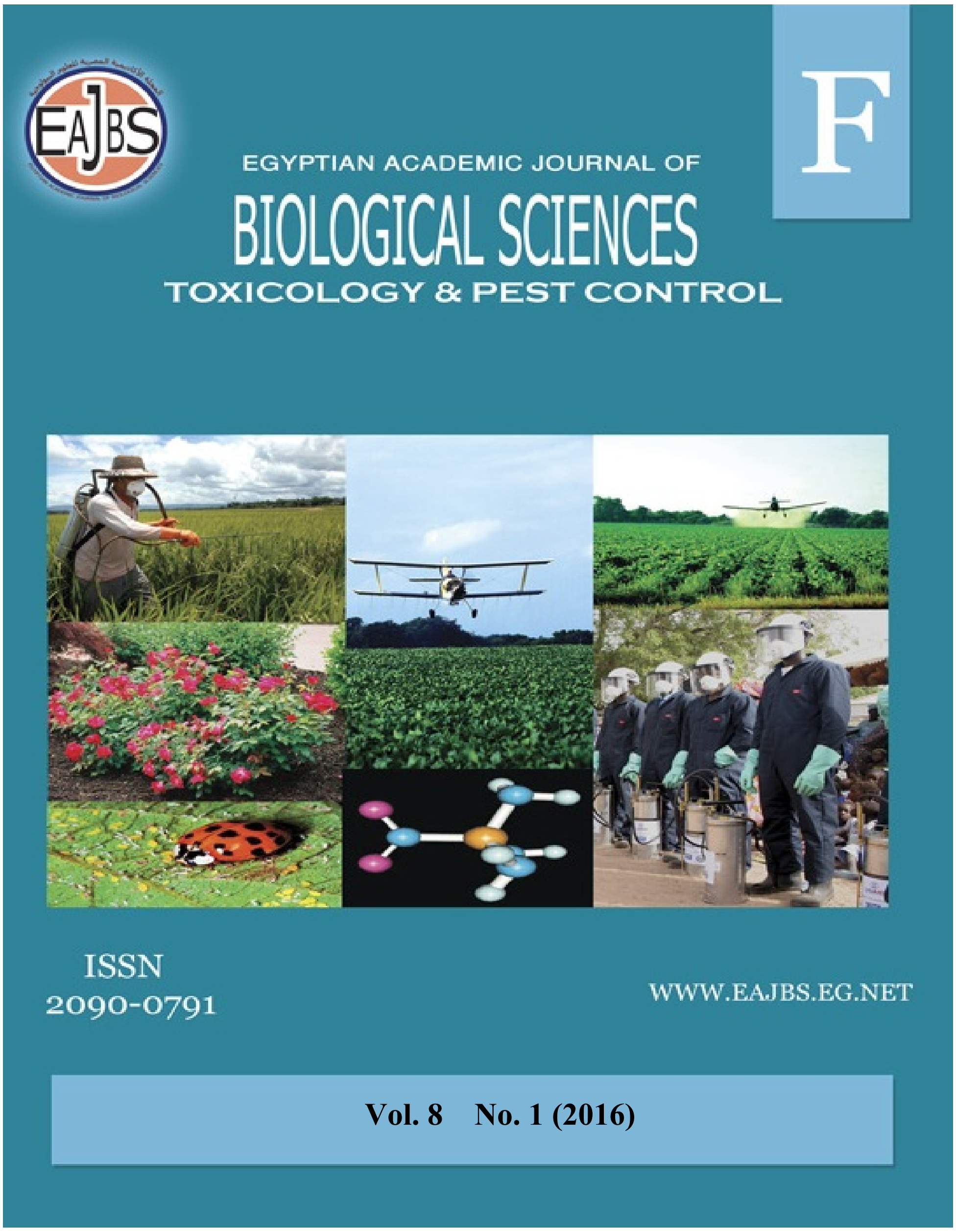

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Nano Silica as A Promising Alternative in Control *Sytophillus oryzae* (L) (Coleoptera: Curculionidae)

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ABSTRACT

Sitophilus oryzae (L.) is a serious primary insect pest of the stored rice, wheat and maize grain. The present study aims to determine the efficiency of the three nano- candidates (hydrophilic, hypophilic and lipophilic silicate). These nano silicate products were tested against the rice weevil *Sitophilus oryzae* under laboratory conditions. Results showed that the number of mortality of *S. oryzae* were 51.0 ± 9.1, 70. 4 ± 9.6 and 73.0 ± 6.2 individuals after investigated with 5 mg/kg-1, the number of mortality scored a higher mortality reached to 99.0 ± 6.7, 89.0 ± 4.2 and 93.00 ± 2.2 with 10 mg/kg^{-1}. In 20 mg/kg^{-1} treatments the number mortality of *S. oryzae* were significantly increased to 100.0 ± 0.0, 100.0 ± 0.0 and 100.0 ± 0.0 14 days post application, as compared to 1.0 ± 2.8, 1.0 ± 5.1 and 0.0 ± 3.1, respectively in the control and 79.0 ± 3.2, 69 ± 2.1 and 98 ± 3.1 with deltamethrin treatments. Number of emerged adults and the original activity remaining OAR% were calculated where it gave with 20 mg/kg^{-1} treatments 95.55, 95.16, 94.59, 74.24, 44.82 and 39.93 OAR% while it gave 94.07, 92.25, 88.54, 42.80, 26.20 and 0.00 OAR% with hydrophobic silicate. With lipophilic silicate the OAR% scored 94.81, 95.16, 92.36, 29.09, 16.55 and 7.29 after 20, 40, 60, 80, 100 and 120 days post treatment in comparison to 95.92, 94.51, 92.01, 60.20, 27.24 and 12.15 with deltamethrin. LC_{50} found to be 160, 220 and 330 mg/kg -1 for hydrophilic, hypophilic and lipophilic silicate; respectively while it is 40 mg/kg -1 in case of Deltamethrin. This investigation lead to open up newer pathway of using nanomaterial-based technology using hydrophilic nano silicate at 20 mg/kg-1 as an efficient candidate to control the rice weevil.

INTRODUCTION

Rice is the most important food crop for more than half of the world's population (Cogburn, 1980). The rice weevil *S. oryzae* (Coleoptera: Curculionidae) is a major pest of stored rice in Egypt, by commerce it has been spread worldwide. Both the adults and larvae feed on whole grains. They attack wheat, corn, oats, rye, barley, sorghum, dried beans and cereal.

It causes extensive losses in the quality and quantity of commercial products (Madrid *et al.,* 1990). It is a primary pest and infesting unbroken grain kernels. Larvae of *S. oryzae* develop within the kernel. *S. oryzae* is becoming resistant to phosphine (Rajendran 1994; 2002; Benhalima *et al.* 2004) Storage and upkeep of agricultural products is very important post harvest activities. Considerable amount of rice grains is being spoiled after harvest due to chemical insecticides compounds (Larsson *et al.,* 1992). However, there are several reasons to search for alternatives to synthetic insecticides: consumer preference for food without insecticide residues, worker safety concerns, resistant insect populations, and deregistration of current synthetic insecticides. Higher plants are a good source of novel insecticides (Prakash & Rao, 1997). Currently, chemical control is the most commonly used strategy against the pests. There are many chemicals that are toxic to stored-grain pests, including insecticides such as organophosphates, pyrethroids and fumigants such as methyl bromide and phosphine (Park *et al.,* 2003; Kljajic and Peric, 2006 and Wadhwa 2009). These chemicals are effective for pest control but have several problems to users (Okonkwo and Okoye, 1996). Nanosilica against different insect species showed up to complete mortality (Debnath *et al.,* 2010). Nano-pesticides and nano-encapsulated pesticides are expected to reduce the volume of application and slow down the fast release kinetics. (Niemeyer and Doz 2001, Leiderer and Dekorsy, 2008). Mode of action occur destruction of the natural water barrier, the waxy layer of the cuticle (Leiderer and Dekorsy, 2008). Nanotechnology gives major impulses to technical innovations in the future (Leiderer and Dekorsy, 2008 Subramanyam and Roesli 2000). The chemistry of silica provides the functionalization opportunity for a variety of surface (Banerjee and Santra, 2009), which can be used to attach the molecules. Tan *et al*. (2004) developed SNPs, functionalized with both streptavidin and antibody. Zhao *et al*. (2004) developed a SNP-based method for the detection of single bacterium.. So these NPs have great potential as DNA or drug delivery agent (Stark 2011; Wang *et al*. 2012). In spite of the versatile application of SNPs, the potential of these NPs in agriculture remains unexplored. (Edibol *et al*. 2003 and Leiderer and Dekorsy, 2008) This investigation focused on the effect of different nano silica as a possible alternative to traditional pesticide to control rice weevil and their effectiveness under stored conditions.

MATERIALS AND METHODS

Insects rearing

S. oryzae was collected from infested rice obtained from Egyptian local market and reared in glass jars under laboratory conditions of 30°C ± 1°C, 75 ± 5% relative humidity (RH) in continuous darkness. The RH was maintained by using saturated solution of sodium chloride (Winston and Bates1960). After the pupal stage the adults less than 24 hrs old were used for the experiments. Hydrophilic, hypophilic and lipophilic silicate are prepared using the method described (Zhang *et al.* 2007).

Recommended pesticide

Deltamethrin (2.5 WP) (40mg/kg seed) and Untreated check (Rajanish and Rohit 2014).

The insecticidal activity of tested nanoparticles

Experiment was designed to test the initial as well as the persistent effect of the tested nanoparticles hydrophilic, hypophilic and lipophilic silicate as cumulative mortality during successive intervals (0, 2, 4, and 7 days).

Bioassay

The bioassay on *S. oryzae* was performed in small plastic screw capped jars. Each jar had a radius of 6 cm and height of 6.5 cm. The caps were perforated to allow aeration. 20 g of rice (IR64) was placed in each jar. Rice in each jar was treated individually with SNPs (hydrophilic, hydrophobic, and lipophilic). Then, the jars were shaken manually for approximately 1 min to achieve equal distribution of nanoparticles on rice (Subramanyam and Roesli 2000). For each dose, there were five replicates. In five additional set no nanoparticle was mixed with rice and this set served as control. The jars were kept for 24 h before 20 unsexed adults of S. oryzae were introduced into each jar. All bioassays were performed at 30°C ± 2°C, 65 ± 5% R. H. Insect mortality was checked after 1, 2, 4, 7, and 14 days. This investigation was carried out at Unit of Virology Department of Entomology and pesticide, Faculty of Agriculture, Cairo University, Egypt.

The persistence of nanoparticles tested during storage

The Original Activity Remaining OAR % was determined to the tested nanoparticles hydrophilic, hypophilic and lipophilic silicate and deltamethrin. Pair of newly emerged beetles, was placed with treated or untreated seeds in glass jars (250 cm^3 capacity) covered with muslin. These jars were kept for certain periods (20,40,60, 80, 100 and 120 days) five jar for each period, after each certain time the beetles were left to lay eggs, and then the newly emerged adults were counted in the treated and untreated jars, and this was our modified technique to Abd El-Aziz and Ismail, 2000 method.

Data analysis

Data were analyzed using ANOVA, and the means were compared by the means-grouping test of Scott and Knott (1974) at $P < 0.5$. Concentration-mortality regressions were calculated according to the method described by Finney (1971). Original Activity Remaining percentages were determined for each tested treatment using both parameters to insure the potential of the material to prolong the persistence as described by Muro and Paul (1985).

RESULTS AND DISCUSSION

Efficacy of SNP as an insecticidal agent was tested on *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae), Moreover, entomotoxicity of SNPs was compared with deltamethrin. 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 (Korunic 1998, Stathers 2004, Majumder *et al*., 2007, Mucha *et al.,* 2008 and Liu and Sun 2010).

The insecticidal activity of tested nanoparticles

Data showing the insecticidal activity illustrated in Tables 1, 2 and 3 Results in Table 1 showed that the number of mortality of *S. oryzae* after investigation with 5mg/kg^{-1}. Where it gave 13.13 ± 5.6, 6.00 ± 7.2, 8.9 ± 3.2 one day post investigation and it gave 24.00 ± 7.7, 36.36 ± 6.3, 51.00 ± 8.4 two days post investigation 36.36 ± 7.7, 55.00 ± 9.4, 64.94 ± 7.4 four days post investigation and 36.36 ± 8.3, 71.71 ± 8.2 and 79.00 ± 6.4 seven days after, with hydrophilic, hydrophobic and lipophilic silicate; respectively. Finally it gave 51.0 ± 9.1, 70. 4 ± 9.6 and 73.0 ± 6.2 two weeks post treatment, the number of mortality scored a higher mortality reached to 72.00 ± 8.4, 11.00 ± 5.7, 11.11 ± 4.2 one day post investigation and it gave 92.07 ± 9.6, 38.61 ± 9.6, 47.00 ± 6.2 two days post investigation 97.00 ± 6.5, 49.00 ± 4.6, 73.73 ± 8.3 four days post investigation and 98.00 ± 4.2, 80.80 ± 8.2 and 88.65 ± 2.3 seven days after, respectively. Finally it gave 99.00 ± 6.7, 89.00 ± 4.2 and 93.00 ± 2.2 with hydrophilic, hydrophobic and lipophilic silicate; respectively. Fourteen days post treatment. In 20 mg/kg^{-1} treatments the number mortality of *S. oryzae* were significantly increased to Where it gave 86.86 ± 7.4, 42.00 ± 7.6, 37.75 ± 8.2 one day post investigation and it gave 88.11 ± 3.5, 76.76 ± 2.7, 46.53 ± 3.2 two days post investigation 93.93 ± 2.7, 80.00 ± 1.2, 69.07 ± 4.2 four days post investigation and 100.00 ± 1.2, 93.93 ± 1.3 and 79.00 ± 2.3 seven days after, respectively. Finally it gave 100.0 ± 0.0, 100.0 ± 0.0 and 100.0 ± 0.0 with hydrophilic, hydrophobic and lipophilic silicate; respectively. 14 days post application, as compared to 1.0 ± 2.8, 1.0 ± 5.1 and 0.0 ± 3.1, respectively in the control and 79.0 ± 3.2, 69 ± 2.1 and 98 ± 3.1 with deltamethrin treatments.

Table 1: Mean mortality (± S.E.) of *S. oryzae* adults exposed for 1, 2, 4, 7, and 14 days on rice treated with three different kind of nano silica at 0.5 g kg-1 with control.

Mortality % per Day	Hydrophilic	Hydrophobic	Lipophilic	Deltamethrin	control
1	13.13 (13/99) ± 5.6 Ba	6.00 (6/100) ±7.2 Aa	8.9 (9/101) ± 3.2 Aa	73.00 (73/100) ± 6.2 Bb	2.00 (2/100) ± 2.7 Aa
2	24.00 (24/100) ± 7.7 Ba	36.00 (36/99) ± 6.3 Ba	51.0 (51/100) ± 8.4 Bb	51.00 (51/100) ± 8.4 Bb	1.00 (1/100) ± 1.3 Aa
4	37.00 (37/100) ± 7.7 Ba	55.00 (55/100) ± 9.4 Bb	64.94 (63/97) ± 7.4 Bb	64.94 (63/97) ± 7.4 Bb	1.00 (1/100) ± 1.3 Aa
7	36.40 (36/99) ± 8.3 Ba	71.70 (71/ 99) ± 8.2 Bb	79.00 (79 / 100) ± 6.4 Bb	60.00 (60 / 100) ± 6.4 Bb	3.00 (3/100) ± 2.3 Aa
14	51.00 (51/100) ± 9.1 Ba	70.40 (69/98) ± 9.6 Bb	73.00 (73/100) ± 6.2 Bb	59.40 (60/101) ± 3.2 Aa	0.00 (0/100) ± 0 Aa

Table 2: Mean mortality (± S.E.) of *S. oryzae* adults exposed for 1, 2, 4, 7, and 14 days on rice treated with three different kind of nano silica at 1 g kg-1 with control.

Mortality % per Day	Hydrophilic	Hydrophobic	Lipophilic	Deltamethrin	control
1	72.00 (72/100) ± 8.4 Ca	11.00 (11/100) ± 5.7 Ab	11.11 (11/99) ± 4.2 Ac	73.00 (73/100) ± 6.2 Bb	2.00 (2/100) ± 2.7 Aa
2	92.07 (93/101) ± 9.6 Cb	38.60 (39/101) ± 9.6 Cb	47.00 (47/100) ± 6.2 Bb	5100 (51/100) ± 8.4 Bb	1.00 (1/100) ± 1.3 Aa
4	97.00 (97/100) ± 6.5 Ca	49.00 (49/100) ± 4.6 Cb	73.73 (73/99) ± 8.3 Bb	64.94 (63/97) ± 7.4 Bb	1.00 (1/100) ± 1.3 Aa
7	98.00 (98/100) ± 4.2 Ca	80.80 (80/ 99) ± 8.2 Ca	88.65 (86/97) ± 2.3 Bb	60.00 (60/100) ± 6.4 Bb	3.00 (3/100) ± 2.3 Aa
14	99.00 (99/100) ± 6.7 Ca	89.00 (89/100) ± 4.2 Ca	91.17 (93/102) ± 2.2 Ca	59.40 (60/101) ± 3.2 Aa	0.00 (0/100) ± 0 Aa

Table 3: Mean mortality (± S.E.) of *S. oryzae* adults exposed for 1, 2, 4, 7, and 14 days on rice treated with three different kind of nano silica at 2 g kg-1 with control.

Mortality % per Day	Hydrophilic	Hydrophobic	Lipophilic	Deltamethrin	Control
1	86.86 (86/99) ± 7.4 Da	42.00 (42/100) ±7.6 Bb	37.75 (37/98)± 8.2 Bb	73.00 (73/100) ± 6.2 Bb	2.00 (2/100) ± 2.7 Aa
2	88.11 (89/101) ± 3.5 Da	76.76 (76/99) ± 2.7 Da	46.53 (47/101) ± 3.2 Ca	51.00 (51/100) ± 8.4 Bb	1.00 (1/100) ± 1.3 Aa
4	93.93 (93/99) ± 2.7 Ca	80.00 (80/100) ± 1.2 Da	69.07 (67/97) ± 4.2 Ca	64.94 (63/97) ± 7.4 Bb	1.00 (1/100) ± 1.3 Aa
7	100.00 (99/99) ± 1.2 Ca	93.93 (93/ 99) ± 1.3 Ca	97.00 (97/100) ± 2.3 Ca	60.00 (60/100) ± 6.4 Bb	3.00 (3/100) ± 2.3 Aa
14	100.00 (100/100) ± 0.0 Ca	100.00 (100/100)0.0 Ca	100.00 (100/100) ± 0.0 Ca	59.40 (60/101) ± 3.2 Aa	0.00 (0/100) ± 0 Aa

The persistence of nanoparticles tested during storage

Number of emerged adults and the original activity remaining OAR% were calculated where it gave with 20 mg/kg^{-1} treatments 95.55, 95.16, 94.59, 74.24, 44.82 and 39.93 OAR% (Table 4) while it gave 94.07, 92.25, 88.54, 42.80, 26.20 and 0.00 OAR% with hydrophobic silicate (Table 5). With lipophilic silicate, as it is demonstrated in Table 6 the OAR% scored 94.81, 95.16, 92.36, 29.09, 16.55 and 7.29 after 20, 40, 60, 80, 100 and 120 days post treatment in comparison to 95.92, 94.51, 92.01, 60.20, 27.24 and 12.15 with deltamethrin. Using silica has been increased in control stored grain products Golob 1997; the micron sized silica has become most popular Mutambuki *et al.,* 2011 as an alternative to the traditional pesticides. This study presents the entomotoxic potential for the most popular nano silica derivates (Hydrophilic, hydrophobic and lipophilic silicate) especially that LC_{50} found to be 160, 220 and 330 mg/kg -1 for hydrophilic, hypophilic and lipophilic silicate; respectively while it is 40 mg/kg -1 in case of Deltamethrin. Besides other investigations showed that silica enhances structural rigidity and plant proliferation (Epstein, 1994) this may be one of the most important reasons for focusing on silica nowadays and previous decade all over the world (Ebeling, 1971 and Hiromi *et al.,* 2011).

Table 4: Effects of hydrophilic silicate at 2 g kg-1 on the *Sitophilus oryzae* number adult newly emerged during storage periods of rice seed compared with deltamethrin.

Interval storage/day	hydrophilic silicate		Deltamethrin		Control Number of emerged adults
	Number of emerged adults	OAR %	Number of emerged adults	OAR %	
20	12	95.55	11	95.92	270
40	15	95.16	17	94.51	310
60	17	94.09	23	92.01	288
80	77	74.24	119	60.20	299
100	160	44.82	211	27.24	290
120	173	39.93	203	12.15	288

Table 5: Effects of hydrophobic silicate at **2 g kg-1** on the *Sitophilus oryzae* number adult newly emerged during storage periods of rice seed compared with deltamethrin

Interval storage/day	hydrophobic silicate		Deltamethrin		Control Number of emerged adults
	Number of emerged adults	OAR %	Number of emerged adults	OAR %	
20	16	94.07	11	95.92	270
40	24	92.25	17	94.51	310
60	33	88.54	23	92.01	288
80	171	42.80	119	60.20	299
100	214	26.20	211	27.24	290
120	311	0.00	203	12.15	288

Table 6: Effects of lipophilic silicate at **2 g kg-1** on the *Sitophilus oryzae* number adult newly emerged during storage periods of rice seed compared with deltamethrin

Interval storage/day	lipophilic silicate		Deltamethrin		Control Number of emerged adults
	Number of emerged adults	OAR %	Number of emerged adults	OAR %	
20	14	94.81	11	95.92	270
40	15	95.16	17	94.51	310
60	22	92.36	23	92.01	288
80	212	29.09	119	60.20	299
100	242	16.55	211	27.24	290
120	267	7.29	203	12.15	288

Mortality percentage found to has ascending relationship with time of exposure and with concentration which agreed with Vanic and Brindhaa, 2013 when they examined silica nano particles against *Corcyra cephalonica* and agreed with Debnath *et al.,* 2010 who evaluated the effect of silica nanoparticles against *sytophilus oryza*. Hydrophilic nanoparticles found to be the most effective for the three investigated nano materials, and it was more effective than malation when concentrations of $1gkg^{-1}$ and 2 gkg^{-1} were applied. This could be due to nano sized silica itself or due to losing water through damage in water barrier that include desiccation. We believe that physical mode of action make using nanocides valuable besides it can be easily removed by conventional milling process (Ebeling, 1971, Debnath *et al*., 2010 and Vanic and Brindhaa, 2013) for, hydrophilic nano silicate will have an excellent potential as seed protecting agent at 2 gkg-1 to control stored grain pest.

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ARABIC SUMMERY

النانو سليكا كبديل محتمل فى مكافحة سوسة الأرز *Sytophillus oryzae* L (Coleoptera: Curculionidae)

حلمى محمد البندارى - ألكسندرا الهلالى *
قسم وقاية النبات – كلية الزراعة- جامعة الفيوم- الفيوم- مصر

*** قسم الحشرات الإقتصادية و المبيدات – كلية الزراعة – جامعة القاهرة**

سوسة الأرز *Sytophillus oryzae* (L) أفة حشرية أولية خطيرة للأرز المخزون، القمح و حبوب الذرة. الدراسة الحالية تهدف لتحديد الكفاءة لثلاث مواد نانوية مختبرة (السليكات المحبة للمياه، الكارهه للمياه و المحبة للدهون). منتجات النانوسليكات السابقة تم إختبارها ضد سوسة الأرز *Sytophillus oryzae* (L) تحت ظروف المعمل. أثبتت النتائج أن نسبة الموت لهذة الحشرات كانت ٥١.٠٠±٩.١، ٧٠.٤± ٩.٦ و ٧٣.٠± ٦.٢ فردا بعد استخدام ٥ ملجم/ كجم$^{-١}$، نسبة الموت سجلت صعودا أعلى حيث وصلت إلى ٩٩.٠±٦.٧ ، ٨٩.٠ ± ٤.٢ و ٩٣.٠٠ ± ٢.٢ مع ١٠ ملجم/ كجم$^{-١}$. فى معاملات ال ٢٠ ملجم/ كجم$^{-١}$ نسبة الموت زادت بشكل ملحوظ لتصل ل ١٠٠.٠٠± ٠.٠، ١٠٠.٠٠± ٠.٠ و ١٠٠.٠٠± ٠.٠ أربعة عشر يوما بعد التطبيق، و تم مقارنة تلك النتائج ب ١.٠ ± ٢.٨، ١.٠ ± ٥.١ و ٠.٠ ± ٣.١ على الترتيب فى الكنترول و ٧٩.٠± ٣.٢، ٦٩± ٢.١ و ٠.٠± ٣.١ مع معاملات الدلتا مثرين . عدد الأفراد الكاملة و الكفاءة الفعلية المتبقية OAR% تم حسابها حيث أعطت مع ٢٠ ملجم/ كجم$^{-١}$ ٩٥.٥٥، ٩٥.١٦، ٩٤.٥٩، ٧٤.٢٤، ٤٤.٨٢ و ٣٩.٩٣ % OAR بينما أعطت ٩٤.٠٧، ٩٢.٢٥، ٨٨.٥٤، ٤٢.٨٠، ٢٦.٢٠ و ٠.٠٠ OAR% مع السليكات النانوية المحبة للماء. مع المحبة للدهون ال OAR% سجلت ٩٤.٨١، ٩٥.١٦، ٩٢.٣٦، ٢٩.٠٩، ١٦.٥٥ و ٧.٢٩ بعد ٢٠، ٤٠، ٦٠،٨٠، ١٠٠ و ١٢٠ يوم بعد المعاملة بالمقارنة مع ٩٥.٩٢، ٩٤.٥١، ٩٢.٠١، ٦٠.٢٠، ٢٧.٢٤ و ١٢.١٥ مع الدلتا مثرين .التركيز النصفى المميت LC_{50} وصل الى ١٦٠، ٢٢٠ و ٣٣٠ ملجم/ كجم$^{-١}$ بالنسبة للنانو سليكات المحبة للماء، الكارهه للماء و المحبة للدهون على الترتيب بينما تصل الى ٤٠ ملجم/ كجم$^{-١}$ فى حالة الدلتامثرين. هذا البحث يشير لفتح اتجاه جديد لاستخدام المواد النانوية باستخدام النانوسليكا المحبة للماء عند ٢٠ ملجم/ كجم$^{-١}$ كمادة كفء لمكافحة سوسة الأرز.