JOURNAL OF ENTOMOLOGICAL SOCIETY OF IRAN
2017, 37(2), 235-247
نامه انجمن حشره‌شناسی ایران ۲۴۷- ۲۳۵ ,(۲)۳۷ ,۱۳۹۶

Synthesis and toxicological evaluation of silica nanoparticles as chlorpyrifos carrier against the beetle pests *Rhyzopertha dominica* and *Tribolium confusum*

Asghar Babamir-Satehi[1], Masumeh Ziaee[1&*] & Ali Ashrafi[2]
1- Department of Plant Protection, Faculty of Agriculture, Shahid Chamran University of Ahvaz, Ahvaz, Iran & 2-Department of Materials Engineering, Isfahan University of Technology, Isfahan, Iran.
* Corresponding author, E-mail: m.ziaee@scu.ac.ir

Abstract
In this study, silica nanoparticles (SNPs) were prepared by sol-gel technique. Chlorpyrifos (40.8% EC) was loaded on the SNPs by immersion loading method. The Specific surface area (S_{BET}) of nanosilica was characterized by BET and recorded 102.24 m^2/g. Loading efficiency of chlorpyrifos-loaded in silica nanoparticles (Ch-SNPs) was measured 86.79% using UV-VIS spectrophotometer. According to FT-IR results, chlorpyrifos properties remained intact after loading on nanosilica because of physical adsorption process of the insecticide in the pores. The residual toxicity of Ch-SNPs was assessed against two stored product insect species, *Rhyzopertha dominica* F. and *Tribolium confusum* Jacquelin du Val. on Petri dish, galvanized steel, mosaic and concrete surfaces. Residual toxicity was evaluated 7, 15, 30, 45 and 60-day post treatment. For each post treatment, the mortality was counted after 6, 24, 48 and 72 h of exposure. The mortality increased with increasing concentration of insecticide and time exposed to each concentration. According to the results, the Ch-SNP was effective against the both pest species, but the toxicity varied depending on the surface material. Ch-SNPs provided long-term protection on petri dishes against the pests, whilst concrete followed by mosaic surfaces with less protection. For instance, *R. dominica* mortality percentage after 24 h exposure to Petri dish, galvanized steel, mosaic and concrete treated with 0.2 mg cm^{-2} Ch-SNP was 100, 82.8, 40 and 1.4%, whereas the mortality was 100, 97.1, 20 and 18.5 % for *T. confusum* at 60-day post-treatment, respectively.
Keywords: Chlorpyrifos, controlled release, insecticidal efficacy, silica nanoparticles, sol-gel technique.

ساخت و ارزیابی سمیت نانوذرات سیلیکا به عنوان حامل کلرپایریفوس برای کنترل سوسک کشیش و شپشه آرد

اصغر بابامیرساطحی[۱]، معصومه ضیائی*[۱] و علی اشرفی[۲]
۱- گروه گیاهپزشکی، دانشکده کشاورزی، دانشگاه شهید چمران اهواز، اهواز، ایران و ۲- گروه مهندسی مواد، دانشگاه صنعتی اصفهان، اصفهان، ایران.
* مسئول مکاتبات، پست الکترونیکی: m.ziaee@scu.ac.ir

چکیده
در این مطالعه، نانوذرات سیلیکا به روش سل-ژل تهیه شد. کلرپایریفوس (۴۰/۸٪ امولسیون) در نانوذرات سیلیکا با روش غوطه‌وری بارگذاری شد. سطح ویژه ذرات (S_{BET}) نانوسیلیکا توسط BET اندازه گیری و ۱۰۲/۲۴ متر مربع بر گرم گزارش شد. کارایی بارگذاری کلروپایریفوس بارگذاری شده با نانوسیلیکا توسط دستگاه اسپکتروفتومتر ماورای بنفش- نور مرئی اندازه‌گیری و ۸۶/۷۹ درصد به‌دست آمد. با توجه به نتایج FT-IR، خواص کلرپایریفوس پس از بارگذاری در نانوذرات به دلیل فرایند جذب فیزیکی حشره‌کش در منافذ، تغییر نکرد. دوام سمیت نانوسیلیکای بارگذاری شده با کلروپایریفوس روی دو گونه از حشرات آفت انباری، سوسک کشیش *Rhyzopertha dominica* F. و شپشه آرد، *Tribolium confusum* Jacquelin du Val. در سطح‌های پتری‌دیش، استیل گالوانیزه، موزاییک و بتن مورد بررسی قرار گرفت. دوام سمیت ۷، ۱۵، ۳۰، ۴۵ و ۶۰ روز بعد از تیمار بررسی شد و در هر فاصله زمانی مربوط به آزمایش دوام، تلفات بعد از گذشت ۶، ۲۴، ۴۸ و ۷۲ ساعت پس از قرار گرفتن حشرات در معرض نانوسیلیکا شمارش شد. تلفات با افزایش غلظت و زمان در معرض قرارگیری در هر غلظت افزایش یافت. با توجه به نتایج به دست آمده کلروپایریفوس

Received: 10 February 2017, *Accepted:* 27 May 2017
Subject Editor: Jafar Mohaghegh-Neyshabouri

بارگذاری شده با نانوسیلیکا علیه هر دو حشره مورد آزمایش موثر بود، اما سمیت آن بسته به نوع مواد سطح متفاوت بود. کلروپایریفوس بارگذاری شده با نانوسیلیکا در پتری دیش باعث حفاظت طولانی مدت علیه سوسک‌های مورد مطالعه شدند؛ در صورتی که مدت زمان حفاظت در بتن و پس از آن در سطح موزاییک کوتاهتر بود. به عنوان مثال، درصد تلفات سوسک کشیش، ٢٤ ساعت پس از در معرض قرارگیری حشرات در سطح‌های پتری‌دیش، استیل گالوانیزه، موزاییک و بتن، ٦٠ روز پس از تیمار با غلظت ٠/٢ میلی گرم بر سانتی‌متر مربع به ترتیب ١٠٠، ٨٢/٨، ٤٠، و ١/٤ درصد بود؛ در صورتی که تلفات برای شپشه آرد ١٠٠، ٩٧/١، ٢٠، و ١٨/٥ درصد بود.

واژگان کلیدی: کلرپایریفوس، رهایش تدریجی، توانایی حشره کشی، نانوذرات سیلیکا، تکنیک سل- زل

دریافت: ١٣٩٥/١٠/٢١، پذیرش: ١٣٩٦/٣/٦.

Introduction

Residual insecticides have been used for protection of storage facilities. They are easily applied, lacking specialized equipment and compatible with international grain trade. They are also cost-effective products that successfully control a wide range of stored product insect pests (Obeng-Ofori, 2010). The application of organophosphate insecticides (pirimiphos-methyl, dimethoate, etc.), pyrethroids (cypermethrin, deltamethrin, b-cyfluthrin, etc.), insect growth regulators (IGRs) (methoprene and pyriproxyfen), and neonicotinoids (thiamethoxam) have been studied for protection of stored products in structures (Andric *et al.*, 2014; Arthur, 2004; Athanassiou *et al.*, 2011; Athanassiou *et al.*, 2015; Ghimire *et al.*, 2016; Velki *et al.*, 2014).

Application of nanopesticides containing inorganic nanoparticles (e.g., silica, titanium dioxide) has been recently tested for controlling pests (Kah and Hofmann, 2014). The large surface area to volume ratio of nanoparticles makes them more toxic against insect pests (Goswami *et al.*, 2010). There have been considerable researches on insecticidal efficacy of silica nanoparticles (Barik *et al.*, 2012; Debnath *et al.*, 2012; Debnath *et al.*, 2011; Rouhani *et al.*, 2012; Sabbour, 2013). The insecticidal mode of action of silica nanoparticles is reported as desiccation and absorption of wax layer of insect cuticle and also cell lysis resulting in insects' death. According to International Agency for the Research of Cancer (IARC), amorphous silica belongs to group 3; and not rated as carcinogen (Barik *et al.*, 2012). Nanopesticides seem to be environmentally friendly (Jiang *et al.*, 2010); however, several factors such as types of nanocarriers used in their formulation, size and release profile of nanoparticles, transport, relocation, and bioavailability of an active ingredient (AI) influence their environmental fate (Kah and Hofmann, 2014).

Porous hollow structure of silica nanoparticles (PHSNs) makes it as a good carrier material for slow and controlled release of active substances such as pesticides (Wang *et al.*, 2014), enhancing pesticides bioavailability and reducing their toxicity to the environment leading to the optimization of synthetic pesticide applications (Liu *et al.*, 2006). PHSNs have been provided as a carrier for the slow release of avermectin by a simple immersion loading method (Li *et al.*, 2006; Wen *et al.*, 2005).

The aims of the current study was to synthesize silica nanoparticles as a carrier to study loading efficiency and residual toxicity of chlorpyrifos-loaded silica nanoparticles (Ch-SNPs), as structural treatment on galvanized steel, mosaic and concrete surfaces for controlling adults of *Rhyzopertha dominica* F. (Coleoptera: Bostrychidae) and *Tribolium confusum* Jacquelin du Val. (Coleoptera: Tenebrionidae).

Materials and methods

Insecticide

Commercial formulation of chlorpyrifos 40.8% EC (Agriphar, Belgium) was applied for the experiments.

Materials

Ethanol (99.9%, Merck), C16TAB (Hexadecyl Trimethyl Ammonium Bromide; Merck KGaA- Germany), ammonium hydroxide (28%, Wako), and TEOS (Tetraethyl orthosilicate; 99.9%, Sigma-Aldrich).

Nano silica preparation

The SNPs were prepared by sol-gel process by the method of Rao *et al.* (2005) with some modifications. At the first step, C16TAB (0.22 g) was added to the ethanol medium (deionized water 133 ml: ethanol 1 ml), as a surfactant, to increases the pore size of nanosilica. Ammonium hydroxide (2 M, 11 ml) was added, as a catalyst, to promote the condensation reaction to sodium acetate (0.034 g) and the solution was added to the ethanol medium and stirred for 10 min with the speed of 200 rpm. Then, TEOS as an organic source of silica, was added dropwisely to the solution and stirring followed for 2.5 h to get a white turbid suspension. The gel suspension was washed in a filter paper with deionized water to remove impurities. All the preparation steps were conducted at room temperature. The gel was dried in an oven set at 70□C for 5 h. The C16TAB and other organic impurities were removed by calcination at 560□C at 1□C per min for 5 h. Silica mesoporous material were prepared and kept in the desiccator until use. Specific surface area (S_{BET}) of SNPs was measured from N_2 adsorption using BET (Brunauer - Emmett - Teller) method by Sorptometer Kelvin 1042 and was obtained 102.24m^2/g.

Characterization of SNPs

Chlorpyrifos loading

Chlorpyrifos loading was performed using immersion loading method described by Wen *et al.* (2005) with some modifications. Chlorpyrifos dissolved in acetone (100gkg^{-1}) and mixed with SNPs at a chlorpyrifos: nanosilica weight ratio of 4:1 and vortexed for 48 h

in a shaker with the speed of 400 rpm at room temperature. The material was washed with deionized water and 30% ethanol solution via *centrifugation* using centrifuge (S 2100 SUV) at 13000 × g, 5° C for 10 min to remove the entrapped chlorpyrifos. The supernatant was stored to determine loading efficiency of chlorpyrifos in SNPs. The powder was then dried in a vacuum oven at 60°C for 6 h and Ch-SNP was prepared.

Loading efficiency of chlorpyrifos was measured using UV-VIS spectrophotometer (UNICO Model 2100 series). Chlorpyrifos was dissolved in distilled water (100 ppm) as a control group. Unloaded SNPs was washed with deionized water and ethanol (30%) via centrifugation and the supernatant was applied as a blank for basic correction. The emission spectrum was collected at the range of 200–400 nm. The absorbance of chlorpyrifos in clear supernatant liquid was determined by UV spectroscopic method. For chlorpyrifos, Makino *et al.* (2009) reported two absorption maxima (229 and 290 nm) within the 210-400 nm range. However, 290 nm was considered for analysis in our study. The process was replicated three times. Loading efficiency was calculated by using following equation (Liu *et al.*, 2005):

$LE = (A-B)/A \times 100$

Where A = the total amount of added chlorpyrifos, and B = the free amount of the chlorpyrifos.

Fourier Transform Infrared Spectroscopy (FT-IR)

FT-IR spectrum of pure SNPs and Ch-SNPs was recorded on the FT-IR (VERTEX 70) spectrometer (Bruker, Germany). About 2 mg of each substance was mixed with KBr powder and ground to approach fine particles. Powder was pressed into a pellet for recording the IR spectrum at the range of 400-4000 cm^{-1}. FT-IR spectrum of chlorpyrifos was obtained from Material Measurement Laboratory of National Institute of Standard and Technology (NIST, 2016).

Insects

Laboratory population of *R. dominica* and *T. confusum* were supplied from a culture in Entomology laboratory-Shahid Chamran University for at least 2 years with no history of exposure to insecticides. *Rhyzopertha dominica* and *T. confusum* were reared on whole wheat (Chamran variety) and wheat flour plus 5% brewer yeast (by weight), respectively. All the species were cultured at 27±1˚C and 65±5% RH and held in continuous darkness. Adults with 7-14 days old and of mixed sexes were used in the experiments.

Insecticidal bioassays

The active ingredient of the Ch-SNPs used in the experiments was 23.6%. The concentrations of 0.01 and 0.2 mg cm^{-2} of Ch-SNP was tested against adults of *R. dominica* and *T. confusum.* According to the active ingredient of Ch-SNP, the used concentrations are

equivalent to 0.00236 and 0.0472 mg (a.i.) cm^{-2}, respectively. Glass petri dishes with an internal radius of 8.8 cm and an area of 62 cm^2 served as the exposure arena with nine replications. The concrete, mosaic and galvanized steel surfaces (30 cm length× 30 cm width) were used in three replications and each surface was treated with different concentrations of Ch-SNP. The dust was distributed uniformly on the surfaces by camel hair brush (No. 2). For petri dishes, 10 adults of each species were introduced in separate dishes and the dishes covered with lids. In the case of other surfaces, three plastic rings (4.5cm diameter and 7.5 cm high) were considered as sub-replicates. Each of the plastic rings was coated with a layer of paraffin on their inner surface to prevent insects crawling up the sides of the plastic rings. Whole wheat (2 g) for *R. dominica* and cracked wheat (Chamran variety) in the case of *T. confusum* was placed on each surface after dusting. Then, 10 adults of each species were placed on each ring. Surfaces treated with unloaded SNPs (at the same concentration) were considered as control. The surfaces were placed in incubator set at 28°C, 65% RH and continuous darkness. Residual toxicity of Ch-SNP was evaluated after 7, 15, 30, 45 and 60-day post treatment. The mortality was recorded after 6, 24, 48 and 72 h of exposure. For each post treatment period, separate series of surfaces were prepared. When no leg or antennal movements were observed, insects considered dead.

Data analysis

There were no mortality in the control groups and no need to correct the mortality data. Mortality percentages were transformed to square root of arcsine to normalize the data, but non-transformed data are presented in the tables. The data was analyzed using the Multivariate Analysis of Variance (MANOVA) Repeated Measures. Tukey-Kramer honestly significant difference (HSD) test was used to determine significant differences between the two tested species at different post exposure times and time exposed to each treatment using SPSS software version 16.0 at P=0.05 (Sokal and Rohlf, 1995; Spss, 2007).

Results

Loading efficiency of chlorpyrifos

The obtained spectra of chlorpyrifos and chlorpyrifos collected from supernatant liquid of Ch-SNPs are presented in Figure 1. The peak absorbance of chlorpyrifos was at 209 nm. Loading efficiency (Mean ± SE) of chlorpyrifos in SNPs was obtained 86.79 ± 0.77%.

Fourier Transform Infrared Spectroscopy (FT-IR)

FT-IR was performed to characterize chemical structures and intermolecular interaction of chlorpyrifos, SNPs and Ch-SNPs. The FT-IR features of chlorpyrifos showed

six peaks at 767, 844 1170, 1432, 1548, and 2990 cm^{-1}. FT-IR spectrum of chlorpyrifos displayed C-Cl with 600–800 cm^{-1} and at 2990 cm^{-1} due to the stretching C-H (Figure 2a). For SNPs, there are peaks at the bands related to Si-O-Si bending at 467 cm^{-1}, Si-O-Si symmetric stretching at 800 cm^{-1} and Si-O-Si asymmetric stretching at 1100 cm^{-1} (Figure 2b). The IR of Ch-SNPs indicated that Ch-SNPs retained most of the major peaks of chlorpyrifos and SNPs. There was a substantial shift for P=S at 1592 cm^{-1} and C–Cl at 763 cm^{-1} attributed to the chlorpyrifos adsorption onto the SNPs' surfaces (Figure 2c).

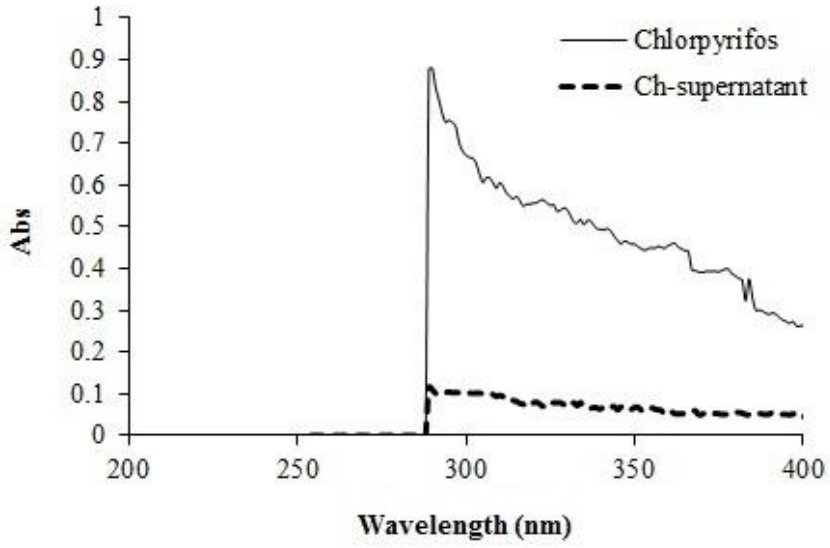

Fig. 1. The spectra of chlorpyrifos and chlorpyrifos collected from supernatant liquid using UV-VIS spectrophotometer. The absorbance of chlorpyrifos was at 290 nm.

Insecticidal bioassays

The mortality of *R. dominica* and *T. confusum* after 6 h exposure at 7-day post treatment on petri dishes was 100% with no difference ($P > 0.05$). At the 15-day post treatment the mortality of *T. confusum* was 100% ($P > 0.05$), even for 6h exposure on petri dishes treated with 0.01 mg cm^{-2}. At the 30, 45 and 60- day experiment, complete mortality was not observed at first hours of exposure to both concentrations of Ch-SNPs, but 100% mortality achieved overtime (Table 1).

The mortality of *R. dominica* and *T. confusum* adults exposed on galvanized steel treated with Ch-SNPs is presented in Table 2. The lower concentration of Ch-SNPs was not very effective after 6 h exposure at all post treatment periods. However, the mortality increased with increasing time of exposure. Results indicated that adults of *T. confusum* were more susceptible to the higher concentration (0.2 mg cm^{-2}) of Ch-SNPs than *R. dominica* and high level of mortality was recorded after an exposure period of only 6 h. However, the toxic effects of Ch-SNPs on *R. dominica* was obtained after a 24 h exposure period during 60-day post treatment (Table 2).

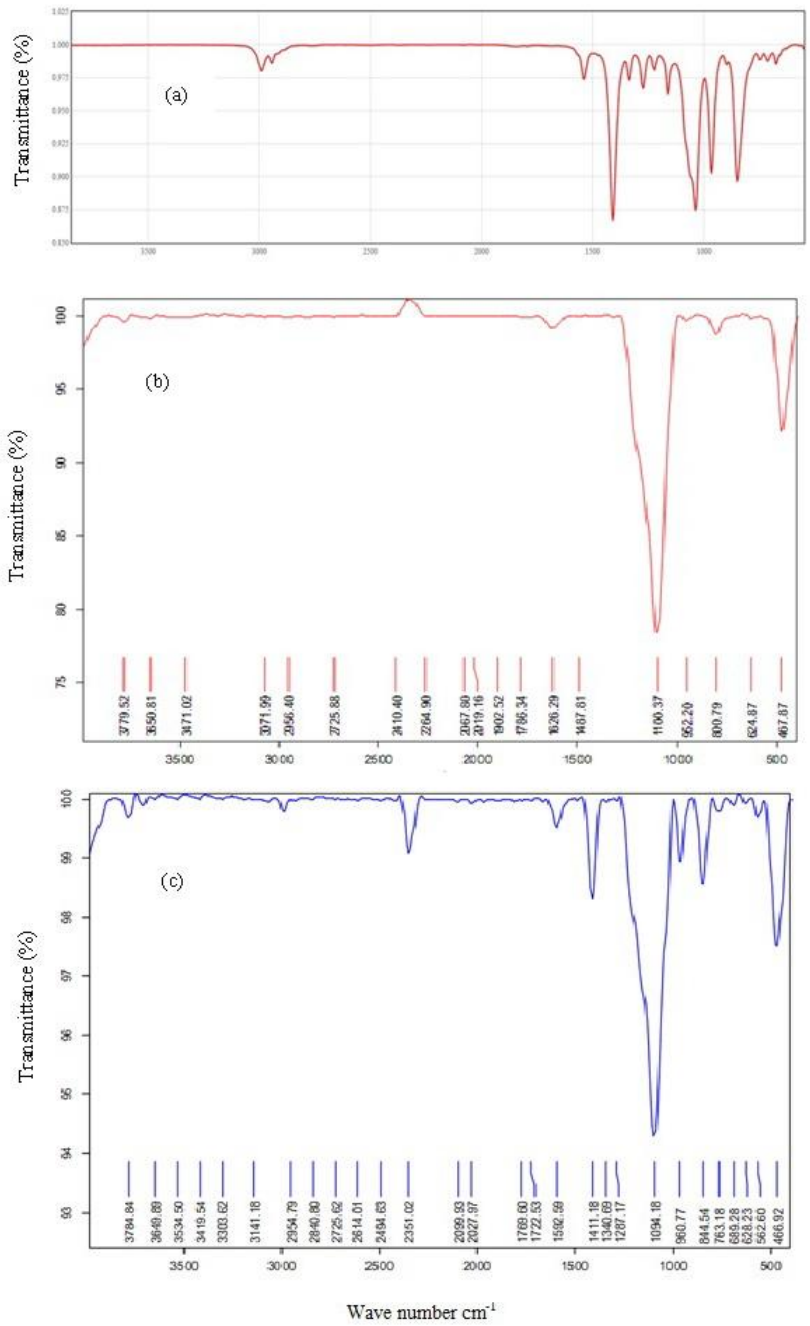

Fig. 2. FT-IR spectra of (a) chlorpyrifos, (b) SNP and (c) Ch-SNP

For *R. dominica* and *T. confusum* exposed to 0.01 mg cm^{-2} Ch-SNPs on mosaic surfaces, the mortality was very low at all post treatment periods. For *R. dominica*, although the Ch-SNPs could not produce 100% mortality at any assessment periods, but the mortality increased with increasing time of exposure. However in the case of *T. confusum*, mortality was very low even after a long period of 72 h. The higher concentration of Ch-SNPs (0.2 mg cm^{-2}) was effective after exposure periods of 24 h (Table 3).

Table 1- Percent mortality (mean ± SE) of *R. dominica* and *T. confusum* on petri dishes treated with Ch-SNPs

			Exposure time (h)				
Species	**Con. (mg cm^{-2})**	**Post-exposure (d)**	**6**	**24**	**48**	**72**	$F_{3,24}$
R. dominica	0.01	7	100±0.0^{a}	100±0.0^{a}	100±0.0^{a}	100±0.0	-
		15	92.8±1.8^{abB}	100±0.0^{aA}	100±0.0^{aA}	100±0.0^{A}	15.0**
		30	71.4±2.6^{cdB}	98.5±1.4^{abA}	100±0.0^{aA}	100±0.0^{A}	89.4**
		45	31.4±2.6^{eC}	57.1±2.8^{cB}	94.2±2.0^{aA}	100±0.0^{A}	219.8**
		60	28.5±4.0^{eD}	42.8±1.8^{dC}	82.8±5.6^{bB}	100±0.0^{A}	86.4**
T. confusum	0.01	7	100±0.0^{a}	100±0.0^{a}	100±0.0^{a}	100±0.0	-
		15	100±0.0^{a}	100±0.0^{a}	100±0.0a	100±0.0	-
		30	80.0±4.3^{bcB}	98.5±1.4^{abA}	100±0.0^{aA}	100±0.0^{A}	18.1**
		45	61.4±5.9^{dB}	88.5±4.6^{bA}	97.1±1.8^{aA}	100±0.0^{A}	20.6**
		60	35.7±3.7^{eC}	54.3±3.7^{cB}	100±0.0^{aA}	100±0.0^{A}	156.6**
$F_{9,60}$			85.2**	105.2**	7.56**	-	
R. dominica	0.2	7	100±0.0^{a}	100±0.0	100±0.0	100±0.0	-
		15	100±0.0^{a}	100±0.0	100±0.0	100±0.0	-
		30	95.7±2.9^{aA}	100±0.0^{A}	100±0.0^{A}	100±0.0^{A}	2.07^{ns}
		45	95.7±2.9^{aA}	100±0.0^{A}	100±0.0^{A}	100±0.0^{A}	2.07^{ns}
		60	95.7±2.9^{aA}	100±0.0^{A}	100±0.0^{A}	100±0.0^{A}	2.07^{ns}
T. confusum	0.2	7	100±0.0^{a}	100±0.0	100±0.0	100±0.0	-
		15	100±0.0^{a}	100±0.0	100±0.0	100±0.0	-
		30	98.5±1.4^{aA}	100±0.0^{A}	100±0.0^{A}	100±0.0^{A}	1.0^{ns}
		45	91.4±3.4^{aB}	100±0.0^{A}	100±0.0^{A}	100±0.0^{A}	6.3**
		60	90.0±3.7^{aB}	100±0.0^{A}	100±0.0^{A}	100±0.0^{A}	7.0**
$F_{9,60}$			2.50^{ns}	-	-	-	

Means followed by the same lower case letter within each column for each concentration and upper case letter in each row are not significantly different using Tukey-Kramer (HSD) test. **: significant at P <0.01, ns: non-significant.

Ch-SNPs at low concentration of 0.01 mg cm^{-2} failed to cause considerable mortality on concrete surface, losing its effectiveness by day 60 for *T. confusum*. The mortality was very low at the higher concentration (0.2 mg cm^{-2}) at short exposure time of 6 h, but increased with increasing time of exposure (Table 4).

Discussion

The FT-IR results indicated that after loading process, SNPs retained its siliceous structure. The peaks indicated that the most chlorpyrifos adsorption in pores of SNPs is probably physical. Wen *et al.* (2005) stated that avermectin properties was unchanged after loading on PHSNs due to physically adsorption of the insecticide. (Liu *et al.*, 2006) prepared PHSNs and loaded with water-soluble pesticide validamycin. The results of FT-IR indicated that all peaks in silica existed in silica-loaded with validamycin fungicide. They concluded that in loading process, the chemical properties of nanoparticles were maintained and validamycin was physically loaded in the nanosilica.

Table 2- Percent mortality (mean ± SE) of *R. dominica* and *T. confusum* on galvanized steel treated with Ch-SNPs

			Exposure time (h)				
Species	**Con. (mg cm^{-2})**	**Post exposure (d)**	**6**	**24**	**48**	**72**	$F_{3,24}$
R. dominica	0.01	7	21.4±2.6^{aC}	65.7±3.7^{aB}	100±0.0^{aA}	100±0.0^{aA}	272.**
		15	14.3±2.0^{bD}	37.1±1.8^{bC}	74.2±3.7^{bB}	88.5±3.4^{abA}	141.5**
		30	8.57±2.6^{bC}	21.4±2.6^{cdBC}	38.5±5.9^{cdAB}	51.4±6.7^{cdeA}	15.1**
		45	1.4±1.4^{cC}	4.3±2.0^{fC}	25.7±3.7^{deB}	44.3±5.3^{deA}	33.9**
		60	0.0±0.0^{cC}	1.4±1.4^{fC}	12.8±1.8^{eB}	35.7±2.8^{eA}	114.5**
T. confusum	0.01	7	0.0±0.0^{cC}	37.1±4.2^{bB}	57.1±7.4^{bcAB}	72.8±6.4^{bcA}	34.4**
		15	0.0±0.0^{cC}	34.3±4.3^{bcB}	65.7±4.8^{bA}	72.8±5.2^{bcA}	64.6**
		30	1.42±1.4^{cB}	18.5±2.6^{deB}	40.0±6.5^{cdA}	51.4±5.08^{cdeA}	25.5**
		45	0.0±0.0^{cC}	5.7±3.7^{efC}	31.4±7.6^{deB}	64.3±7.1^{cdA}	27.6**
		60	0.0±0.0^{cC}	1.42±1.4f^{C}	21.4±5.0^{deB}	42.8±4.2^{deA}	35.6**
$F_{9,60}$			26.46**	49.6**	27.0**	17.5**	
R. dominica	0.2	7	38.6±2.6^{cB}	100±0.0^{aA}	100±0.0^{aA}	100±0.0^{A}	554.0**
		15	38.6±2.6^{cB}	100±0.0^{aA}	100±0.0^{aA}	100±0.0^{A}	554.0**
		30	30.0±2.1^{cdB}	98.5±1.4^{aA}	100±0.0^{aA}	100±0.0^{A}	710.0**
		45	34.2±3.6^{cdB}	97.1±1.8^{aA}	100±0.0^{aA}	100±0.0^{A}	247.7**
		60	24.3±2.02^{dC}	82.8±1.8^{bB}	97.1±1.8^{bA}	100±0.0^{A}	458.7**
T. confusum	0.2	7	97.1±1.8^{aA}	100±0.0^{aA}	100±0.0^{aA}	100±0.0^{A}	2.4^{ns}
		15	97.1±1.8^{aA}	100±0.0^{aA}	100±0.0^{aA}	100±0.0^{A}	2.4^{ns}
		30	97.1±1.8^{aA}	100±0.0^{aA}	100±0.0^{aA}	100±0.0^{A}	2.4^{ns}
		45	90.0±2.1^{aB}	98.5±1.4^{aA}	100±0.0^{aA}	100±0.0^{A}	13.6**
		60	68.5±3.4^{bB}	97.1±1.8^{aA}	100±0.0^{aA}	100±0.0^{A}	62.5**
$F_{9,60}$			156.9**	19.3**	2.40**	-	

Means followed by the same lower case letter within each column for each concentration and upper case letter in each row are not significantly different using Tukey-Kramer (HSD) test. **: significant at P <0.01, ns: non-significant.

In the study by Rumbos *et al.* (2016), *Sitophilus oryzae* (L.), *T. confusum* and *R. dominica* were exposed to wheat treated with pirimiphos-methyl. Adults of *S. oryzae* followed by *T. confusum* were more susceptible to pirimiphos-methyl and *R. dominica* was the most resistant species. In that study, the authors attributed high susceptibility of *T. confusum* to the unfitness of wheat kernels as a food source that reduces recovery and survival rates of this species. In addition, immature stages of *R. dominica* develop inside grain kernels and are not exposed to the insecticides. In the current study, adults of *T. confusum* showed more vulnerability than *R. dominica.* Another possible explanation can be the larger body surface area of *T. confusum* which increases the contact surface of the insecticide particles resulting in high level of mortality.

Our results indicated that the toxicity of Ch-SNPs was as follows: petridishes > galvanized steel > mosaic> concrete. Nayak *et al.* (2003) investigated structure treatment of concrete and galvanized steel surfaces with organophosphorus insecticides including azamethiphos, chlorpyrifos-methyl, fenitrothion and pirimiphos-methyl against three psocid species. They concluded that mortality was lower on concrete than galvanized steel. The low levels of mortality on concrete and mosaic surfaces could attribute to their structure; the surfaces with higher pores absorb more insecticide particles and lower the insecticidal potency.

Table 3- Percent mortality (mean ± SE) of *R. dominica* and *T. confusum* on mosaic treated with Ch-SNPs

			Exposure time (h)				
Species	**Con. (mg cm^{-2})**	**Post exposure (d)**	**6**	**24**	**48**	**72**	***F*** $_{3,24}$
R. dominica	0.01	7	0.0±0.0^{aC}	31.4±3.4^{aB}	74.2±4.2^{aA}	84.2±3.7^{aA}	140.4**
		15	1.4±1.4^{aC}	25.7±2.9^{aB}	54.2±4.3^{bA}	62.8±4.7^{bA}	60.7**
		30	1.4±1.4^{aC}	11.4±2.6^{bB}	41.4±4.5^{bcA}	48.5±3.4^{bcA}	50.2**
		45	0.0±0.0^{aC}	4.2±2.9^{bcC}	34.2±2.0^{cdB}	51.4±4.6^{bcA}	71.1**
		60	0.0±0.0^{aC}	1.4±1.4^{bcC}	21.4±2.6^{deB}	35.7±2.9^{cdA}	66.3**
T. confusum	0.01	7	2.85±1.8^{aB}	4.3±2.9^{bcB}	12.8±3.6^{efAB}	22.8±1.8^{defA}	11.9**
		15	0.0±0.0^{aB}	1.4±1.4^{bcAB}	2.8±1.8^{fAB}	8.5±3.4^{fA}	3.32**
		30	0.0±0.0^{aA}	2.85±1.8^{bcA}	5.7±2.9^{fA}	8.5±2.6^{fA}	2.8**
		45	0.0±0.0^{aB}	1.4±1.4^{bcB}	5.7±2.0^{fB}	27.1±4.2^{deA}	26.7**
		60	0.0±0.0^{aB}	0.0±0.0^{cB}	2.8±1.8^{fB}	15.7±2.0^{efA}	30.0**
$F_{9,60}$			1.33^{ns}	22.87**	60.55**	51.50**	
R. dominica	0.2	7	14.3±2.02^{bC}	88.5±2.6^{aB}	100±0.0^{aA}	100±0.0^{aA}	627.0**
		15	12.8±1.8^{bcC}	68.5±5.5^{bB}	100±0.0^{aA}	100±0.0^{aA}	198.6**
		30	0.0±0.0^{dC}	70.0±6.1^{bB}	98.5±1.4^{aA}	100±0.0^{aA}	218.7**
		45	0.0±0.0^{dD}	48.5±4.0^{cC}	88.5±1.4^{bB}	100±0.0^{aA}	446.0**
		60	0.0±0.0^{dD}	40.0±3.0^{cC}	75.7±2.9^{cB}	100±0.0^{aA}	413.7**
T. confusum	0.2	7	58.5±5.5^{aB}	97.1±2.8^{aA}	100±0.0^{aA}	100±0.0^{aA}	42.4**
		15	61.4±4.5^{aB}	100±0.0^{aA}	100±0.0^{aA}	100±0.0^{aA}	70.5**
		30	2.8±1.8b^{cdC}	48.5±5.0^{cB}	97.1±1.8^{aA}	100±0.0^{aA}	259.6**
		45	1.4±1.4^{cdC}	35.7±2.9^{cdB}	94.3±2.02^{abA}	100±0.0^{aA}	603.8**
		60	0.0±0.0^{dD}	20.0±3.8^{dC}	75.7±3.7^{cB}	98.5±1.4^{aA}	285.6**
$F_{9,60}$			90.9**	47.6**	28.1**	1.0^{ns}	

Means followed by the same lower case letter within each column for each concentration and upper case letter in each row are not significantly different using Tukey-Kramer (HSD) test. **: significant at P <0.01, ns: non-significant.

Song *et al.* (2012) provided silica nanoparticles and loaded with chlorfenapyr to increase the insecticidal toxicity of chlorfenapyr-loaded nanoparticles. In our study, Ch-SNPs were effective against both tested species even at the lowest rate of 0.01 mg cm^{-2}. Wen *et al.* (2005) stated that PHSN was effective for controlled release of avermectin and can be recommended as a carrier of pesticides. Wang *et al.* (2014) also designed a novel hydrophilic delivery system through loading Abamectin with porous silica nanoparticles (Abam-PSNs). Abam-PSNs improve the chemical stability, dispersity, and the controlled release of Abamectin.

Silica nanoparticles can serve as effective tools in pesticide delivery systems as they improve stability and controlled release of pesticides and decrease the recommended application rate.

Table 4- Percent mortality (mean ± SE) of *R. dominica* and *T. confusum* on concrete treated with Ch-SNPs

Species	Con. (mg cm^{-2})	Post exposure (d)	Exposure time (h) 6	24	48	72	$F_{3,24}$
R. dominica	0.01	7	0.0±0.0^{bC}	25.7±2.9^{bcaB}	61.4±5.0^{aA}	71.4±3.4^{aA}	93.6**
		15	0.0±0.0^{bC}	8.57±2.6^{bcB}	15.7±2.0^{bA}	18.5±1.4^{bA}	21.2**
		30	0.0±0.0^{bB}	1.42±1.4^{bcB}	11.4±2.6^{bcdA}	17.1±2.8^{bcA}	15.8**
		45	0.0±0.0^{bB}	0.0±0.0^{cB}	5.71±2.0^{bcAB}	10.0±2.1^{bcdeA}	10.6**
		60	0.0±0.0^{bB}	0.0±0.0^{cB}	0.0±0.0^{dB}	5.71±2.0^{cdeA}	8.0**
T. confusum	0.01	7	2.8±1.8^{aB}	10.0±3.0^{bAB}	12.8±1.8^{bcA}	14.2±2.0^{bcdA}	5.0**
		15	0.0±0.0^{bA}	2.8±2.8^{bcA}	2.8±2.8^{cdA}	4.3±4.3^{deA}	0.37^{ns}
		30	0.0±0.0^{bA}	2.8±2.8^{bcA}	4.3±4.3^{bcdA}	5.7±4.3^{cdeA}	0.67^{ns}
		45	0.0±0.0^{bA}	0.0±0.0^{cA}	0.0±0.0^{dA}	1.4±1.4^{eA}	1.0^{ns}
		60	0.0±0.0^{bA}	0.0±0.0^{cA}	0.0±0.0^{dA}	0.0±0.0^{eA}	-
$F_{9,60}$			2.4**	15.0**	55.7**	59.3**	
R. dominica	0.2	7	21.4±2.6^{cC}	68.5±4.0^{bB}	91.4±2.6^{aA}	100±0.0^{aA}	165.7**
		15	12.8±2.8^{cdD}	25.7±2.0^{cdC}	57.1±1.8^{cB}	91.4±3.4^{abA}	181.0**
		30	2.8±1.8^{dD}	14.2±2.0^{deC}	71.4±2.6^{bB}	80.0±2.1^{bcd}	322.7**
		45	0.0±0.0^{dB}	8.5±1.4^{efB}	71.4±3.4^{bA}	77.1±3.5^{cde}	248.9**
		60	0.0±0.0^{dC}	1.4±1.4^{fC}	48.5±3.4^{cdB}	67.1±2.8^{eA}	210.6**
T. confusum	0.2	7	41.4±2.6^{bB}	95.7±2.9^{aA}	100±0.0^{aA}	100±0.0^{aA}	209.7**
		15	58.5±7.3^{aB}	97.1±1.8^{aA}	100±0.0^{aA}	100±0.0aA	28.4**
		30	0.0±0.0^{dD}	30.0±3.0^{cC}	41.4±3.4^{deB}	85.7±2.9^{bcA}	168.8**
		45	0.0±0.0^{dC}	28.6±3.4^{cB}	32.8±2.8^{eB}	75.7±3.7^{cdeA}	117.0**
		60	0.0±0.0^{dD}	18.5±2.6^{cdeC}	35.7±2.9^{eB}	68.5±2.6^{deA}	151.3**
$F_{9,60}$			54.4**	181.1**	97.1**	24.6**	

Means followed by the same lower case letter within each column for each concentration and upper case letter in each row are not significantly different using Tukey-Kramer (HSD) test at $P = 0.05$.

Acknowledgments

The authors are grateful to Shahid Chamran University for its financial and logistic support during the course of this project.

References

Andric, G., Kljajic, P. & Prazic-Golic, M. (2014) Residual efficacy of cypermethrin and pirimiphos-methyl against *Sitophilus granarius* (L.) and *Plodia interpunctella* (Hübner) on concrete surface. *Pesticidi i fitomedicina* 29, 275-281.

Arthur, F. H. (2004) Evaluation of methoprene alone and in combination with diatomaceous earth to control *Rhyzopertha dominica* (Coleoptera: Bostrichidae) on stored wheat. *Journal of Stored Products Research* 40, 485-498.

Athanassiou, C. G., Arthur, F. H. & Throne, J. E. (2011) Efficacy of layer treatment with methoprene for control of *Rhyzopertha dominica* (Coleoptera: Bostrychidae) on wheat, rice and maize. *Pest Managment Science* 67, 380-384.

Athanassiou, C. G., Kavallieratos, N. G., Boukouvala, M. C., Mavroforos, M. E. & Kontodimas, D. C. (2015) Efficacy of alpha-cypermethrin and thiamethoxam against *Trogoderma granarium* Everts (Coleoptera: Dermestidae) and *Tenebrio molitor* L. (Coleoptera: Tenebrionidae) on concrete. *Journal of Stored Products Research* 62, 101-107.

Barik, T., Kamaraju, R. & Gowswami, A. (2012) Silica nanoparticle: a potential new insecticide for mosquito vector control. *Parasitology Research* 111, 1075-1083.

Debnath, N., Das, S., Seth, D., Chandra, R., Bhattacharya, S. & Goswami, A. (2011) Entomotoxic effect of silica nanoparticles against *Sitophilus oryzae* (L.). *Journal of Pest Science* 84, 99-105.

Debnath, N., Das, S., Patra, P., Mitra, S. & Goswami, A. (2012) Toxicological evaluation of entomotoxic silica nanoparticle. *Toxicological & Environmental Chemistry* 94, 944-951.

Ghimire, M. N., Arthur, F. H., Myers, S. W. & Phillips, T. W. (2016) Residual efficacy of deltamethrin and β-cyfluthrin against *Trogoderma* variabile and *Trogoderma inclusum* (Coleoptera: Dermestidae). *Journal of Stored Products Research* 66, 6-11.

Goswami, A., Roy, I., Sengupta, S. & Debnath, N. (2010) Novel applications of solid and liquid formulations of nanoparticles against insect pests and pathogens. *Thin Solid Films* 519, 1252-1257.

Jiang, L., Zheng, F., Leng, P. F., Xu, M. & Zhao, M. J. (2010) Advances in research of nano-pesticide. *Guangdong Agricultural Science* 5, 97-100. [In Chinese with English summary].

Kah, M. & Hofmann, T. (2014) Nanopesticide research: Current trends and future priorities. *Environment International* 63, 224-235.

Li, Z. Z., Xu, S. A., Wen, L. X., Liu, F., Liu, A. Q., Wang, Q., Sun, H. Y., Yu, W. & Chen, J.F. (2006) Controlled release of avermectin from porous hollow silica nanoparticles: Influence of shell thickness on loading efficiency, UV-shielding property and release. *Journal of Controlled Release* 111, 81-88.

Liu, C. G., Desai, K. G. H., Chen, X. G. & Park, H. J. (2005) Linolenic acid-modified chitosan for formation of self-assembled nanoparticles. *Journal of Agricultural and Food Chemistry* 53, 437-441.

Liu, F., Wen, L.-X., Li, Z.-Z., Yu, W., Sun, H.-Y. & Chen, J.-F. (2006) Porous hollow silica nanoparticles as controlled delivery system for water-soluble pesticide. *Materials Research Bulletin* 41, 2268-2275.

Makino, Y., Oshita, S., Murayama, Y., Mori, M., Kawagoe, Y. & Sakai, K. (2009) Nondestructive analysis of chlorpyrifos on apple skin using UV reflectance. *Transactions of the ASABE* 52, 1955-1960.

Nayak, M. K., Collins, P. J. & Kopittke, R. A. (2003) Residual toxicities and persistence of organophosphorus insecticides mixed with carbaryl as structural treatments against three liposcelidid psocid species (Psocoptera: Liposcelididae) infesting stored grain. *Journal of Stored Products Research* 39, 343-353.

NIST, (2016) Chlorpyrifos [WWW document]. Available in: http://webbook.nist.gov/cgi/cbook.cgi?ID=C2921882&Mask=200.

Obeng-Ofori, D., (2010) Residual insecticides, inert dusts and botanicals for the protection of durable stored products against pest infestation in developing countries. In: Proceedings of the Tenth International Working Conference on Stored Product Protection. Ed. by M O Carvalho, P G Fields, C S Adler, F H Arthur, C G Athanassiou, J F Campbell, F Fleurat-Lessard, P W Flinn, R J Hodges, A A Isikber, S Navarro, R T Noyes, J Riudavets, K K Sinha, G R Thorpe, B H Timlick, P Trematerra & N D G White, Julius-Kühn-Archiv, Estoril, Portugal, 774-788.

Rao, K. S., El-Hami, K., Kodaki, T., Matsushige, K. & Makino, K. (2005) A novel method for synthesis of silica nanoparticles. *Journal of Colloid and Interface Science* 289, 125-131.

Rouhani, M., Samih, M. & Kalantari, S. (2012) Insecticidal effect of silica and silver nanoparticles on the cowpea seed beetle, *Callosobruchus maculatus* F.(Col.: Bruchidae). *Journal of Entomological Research* 4, 297-305.

Rumbos, C. I., Dutton, A. C. & Athanassiou, C. G. (2016) Insecticidal efficacy of two pirimiphos-methyl formulations for the control of three stored-product beetle species: Effect of commodity. *Crop Protection* 80, 94-100.

Sabbour, M. (2013) Entomotoxicity assay of nanoparticle 4-(silica gel Cab-O-Sil-750, silica gel Cab-O-Sil-500) against *Sitophilus oryzae* under laboratory and store conditions in Egypt. *Scientific Research Reports* 1, 67-74.

Sokal, R. R. & Rohlf, F. J., (1995) Biometry: the principles and practice of statistics in biological research. Third edition. 887 pp.

Song, M.-R., Cui, S. M., Gao, F., Liu, Y. R., Fan, C. L., Lei, T. Q. & Liu, D. C. (2012) Dispersible silica nanoparticles as carrier for enhanced bioactivity of chlorfenapyr. *Journal of Pesticide Science* 37, 258-260.

SPSS, (2007) SPSS 16 for Windows User's Guide Release, Spss Inc, Chicago

Velki, M., Plavšin, I., Dragojević, J. & Hackenberger, B. K. (2014) Toxicity and repellency of dimethoate, pirimiphos-methyl and deltamethrin against *Tribolium castaneum* (Herbst) using different exposure methods. *Journal of Stored Products Research* 59, 36-41.

Wang, Y., Cui, H., Sun, C., Zhao, X. & Cui, B. (2014) Construction and evaluation of controlled-release delivery system of Abamectin using porous silica nanoparticles as carriers. *Nanoscale Research Letters* 9, 655.

Wen, L. X., Li, Z. Z., Zou, H. K., Liu, A. Q. & Chen, J. F. (2005) Controlled release of avermectin from porous hollow silica nanoparticles. *Pest Management Science* 61, 583-590.