contrast, (Wazid *et al*.,2020) mentioned that the concentration of 1500ppm of silica nanoparticlis proved to be superior by recording highest mortality, lowest number of eggs, least seed damage (sorghum and chickpea) up to five months of storage, showed superiority in preventing the adults emergence of rice weevil *Sitophilus oryza* and *Callsobruchus analis*.

The comparative assessment of persistence indicated that SNPs remained effectives untile7days exposure to different seeds treated with 200 ppm concentration was totally controlled the adults consequently its progeny production in f1- generation of *C. maculatus* non occurred specially on treated kidney bean, this according to the efficacy and insecticidal potential of SNPs which depending on the rate dose, exposure intervals and the particles size, more over the physical characteristics of the seeds are also an important factors for the attachment of nanoparticles on their surface, in addition insect species and their development stage (Bohinc *et al*., 2018, Bohinc and Trdan ,2017) .

On the other hand, Fields and Korunic (2000) indicated that certain factors which are typical for inert dust such as temperature and relative humidity may influence the mortality of different species of stored product pests.

Though SPNs used in our experiment are amorphous in nature and silica is considered less reactive material (Debanath *et al*., 2011). Phytotoxic studies, seed coats play a very important role in protecting the embryo and can have selective permeability to the external factors (Wierzbicka and Obidzinska, 1998).

In the present study, toxicity of silica nanoparticles on the germination and growth of pulse seed of the three varieties (cow pea, kidney bean and chick pea) was studied and observed that SNPs was enhanced seed potential by increasing characteristics of seed germination with cow pea and chick pea except kidney bean at the application of the high rate of concentration 200ppm of SNPs proved best by giving the highest values for percent seed germination. These results agree with the finding of Nair *et al*. (2011) observed better germination of seeds of rice in presence of FITC-labeled silica nanoparticles.

(Arumugam *et al.,* 2016) indicated that SNPs treated seeds of six varieties of pulses revealed no effect on growth of seeds in term of seed germination, growth rate of root and shoot.
Increase in germination may be due to the absorption and utilization of SNPs by seeds, in addition, the physical characteristics seeds play a significant role in limiting the coating maximum surface area on the seeds by silica nanoparticles (Suriyaprabha *et al*., 2012).

In this study, all tested concentrations used had no affected shoot and root length of seedling of chick pea and kidney bean.

Similarly, silica nanoparticles significantly enhanced the seed germination characteristics in tomato such as %of germination and growth seedling by their direct and indirect involvement in the root and shoot growth and invites (permition)that the nanosilica used as fertilizer for crop improvement (Siddiqui and Al-whaibi 2014, Haghighi *et al*., 2012) while, Khalaki *et al*., (2016), compared the effects of silica nanoparticles and silver nanoparticles at 20and 60%in concentrations percent, root and shoot length of *Thymus Kotschyanus* seed germination was controlled for 14 days. Results showed that the increasing silica nanoparticles concentration had enhanced the seed germination, in contrast, the increase of silver nanoparticles concentration had decreased the germination traits and concluded that, the property or efficacy of nanoparticles based on their size and its type.

In our study, the scanning electron microscopic (SEM) was used to observe the surface and morphology of insect adults of *C. maculatus* introduced into the SNPs treated seeds clearly showed attachment of nanoparticles all over the body of the pest with scratches and splits on the cuticle adhered to all body parts including the head, thorax, abdomen and elytra in addition lost the large parts of its mouth parts. These results agree with Sayed and Salem, (2019), while Arumugam *et al*., (2016) and Mesbah *et al*., 2017 reported that the nanoparticles were more distributed over the insect body affecting more surface area and increasing the effects caused drying activity of insect treated with silica, the effect on the wax layer increased thus increasing the loss of water through dehydration as the water barrier is damaged and die out of desiccation which eventually led to death of the pest Stated by Ebeling (1971).

5. Conclusions

In this study, it could be concluded that silica nanoparticles would be effective to be applied for protection of stored grains against those insect pests attacking grains and can be used at low concentration considering the moisture content of accommodation which the affect the activity of silica, in addition nano silica can be used as a safe and low cost nanocide to control *C. maculatus,* and its efficacy varied depending on the dose rates , exposure intervals and the type of insect species.

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