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EFFICACY OF SILICA NANOPARTICLES ON GROUNDNUT BRUCHID, *CARYEDON SERRATUS* (OLIVIER) (COLEOPTERA, BRUCHIDAE)

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ABSTRACT

In Senegal, post-harvest losses caused by groundnut bruchid, *Caryedon serratus*, are detrimental to farmers and affect national economy. The objectivie of this study was to investigate the effectiveness of silica nanoparticles against *C. serratus*. Silica nanoparticles were synthesized by reverse microemulsion and different concentrations (0.17, 0.33, 0.67 and 1.7 mg kg^{-1}) were tested on mortality and fecundity of *C. serratus*. Result showed that silica nanoparticles had high toxicity on *C. serratus* adults. Adults' mortality increased with concentrations and time of exposure to each concentration. Up to 100% mortality was observed with high concentrations (0.67 and 1.7 mg kg^{-1}) after 6 and 7 days post exposure, respectively. Additionally, fecundity potential of *C. serratus* was significantly ($P<0.05$) reduced by silica nanoparticles. Results of this study clearly demonstrated that silica nanoparticles can be used as a valuable tool for the control of *C. serratus* in stored groundnut (*Arachis hypogaea* L.) .

Key Words: Pests, post-harvest, reverse microemulsion

RÉSUMÉ

Au Sénégal, les pertes post-récolte causées par la bruche de l'arachide, *Caryedon serratus*, sont préjudiciables aux agriculteurs et affectent l'économie nationale. L'objectif de cette étude était d'étudier l'efficacité des nanoparticules de silice contre *C. serratus*. Des nanoparticules de silice ont été synthétisées par microémulsion inverse et différentes concentrations (0,17 ; 0,33 ; 0,67 et 1,7 mg kg^{-1}) ont été testées sur la mortalité et la fécondité de *C. serratus*. Les résultats ont montré que les nanoparticules de silice présentaient une toxicité élevée chez les adultes de *C. serratus*. La mortalité des adultes augmentait avec les concentrations et le temps d'exposition à chaque concentration. Une mortalité allant jusqu' à 100% a été observée avec les plus grandes concentrations (0,67 et 1,7 mg kg^{-1}) respectivement après 6 et 7 jours d'exposition. De plus, le potentiel de fécondité de *C. serratus* était significativement (P <0,05) réduit par les nanoparticules de silice. Les résultats de cette étude ont clairement démontré que les nanoparticules de silice peuvent être utilisées comme un outil précieux pour lutter contre *C. serratus* dans les arachides stockées (*Arachis hypogaea* L.).

Mots Clés : Organismes nuisibles, post-récolte, microémulsion inverse

INTRODUCTION

The food situation in sub-Saharan Africa is characterised by high post-harvest losses due largely to insect pests, in the field and in storage sites (Cissokho *et al.*, 2015). A number of bruchids are well known to cause damage on legume seeds in storage. Among those insects, are beetles which belong to bruchidae family: *Caryedon serratus* (Olivier), more commonly called "the groundnut bruchid" (Delobel *et al.*, 1995). In the wild, this bruchid can develop on several host plants of the family of Caesalpiniaceae, belonging mainly to genera *Bauhinia*, *Cassia*, *Piliostigma* and *Tamarindus* (Sembène *et al.*, 2012a).

In Senegal, attack of groundnut (*Arachis hypogaea* L.) crops by *Caryedon serratus* is a major national problem. Groundnut occupies an important place in the country's economic system and generates about US$138 million every year, which represents 40% of the global exportat of the country (Sembène *et al.*, 2012b). It is a legume that is rich in protein (25% of its composition) and fat (50% of its composition); and is a very important nutrient source for local populations (Schilling *et al.,* 1996). Losses caused by the bruchid on groundnut do not only harm the farmers; but also affect the national economy. These can go up to 83% of quantitative loss for a storage period of 4 months (Thiaw *et al.*, 2007; Thiaw and Sembène, 2010). Thus, special attention should be paid to the management of storage areas in order to limit post-harvest losses.

Although chemicals have been used to control insect pests (Gueye *et al.*, 2011), an alternative that does not create a health problem or harm to consumers and the environment, must be found. Thus, nanotechnology holds great promise for protection of crops and foodstuffs (Stadler *et al.*, 2010). Researchers believe that nanotechnologies will revolutionise agriculture, including pest management in the near future (Bhattacharyya *et al.*, 2010). Nanoparticles represent a new generation of technology that could bring an economic and environmental solution (Ali *et al.*, 2014).

Silica nanoparticles have drawn attention in scientific researches, because they are easily prepared and due to their great range of industrial products and also their application to biology (Debnath *et al.*, 2012). However, few studies have been done on the toxicity of nanoparticles on insects. Nevertheless, the insecticidal activity of silica nanoparticles has been successfully tested on the pests such as *Callosobruchus maculatus* (F.) and *Tribolum castaeneum* (Herbst) (Yang *et al.*, 2009; Rouhani *et al.*, 2012). The objective of this study, therefore was to evaluate the biocidal effect of silica nanoparticles for the prevention of postharvest groundnut losses caused by *Caryedon serratus*.

MATERIALS AND METHODS

Synthesis of silica nanoparticles. Components of the reverse microemulsion, Triton X-100 (1.77 ml), hexanol (1.8 ml), cyclohexane (7.5 ml) and water (0.48 ml), were introduced at room temperature, while stirring in a 30 ml bottle. Then, tetraethylorthosilicate (0.10 ml), an organic source of silica, was introduced using a micropipette. After stirring for 30 minutes, ammonium hydroxide (0.06 ml) was added to the bottle. The bottle mixtures were left to stir on magnetic agitator overnight. Nanoparticles were separated from the reaction medium by addition of equal volume of ethanol, prior to centrifugation. With the supernatant removed, the nanoparticles were washed four times with ethanol, before being left to dry outdoors. To increase porosity of silica, nanoparticles were calcined at 200 ° C in an oven overnight.

Insect and bioassay. The effectiveness of silica nanoparticles was evaluated against *C. serratus,* using 30 g of groundnuts seeds were poured into glass vials. The seeds were treated with 0.17, 0.33, 0.67 and 1.7 mg kg^{-1} of silica nanoparticles, in three replications, and a control group (without nanoparticles). The vials were shaken for 5 min to achieve equal distribution of the nanoparticles over the entire seed mass. Then, 5 couples of *C. serratus* were added into each vial. The vials were covered with muslin cloth for sufficient ventilation, and placed on a bench at room temperature. Mortality and fecundity of *C. serratus* were counted after 1, 2, 3, 4, 5, 6 and 7 days of exposure. Insects were considered dead when no leg or antenna movements were observed after prodding with a fine brush.

Fecundity assessment was done by counting hatched eggs and unhatched eggs, using a binocular loupe.

Statistical tests. Data were analysed using analysis of variance (ANOVA) using SPSS software Version 16, and means were separated using Tukey-Kramer (HSD) test at $P < 0.05$, using SPSS software Version 16.

RESULTS

Silica nanoparticles. Shape and size of the nanoparticles prepared in this study were checked by Transmission Electron Microscopy (TEM). Figure 1 indicates that morphology of silica nanoparticles was spherical, with the average diameter of 30 nm.

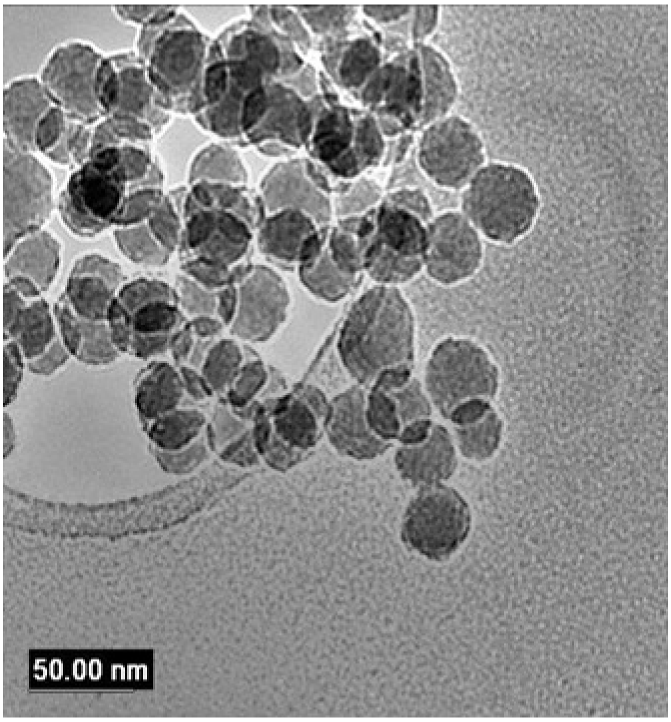

Figure 1. Transmission electron microscopy microscopic images of silica nanoparticles used as a pesticide in the study with the groundnut bruchid.

Adulticidal activity of silica nanoparticles. The results of adulticidal activity of silica nanoparticles against adult of *C. serratus* are presented in Table 1. Data clearly showed that the mortality of *C. serratus* adults increased with increasing concentrations and time exposed to each concentration. Twenty-four hours after introduction of adults, the mortality was low, but rose to 100% after 6 and 7 days of exposure to 0.67 and 1.7 mg kg^{-1} of silica nanoparticles, respectively. There were significant (P<0.05) differences between silica concentrations.

Effect on the fecundity. The effect of treatment of silica nanoparticles with the groundnuts seeds on *Caryedon serratus* adults is presented in Table 2. It was revealed that the fecundity potential of *C. serratus* was significantly (P<0.05) reduced by silica nanoparticles. Fecundity decreased as concentrations and time after treatment

TABLE 1. Mortality (%±SE) of *Caryedon serratus* adults, exposed on groundnut seed treated with silica nanoparticles

Exposure time (d)	Concentration (mg kg^{-1})				
	0	0.17	0.33	0.67	1.7
1	0^{a} ±0	0^{a} ±0	13^{a} ±4.8	30^{a} ±6.4	30^{a}±6.8
2	13^{a} ±4.5	7^{ab} ±3.4	37^{abc} ±5.7	57^{bc} ±5.5	70^{c} ±6.2
3	13^{a} ±4.5	17^{ab} ±5.2	70^{abc} ±6.2	73^{bc}±5.3	87^{c} ±4.7
4	16^{a} ±5.7	17^{a} ±5.2	77^{b} ±4.2	77^{b}±4.2	90^{b} ±5.3
5	20^{a} ±6.3	20^{a} ±6.3	80^{b}±3.7	80^{b}±3.7	100^{b}±0
6	23^{a} ±4.6	27^{a} ±5.4	83^{b}±3.2	90^{b}±5.3	100^{b}±0
7	27^{a} ±5.4	30^{a} ±3.7	83^{b} ±3.2	100^{b} ±00	100^{b}±0

Means followed by the same lowercase letter in each row are not significantly different using Turkey's test at P < 0.05

TABLE 2. Fecundity (±SE) of *Caryedon serratus* adults, exposed on groundnut seed treated with silica nanoparticles

Exposure time (d)	Concentration (mg kg^{-1})				
	0	0.17	0.33	0.67	1.7
1	35^{a} ± 5.2	25^{a} ± 5.3	10^{a} ± 2.5	13^{a} ± 3.2	8^{a} ± 2.1
2	16^{a} ± 3.8	11^{a} ± 2.3	5^{a} ± 2.4	4^{a} ± 1.3	5^{a} ± 2.4
3	10^{a} ± 3.2	8^{a} ± 2.1	5^{a} ± 2.4	2^{a} ± 1.2	7^{a} ± 3.5
4	8^{a} ± 2.1	12^{a} ± 3.4	3^{a} ± 1.6	0^{a} ± 0	0^{a} ± 0
5	10^{a} ± 3.2	3^{a} ± 1.6	2^{a} ± 1.2	2^{a} ± 1.2	1^{a} ± 0.6
6	6^{a} ± 2.1	10^{a} ± 3.2	2^{a} ± 1.2	0^{a} ± 0	0^{a} ± 0
7	4^{a} ± 1.3	6^{ab} ± 2.1	1^{b}±0.6	0^{b} ± 0	0^{b} ± 0

Means followed by the same lowercase letter in each row are not significantly different using Turkey's test at P < 0.05

increased. There were very few of eggs at the lowest concentrations of silica nanoparticles, after 7 days post exposure, and there were no eggs laid at the highest concentrations (0.67 and 1.7 mg kg^{-1}) after 6 and 7 days post exposure, respectively.

DISCUSSION

Results from the bioassay (Table 1) revealed that silica nanoparticles are an effective pest control approach against *C. serratus*. The mortality of *C. serratus* adults increased with concentrations and time exposed to each concentration, implying that application of silica nanoparticles could significantly increases mortality effect of nanoparticles with increasing the time after application or concentrations (Debnath *et al*., 2012; Rouhani *et al*., 2012). Additionally, Debnath *et al*. (2012) and Rouhani *et al*. (2012) reveal that silica nanoparticles has a high potential as pesticide. Silica nanoparticles can be applied to protect plant crops in fields; because they do not have adverse effects on plant growth, but also enhance structural rigidity and strength of plants (Debnath *et al*., 2011). On the basis of our results, silica nanoparticles have high toxicity on *C. serratus* adults. Silica nanoparticles is a physically active material; thus these nanoparticles caused damage to cuticular water barrier of the insect mostly by abrasion (Osman *et al*., 2015), or by absorbance of this nanoparticles into the cubicula lipids of the insect, resulting in damage in the protective wax layer and induces death by desiccation (Rahman *et al.,* 2009).

Silica nanoparticles used in this study seem more effective that those used in most previous studies. Rouhani *et al*. (2012) reported that 2.5 mg kg^{-1} of silica nanoparticles caused 96% mortality of *Callosobruchus maculatus* after 7 days of exposure. Goswami *et al*. (2010) obtained, after 7 days of exposure, 100% mortality with the silica nanoparticles with 2 mg kg^{-1}. However, according to our results, 100% mortality of *C. serratus* was observed on groundnut treated with 0.67 mg kg^{-1} tested silica nanoparticles, after 7 days of exposure (Table 1). The high insecticidal potential of silica nanoparticles could be attributed to the nanoparticles calcined. High temperature calcination increases the porosity of the silica, resulting in better absorption of the insect wax layer, causing rapid death through desiccation and to a lesser degree by abrasion (Ertan *et al*., 2009).

Silica nanoparticles concentration drastically reduced the fecundity of *C. serratus* (Table 2). Suffering of adult insects from desiccation and spiracular blockage by exposure of silica nanoparticle, could be the cause for this reduced fecundity. This could have prevented mating as the treated silica nanoparticles attached all over the body of adult beetles. Insects are supposed to release a greasy layer on their body surface (Voigt *et al*., 2009), which may be involved in physical interactions between the organisms, especially during mating. In mating, males frequently attach to the female's dorsal body by means of their feet, where grease should play an important role for the attachment of feet (Voigt *et al*., 2009). This could have caused incomplete mating in the case of lower concentrations, and prevented mating at higher concentrations of silica nanoparticles treatments. It may not only have been desiccation or blockage of spiracles that did cause a reduction in fecundity, but also the surface enlargement of the integument as a consequence of dehydration (Arumugam *et al*., 2016).

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